Parental support and cytokine activity in childhood asthma: The role of glucocorticoid sensitivity

Gregory E. Miller, PhD, Alexandra Gaudin, BA, Eva Zysk, BA, and Edith Chen, PhD *Vancouver, British Columbia, Canada*

Background: Stress is known to worsen the course of asthma, but the underlying mechanisms are poorly understood. This problem is especially difficult because stress elicits secretion of cortisol, a hormone that dampens airway inflammation and ameliorates asthma symptoms.

Objective: This article proposes that stress affects asthma by inducing resistance to the anti-inflammatory properties of glucocorticoids. To evaluate this hypothesis, we examine whether a particular kind of stress in children's lives, not feeling supported or understood by parents, is associated with *in vitro* measures of lymphocyte resistance to glucocorticoids and indices of eosinophil mobilization and activation.

Methods: Children with asthma (n = 67) and medically healthy children (n = 76) completed standardized questionnaires about support from their parents. PBMCs were collected and incubated with a mitogen cocktail in the presence of physiologic concentrations of hydrocortisone. Production of IL-5, IL-13, and IFN-γ was measured by means of ELISA. Circulating eosinophils were enumerated with a hematology analyzer, and the extent of their activation was indexed by means of ELISA for eosinophil cationic protein.

Results: To the extent that children with asthma perceived low support from their parents, children were more resistant to hydrocortisone's anti-inflammatory effects on IL-5 and IFN-γ production and had higher circulating levels of eosinophil cationic protein. These associations were independent of socioeconomic conditions, cigarette exposure, disease severity, and medication use.

Conclusions: These patterns suggest the hypothesis that strained parent-child relations, and perhaps stress more generally, brings about adverse outcomes in asthma by diminishing cortisol's ability to regulate cytokine activity and subsequent airway inflammation. (J Allergy Clin Immunol 2009;123:824-30.)

Key words: *Social support, cortisol, cytokines, stress, glucocorticoid receptor*

At least since the time of the 12th-century physician-philosopher Maimonides,[1] doctors and patients have believed that stress contributes to asthma.[2,3] However, it has only been in recent decades that evidence has emerged to support this view.[4-6] Stress that arises from turmoil within the family seems to be particularly detrimental.[7] For example, prospective studies have found that wheezing and asthma are more likely to develop in children whose families report parenting difficulties, high stress, or mood problems.[8-11] Also, in studies of children with existing asthma, family difficulties have been linked with increased symptom expression, worse pulmonary function, and the onset of attacks.[12,13]

The mechanisms responsible for these associations are not well understood.[14] Historically, much of the blame for stress-related diseases has been assigned to the hypothalamic-pituitary-adrenocortical axis,[15] which releases cortisol into the circulation after many psychologically demanding experiences.[16,17] Cortisol has a wide variety of physiologic consequences, including mobilization of glucose, regulation of vascular tone and fluid volume, and modulation of immune function.[18] This chain of events seems unlikely to contribute to stress-related asthma, however, because one of cortisol's major actions in the immune system is to suppress inflammation.[19] In fact, to the extent that stress provokes a cortisol surge, it might theoretically be expected to dampen airway inflammation and by doing so ameliorate symptoms of asthma, as it does when administered therapeutically as inhaled or systemic corticosteroid. Of course, evidence suggests just the opposite pattern: that stress amplifies the immune response to allergens and irritants[20,21] and increases the frequency and severity of symptoms.[4,5]

To resolve this paradox and identify underlying mechanisms, we have advanced the hypothesis that chronic stress fosters resistance to glucocorticoids.[22,23] This view suggests that chronic stress triggers persistent secretion of cortisol, which leads to compensatory downregulation of glucocorticoid receptor (GR) expression and functioning. In patients with asthma, such dynamics could enable airway inflammation to flourish and also diminish the efficacy of therapeutics that work through GRs. In an earlier project with asthmatic patients, we found evidence consistent with this scenario: high levels of chronic turmoil in the family, especially when coupled with other stressors, were linked with a 5.5-fold reduction in leukocyte GR mRNA.[24] Although these findings suggest that family turmoil can downregulate GR expression, it remains unclear whether it also has functional implications for the cellular processes that drive asthma.

The current project enrolled children with asthma and healthy control subjects to answer this question. We assessed an important

From the Department of Psychology, University of British Columbia.

Supported by grants from the Michael Smith Foundation; the National Heart, Lung, and Blood Institute; the National Institute of Child Health and Human Development; the Canadian Institutes of Health Research; and the William T. Grant Foundation.

Disclosure of potential conflict of interest: All of the authors have received research support from the National Institutes of Health, the Canadian Institutes of Health Research, Allergen NCE, and the Heart and Stroke Foundation of Canada.

Received for publication September 18, 2008; revised November 12, 2008; accepted for publication December 8, 2008.

Available online February 2, 2009.

Reprint requests: Gregory E. Miller, PhD, 2136 West Mall Ave, Vancouver BC V6T1Z4 Canada. E-mail: gemiller@psych.ubc.ca.

0091-6749/$36.00

doi:10.1016/j.jaci.2008.12.019

Abbreviations used
ECP: Eosinophil cationic protein
GR: Glucocorticoid receptor

component of children's familial relationships: how much support and understanding they felt from their parents. We also collected PBMCs and, after incubating them with mitogens and cortisol, measured production of T_H1 (IFN-γ) and T_H2 (IL-5 and IL- 13) cytokines that regulate allergic immune responses. We predicted that among children with asthma, low family support would be associated with decreased sensitivity to cortisol's suppressive influences on cytokine production. We also predicted that as a downstream consequence of this dynamic, children with low support would show higher eosinophil counts and more eosinophil cationic protein (ECP). ECP is a cytotoxic protein released from the granules of activated eosinophils and is considered a surrogate marker of inflammation in the airways.

METHODS

Sample

The sample consisted of 67 children with asthma and 76 medically healthy children. All were recruited from Vancouver, British Columbia, through advertisements in physician's offices, newspapers and magazines, and community settings. Children were eligible if they were 9 to 18 years of age, fluent in English, and free of upper respiratory tract illness for the past 4 weeks. Children in the asthma group had to have a physician's diagnosis and be free of other chronic medical illnesses. Healthy control subjects were required to have a history without chronic illness. The protocol was approved by the University of British Columbia's Research Ethics Board. All children provided written assent before participating and had a parent or guardian provide written consent.

Parental support

Children completed the family subscale of the Social Support Scale for Children, a self-report instrument that has been extensively validated.[12,25,26] They were asked to rate the degree to which 6 statements described their parental relationships, such as "Some kids have parents who care about their feelings" and "Some kids have parents who like them as they are." Responses were given on a 4-point scale. The scale's items were internally consistent (Cronbach $\alpha = .73$), but the distribution was skewed, with most children having scores on the higher end. We therefore stratified the sample into 4 groups ranging from low (1) to high (4). This variable was normally distributed.

Glucocorticoid sensitivity

When participants arrived, a topical anesthetic (Eutectic Mixture of Local Anesthetics, APP Pharmaceuticals, Schaumburg, Ill) was applied to minimize discomfort from venipuncture. One hour later, 10 mL of blood was drawn from the antecubital vein into Cell Preparation Tubes (Becton Dickinson, Franklin Lakes, NJ). PBMCs were then isolated and resuspended in RPMI-1640 medium with HEPES supplemented with 10% FCS (Sigma-Aldrich, ST Louis, Mo) at a concentration of 3×10^6 cells/mL. PBMCs were then incubated at 37°C and 5% CO_2 with 25 ng/mL phorbol 12-myristate 13-acetate and 1 μg/mL ionomycin calcium salt and either 0 or 10 ng/mL hydrocortisone (all from Sigma-Aldrich). The final in-well concentration of hydrocortisone was 28 nmol/L. (In pilot studies we experimented with hydrocortisone doses ranging from 0.3 to 300 nmol/L. Because cytokine responses were strongly correlated across doses, with r values ranging from 0.79 for IL-13 to 0.91 for IFN-γ, we opted to use a single 28 nmol/L administration in the final study. This dose is roughly similar to the values in human blood during mild psychologic stress.) After 48 hours of culture, cell suspensions were centrifuged, and supernatants were collected and frozen at −80°C. Concentrations of IL-5, IL-13, and IFN-γ were quantified by means of ELISA (R&D Systems, Minneapolis, Minn). Coefficients of variation averaged less than 8%. For each cytokine, we later computed a variable reflecting "percentage resistance to glucocorticoid inhibition" by dividing values in hydrocortisone-treated wells by those in saline-treated wells.

To evaluate the reproducibility of these procedures, we drew blood from 3 technicians on 3 consecutive days and then divided each sample into 3 aliquots. The assay was then run on 9 samples. Analyses revealed that within-person coefficients of variation averaged 9.7%, 11.4%, and 16.6% for IL-5, IL-13, and IFN-γ, respectively. These data suggest that our procedures are reproducible and that patient responses are fairly stable over short timeframes.

Eosinophil counts and activation

Eosinophil counts were measured as part of a 5-part blood count and differential with a Bayer Advia 70 hematology analyzer (Diamond Diagnostics, Holiston, Mass). ECP levels were measured by using the ImmunoCAP system (PhadiaAB, Uppsala, Sweden) with reagents from Somagen (Edmonton, Alberta, Canada). Ten milliliters of blood was drawn into Serum Separator Tubes (BD, Franklin Lakes, NJ) and incubated at room temperature for 90 minutes. During this interval, activated eosinophils released ECP. The tubes were then centrifuged. After serum had been aspirated, it was stored at −80°C.

Potential confounders

To determine whether behavioral and biomedical characteristics might be acting as confounders, we collected information regarding the children's age, sex, family income, cigarette exposure, pulmonary function, and medication use. Each of these factors has been linked to family support, cytokine production in past work, or both.[12,27,28] Demographic information was collected during parental interviews. Personal use of cigarettes and secondhand exposure were ascertained by means of a questionnaire. Because only 1 child was a regular smoker, we used hours per week of exposure to secondhand smoke as a covariate in analyses. Pulmonary function was indexed by FEV_1 during a spirometric examination. Spirometry was conducted at least 4 hours after the last use of a short-acting bronchodilator and performed according to guidelines used in large multisite clinical trials in patients with asthma.[29] FEV_1 values were calculated as a percentage of predicted value based on child age, sex, ethnicity, and height. Medication use was assessed by having parents bring all of their child's medications to the research center. The name and dosage of each medication was recorded directly from the label, and the frequency of use in the past 2 weeks was ascertained. In analyses we included variables reflecting days of treatment with inhaled corticosteroids and β-agonists as covariates.

Statistical analyses

Hypotheses were evaluated in a series of regression equations. Each biologic outcome was predicted from 3 blocks of variables entered consecutively. They consisted of (1) potential confounders, (2) indicators of parental support and asthma status, and (3) a product term representing the interaction of the latter factors. When a statistically significant interaction emerged, it was decomposed according to standard procedures.[30] Simple slopes within each group were computed. Percentage resistance was then estimated at low, medium, and high levels of support (defined as 1 SD below, the sample mean, and 1 SD above, respectively) and then plotted separately for the asthmatic and healthy children. For all statistical analyses, α was set to .05, and 2-tailed tests of significance were performed.

RESULTS

Preliminary analyses

Table I displays the sample characteristics. The children with asthma and medically healthy children were similar in terms of age, ethnic background, family income, and cigarette exposure ($P > .35$). However, there was a greater proportion of male subjects in the asthmatic sample ($\chi^2 = 5.74$, $P = .03$).

TABLE I. Characteristics of the sample

	Asthmatic subjects (n = 67)	Healthy subjects (n = 76)
Age (y)	13.3 ± 0.4	13.4 ± 0.3
Sex (male)*	49 (73.2%)	40 (52.6%)
Descent (European)	39 (58.2%)	43 (57.3%)
Descent (Asian)	22 (32.8%)	20 (26.3%)
Annual family income (thousands)	68.7 ± 2.7	63.8 ± 2.6
Low income (<$35,000 annually)	9 (13.4%)	13 (17.1%)
FEV1 (% predicted)*	97.1 ± 1.3	103.2 ± 1.9
Cigarette exposure (h/wk)	1.2 ± 0.2	1.0 ± 0.2
Inhaled corticosteroids (last 2 wk)	25 (37.3%)	—
Bronchodilator (last 2 wk)	28 (41.8%)	—
Asthma severity (moderate or severe)	33 (49.3%)	—
Family support (1-4)	2.5 ± 0.1	2.5 ± 0.1
IL-5 production, saline (pg/mL)	73.3 ± 9.1	72.9 ± 7.6
IL-13 production, saline (pg/mL)	259.6 ± 27.3	311.9 ± 21.2
IFN-γ production, saline (pg/mL)†	6,392.9 ± 962.9	16,523.3 ± 1,519.9
IL-5 production, hydrocortisone (pg/mL)	46.8 ± 5.5	49.7 ± 5.3
IL-13 production, hydrocortisone (pg/mL)	219.7 ± 21.8	267.1 ± 19.6
IFN-γ production, hydrocortisone (pg/mL)†	6,385.8 + 1,004.7	13,215.1 + 1,268.6
Circulating eosinophils ($\times 10^9$ cells/L)†	0.35 ± 0.04	0.17 ± 0.02
ECP (μg/L)†	16.2 ± 1.5	11.5 ± 1.0

Values are presented as means ± SE or numbers (percentages). Symbols indicate that the asthmatic and healthy groups differ.
*$P < .05$.
†$P < .01$.

Children with asthma had lower FEV_1 percentiles than healthy control subjects ($t = 2.82$, $P = .007$). About 40% of them had used inhaled corticosteroids and β-agonists in the past 2 weeks. Using guidelines from the National Asthma Education and Prevention Program's Expert Panel Report 2, based on the higher of symptom frequency and medication use,[31] 11 (7.6%) of the children with asthma had mild intermittent disease, and the others had persistent disease that was classified as mild (23 [15.9%]), moderate (22 [15.2%]), or severe (11 [7.6%]). Fifty-seven (85.1%) of the children with asthma (85.1%), and 32 (42.1%) of the healthy children were atopic, as determined by means of screening of serum IgE antibodies to common allergens (ImmunoCAP 100€; Phadia AB, Uppsala, Sweden).

Table I shows that the groups perceived similar levels of support from their parents ($t = 0.15$, $P = .89$), and their PBMCs produced similar amounts of IL-5 and IL-13 when stimulated *in vitro* ($t < 1.40$, $P > .16$). Children with asthma produced less IFN-γ after stimulation than healthy control subjects ($t = 5.49$, $P < .001$). The same patterns emerged in wells treated with hydrocortisone. The groups had similar concentrations of IL-5 and IL-13 ($t < 1.53$, $P > .13$), but children with asthma continued to produce less IFN-γ ($t = 4.44$, $P < .001$). Children with asthma had higher eosinophil counts and more circulating ECP than healthy children ($P < .01$).

Further analyses indicated that coincubation with hydrocortisone significantly reduced IL-5, IL-13, and IFN-γ production relative to saline ($t > 4.99$, $P < .001$). The suppressive effects of hydrocortisone were similar in magnitude for the asthma and control groups for IL-5 and IL-13 ($t < 1.0$, $P > .38$). However, hydrocortisone's effects for IFN-γ production were significantly more pronounced in healthy children ($F = 22.65$, $P < .001$); that is, hydrocortisone reduced IFN-γ production by 20.0% relative to saline treatment in healthy children ($t = 6.88$, $P < .001$) but by less than 0.1% in those with asthma ($t = 0.13$, $P > .90$).

Parental support and glucocorticoid sensitivity

Table II presents regression analyses for each cytokine. For IL-5, subjects from lower-income families were more resistant to the inhibitory properties of hydrocortisone, but none of other covariates emerged as significant predictors. There were also no main effects of asthma status or parental support. However, a significant interaction between these variables emerged ($P = .03$; Fig 1, upper panel). Asthmatic patients with less family support were more resistant to hydrocortisone inhibition of IL-5 production, but there was no association between these factors among the healthy children.

For IFN-γ, none of the covariates was a predictor of glucocorticoid sensitivity, and asthmatic patients displayed greater resistance to inhibition than healthy children. There was also a significant interaction between asthma status and parental support ($P = .03$) that was similar to the IL-5 effect (Fig 1, lower panel); that is, among children with asthma, low parental support was associated with more resistance to hydrocortisone inhibition, whereas these factors were unrelated in healthy children.

For IL-13, the only variable to predict glucocorticoid sensitivity was FEV_1 percentage: children with lower pulmonary function were more resistant to hydrocortisone. The main effects of asthma status and parental support and their interaction were nonsignificant.

To further evaluate the role of inhaled corticosteroids in these associations, we redid analyses in the subgroup of asthmatic patients (n = 42) and healthy control subjects (n = 76) who had not used them in the last 2 weeks. The results were identical to those presented above. For both IL-5 and IFN-γ, there were interactions between asthma and support (for IL-5 interaction: B = −0.14, SE = 0.07, $P = .05$; for IFN-γ interaction: B = −0.16, SE = 0.08, $P = .05$). In both cases lower support was associated with greater resistance to hydrocortisone inhibition among children with asthma. In the healthy control subjects family support was unrelated to IL-5 hydrocortisone sensitivity but showed a weak positive relation with IFN-γ, which was not significant (P for simple slope = .61). These findings are displayed in Fig 2. There was no interaction for IL-13.

Seven asthmatic patients had taken a course of oral steroids within the last 6 months. The pattern of results was similar when they were removed from the analyses (for IL-5 interaction: B = −0.13, SE = 0.05, $P = .02$; for IFN-γ interaction: B = −0.14, SE = 0.07, $P = .04$), suggesting that use of these medications was not contributing to the findings.

Eosinophil mobilization and activation

To evaluate potential downstream consequences of the glucocorticoid resistance, we examined relations between parental support and eosinophil mobilization and activation. As Table III shows, eosinophil counts were higher among children with asthma and those using regular medications but unrelated to familial support. ECP levels were higher in asthmatic patients and showed a significant medical status-by-family support interaction similar to IL-5 and IFN-γ (Fig 3). Thus in children with asthma, low parental

TABLE II. Regression analyses relating family support to cytokine production

	IL-5				IFN-γ				IL-13			
Predictor	**B**	**IL-5 SE B**	**β**	***P* value**	**B**	**IFN-γ SE B**	**β**	***P* value**	**B**	**IL-13 SE B**	**β**	***P* value**
Step 1												
Age	0.01	0.01	0.05	.55	−0.01	0.01	−0.02	.83	0.01	0.02	0.06	.48
Sex	−0.04	0.06	−0.05	.55	−0.10	0.07	−0.12	.18	−0.04	0.08	−0.04	.63
Income	−0.03	0.02	−0.17	.05	0.01	0.02	0.02	.80	0.03	0.02	0.13	.14
Smoke exposure	−0.01	0.02	−0.01	.94	0.02	0.02	0.01	.91	−0.03	0.03	−0.12	.19
FEV_1	−0.01	0.01	−0.07	.41	0.01	0.01	0.02	.86	−0.01	0.01	−0.20	.03
Steroids	−0.23	0.16	−0.21	.15	0.28	0.20	0.21	.17	0.05	0.23	0.03	.83
β-Agonists	0.16	0.16	0.15	.31	−0.19	0.20	−0.14	.34	0.32	0.22	0.22	.15
Step 2												
Asthma	−0.06	0.07	−0.09	.33	0.16	0.08	0.20	.05	−0.10	0.09	−0.11	.28
Support	−0.01	0.04	−0.02	.84	0.03	0.05	0.07	.54	0.04	0.05	0.09	.40
Step 3												
Asthma × support	−0.13	0.06	−0.27	.03	−0.15	0.07	−0.25	.03	−0.06	0.08	−0.09	.47

In the IL-5 equation the cumulative percentage variance explained ($100 \times R^2$) at steps 1, 2, and 3 was 4.3, 9.4, and 13.7. The parallel figures for the IFN-γ and IL-13 equations were 7.8, 10.6, and 13.9 and 9.6, 10.7, and 11.2, respectively.

B, Unstandardized regression coefficient; *SE B*, standard error of regression coefficient; β, standardized regression coefficient.

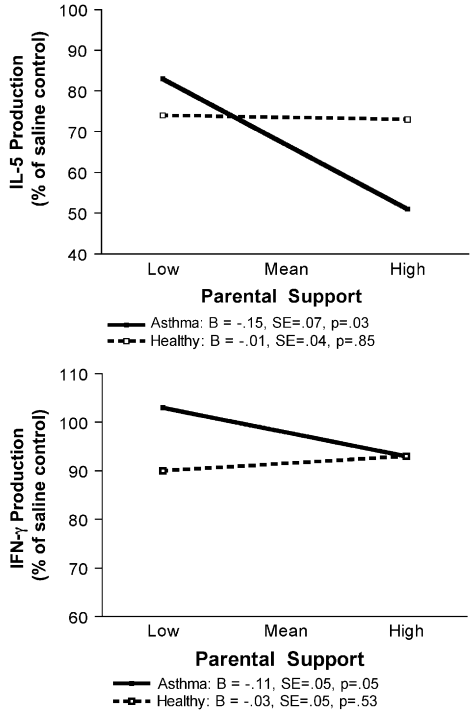

FIG 1. Cytokine production as a function of family support. To the extent that they perceived low support from their parents, children with asthma were more resistant to hydrocortisone's anti-inflammatory effects on production of IL-5 and IFN-γ. These factors were unrelated in healthy control subjects.

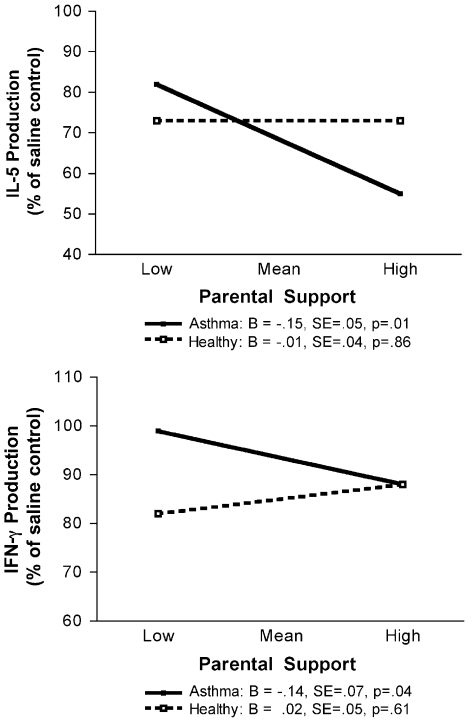

FIG 2. Cytokine production as a function of family support in children who do not use inhaled corticosteroids regularly. To the extent that they perceived low support from their parents, inhaled corticosteroid–free children with asthma were more resistant to hydrocortisone's anti-inflammatory effects on production of IL-5 and IFN-γ. These factors were unrelated in healthy control subjects.

support was associated with higher ECP concentrations, whereas in healthy children these variables were unrelated.

DISCUSSION

Researchers focusing on stress and asthma have long struggled to resolve a paradox: How can stress worsen the disease if it also triggers secretion of cortisol, a hormone that potently suppresses inflammation and is used clinically to ameliorate symptoms? The data from this study suggest that glucocorticoid insensitivity might be one of the mechanisms. We found that to the extent that children with asthma perceived low support from their parents, they were more resistant to hydrocortisone's anti-inflammatory effects on IL-5 and IFN-γ production. They also had greater eosinophil activation, as manifested by ECP concentration. This might have been a downstream consequence of the IL-5 dysregulation because this cytokine is pivotal in recruiting eosinophils to the airways and inducing them to degranulate. Together, these patterns suggest the hypothesis that strained

TABLE III. Regression analyses relating family support to eosinophil mobilization and activation

	Eos				ECP			
Predictor	**B**	**Eos SE B**	**β**	***P* value**	**B**	**ECP SE B**	**β**	***P* value**
Step 1								
Age	−0.01	0.01	−0.14	.06	−0.59	0.49	−0.10	.24
Sex	−0.01	0.04	−0.02	.81	−0.36	0.60	−0.01	.89
Income	−0.02	0.02	−0.06	.42	1.61	1.63	0.08	.33
Smoke exposure	−0.01	0.01	−0.01	.88	0.46	0.75	0.62	.54
FEV_1	−0.01	0.01	−0.03	.70	−0.10	0.10	−0.04	.64
Steroids	0.27	0.10	0.39	.01	−12.63	7.46	−0.27	.10
β-Agonists	−0.22	0.10	−0.31	.04	−9.86	7.65	−0.21	.19
Step 2								
Asthma	0.18	0.04	0.38	.01	5.72	2.76	0.19	.04
Support	−0.01	0.02	−0.06	.52	1.61	1.61	0.11	.32
Step 3								
Asthma × support	−0.04	0.03	−0.11	.27	−5.79	2.37	−0.27	.02

In the eosinophil equation the cumulative percentage variance explained ($100 \times R^2$) at steps 1, 2, and 3 was 17.3, 28.9, and 29.5. The parallel figures for the ECP equation were 2.2, 5.3, and 8.8.

Eos, Eosinophil counts in peripheral blood; *B*, unstandardized regression coefficient; *SE B*, standard error of regression coefficient; β, standardized regression coefficient.

parent-child relations, and perhaps stress more generally, worsens asthma by diminishing cortisol's ability to regulate IL-5 activity and subsequent eosinophilic inflammation.

IFN-γ's role in this chain of events is unclear. We found that among children with low parental support, hydrocortisone was less able to suppress IFN-γ production. If this dynamic increases levels of IFN-γ in the airways, it might be expected to positively influence disease because this molecule is a potent inhibitor of T_H2 cytokines, such as IL-5, that promote eosinophilic inflammation. However, recent evidence indicates that IFN-γ is often present at fairly high levels in the airways of asthmatic patients, and some have argued that it even contributes to asthma pathogenesis by coordinating epithelial responses to viral infection.[32,33] Thus another possibility is that by diminishing cortisol's ability to regulate the production of IFN-γ, strained parent-child relations could amplify the antiviral immune response and thereby contribute to inflammation and obstruction of the airways. Consistent with this reasoning, studies indicate that stress accentuates the proinflammatory cytokine response to respiratory pathogens.[34,35]

These data highlight the importance of family relationships in asthma. Parental support has long been viewed as an essential component of childhood asthma management, but the prevailing assumption has been that its benefits are primarily attributable to improved compliance with medication and avoidance of asthma triggers. However, these data add to growing evidence that familial relations also have a more direct influence on the biologic processes that drive asthma pathogenesis.[7,36] For example, other studies have shown that in children with asthma, impaired family functioning is related to higher levels of IgE, greater lymphocyte proliferative responses to allergic triggers, and heightened *in vitro* production of T_H2 cytokines,[12,28,37] as well as to clinical outcomes, such as the onset of wheezing and asthma and the expression of symptoms in daily life.[8-12] A lack of parental support might be one of the critical mechanisms underlying these associations because of its importance in facilitating effective coping, emotional adjustment, and treatment adherence.[38]

These data also converge with research indicating that stress alters cortisol-signaling dynamics in children with asthma. For example, asthmatic children display blunted cortisol responses to acute psychologic stress[39] and, when they are exposed to familial turmoil and other life events, express markedly less GR mRNA in leukocytes.[24] In animal models stress-induced blunting of cortisol output has been linked with greater bronchial reactivity and more cellular infiltration of the airways.[40] Collectively, these findings suggest that stress has the capacity to broadly impair cortisol-related signaling in asthma, not only by reducing the expression of relevant ligands and receptors but also by impairing lymphocytes' ability to register and respond to signals from cortisol.

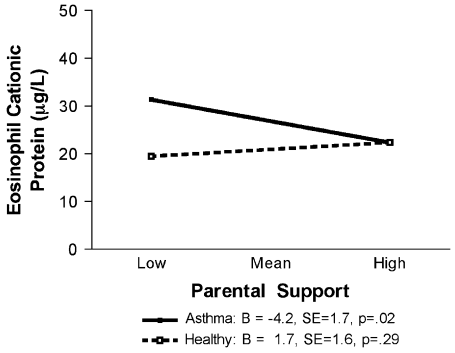

FIG 3. ECP as a function of family support. To the extent that they perceived low support from their parents, children with asthma had higher circulating concentrations of ECP. These factors were unrelated in healthy control subjects.

Thus our findings might have implications for understanding why some asthmatic patients have suboptimal responses to glucocorticoid therapy.[41] Most of these patients have an acquired form of glucocorticoid resistance thought to arise from heavy exposure to allergens, infection with microbial superantigens, or genetic predisposition.[42] Our data suggest that unsupportive parental relationships could play a role in the response to glucocorticoid therapy as well. The mechanisms underlying such an effect are not clear but could include poor medication compliance or stress-related downregulation of GR.[24] It is also interesting to note that interpersonal difficulties are associated with increased activation of nuclear factor κB–dependent genes in monocytes,[43,44] a pattern that is also seen in the bronchoalveolar lavage fluid of glucocorticoid-resistant asthmatic patients.[45] Thus upregulation of the classical macrophage activation pathway is another potential mechanism contributing to the findings herein.

Our data also suggest that familial dynamics relate to cytokine activity differently in healthy children and asthmatic patients. Although low parental support was associated with hydrocortisone resistance in the asthmatic sample, these factors were unrelated in control subjects. This pattern of findings was unexpected. Other studies have found linkages between difficult family situations and resistance to glucocorticoids in healthy subjects.[22,43] However, in these projects the stressors were severe and chronic; for example, the subjects had spouses or children with cancer. Thus it might be that healthy subjects have a relatively high threshold for stress-related changes in glucocorticoid sensitivity compared with asthmatic patients.

This study has several limitations. First, it is based on a small group of children who generally had well-controlled asthma and good family functioning, and as such, its generalizability might be limited.

Second, it focused on support from parents, which is just one of several important dimensions of family functioning in asthma, along with family conflict, parenting difficulties, and mental health status.[38] Future work will need to explore the relative contributions of these processes and pay more careful attention to issues such as chronicity of exposure and variation across demographic groups.[46]

Third, the assays used to index glucocorticoid sensitivity were carried out *in vitro* and made use of a nonspecific mitogen cocktail. Future research will need to substantiate these findings in conditions that more closely approximate the airway milieu.

Finally, because the project's design was cross-sectional, it is impossible to make causal inferences from the data. We have ruled out some of the most plausible alternative explanations for the observed patterns, including differences in socioeconomic conditions, cigarette exposure, disease severity, and medication use. Nonetheless, other (unmeasured) confounders could have contributed to the associations.

Despite these limitations, the study begins to shed light on a long-standing paradox about the role of cortisol in linking stress to asthma and in doing so provides some clues about how the social world might affect airway disease. Of course, there are likely to be additional mechanisms involved, such as stress-related shifts in the balance of T_H1/T_H2 cytokines,[47] and changes in medication adherence, health behavior, and symptom perception, and future research will need to evaluate the relative importance of these pathways. Such work will provide a deeper understanding of potential mechanisms and might inform the development of interventions to improve patient outcomes.

Clinical implications: These findings suggest that strained family relations diminish cortisol's ability to regulate cytokines that orchestrate inflammation. By assessing the familial environment and making necessary referrals, clinicians might be able to improve patient outcomes.

REFERENCES

1. Muntner S. Maimonides' treatise on asthma. Dis Chest 1968;54:128-32.
2. French TM, Alexander F. Psychogenic factors in bronchial asthma. In: Psychosomatic medicine monograph IV, part I. Washington (DC): National Research Council; 1941.
3. Wright RJ, Rodriquez MS, Cohen S. Review of psychosocial stress and asthma: an integrated biopsychosocial approach. Thorax 1998;53:1066-74.
4. Wright RJ, Cohen RT, Cohen S. The impact of stress on the development and expression of atopy. Curr Opin Allergy Clin Immunol 2005;5:23-9.
5. Bloomberg GR, Chen E. The relationship of psychologic stress with childhood asthma. Immunol Allergy Clin North Am 2005;25:83-105.
6. Marshall GD, Roy SR. Stress and allergic diseases. In: Ader R, editor. Psychoneuroimmunology. 4th ed. Boston: Elsevier, 2007. p. 799-824.
7. Wright RJ. Prenatal maternal stress and early caregiving experiences: implications for childhood asthma risk. Paediatr Perinat Epidemiol 2007;21(suppl 3):8-14.
8. Mrazek DA, Klinnert M, Mrazek PJ, Brower A, McCormick D, Rubin B, et al. Prediction of early-onset asthma in genetically at-risk children. Pediatr Pulmonol 1999;27:85-94.
9. Klinnert MD, Nelson HS, Price MR, Adinoff AD, Leung DY, Mrazek DA. Onset and persistence of childhood asthma: predictors from infancy. Pediatrics 2001; 108:E69.
10. Kozyrskyj AL, Mai XM, McGrath P, Hayglass KT, Becker AB, Macneil B. Continued exposure to maternal distress in early life is associated with an increased risk of childhood asthma. Am J Respir Crit Care Med 2008;177:142-7.
11. Wright RJ, Cohen S, Carey V, Weiss ST, Gold DR. Parental stress as a predictor of wheezing in infancy: a prospective birth-cohort study. Am J Respir Crit Care Med 2002;165:358-65.
12. Chen E, Chim LS, Strunk RC, Miller GE. The role of the social environment in children and adolescents with asthma. Am J Respir Crit Care Med 2007;176:644-9.
13. Sandberg S, Paton JY, Ahola S, McCann DC, McGuinness D, Hillary CR, et al. The role of acute and chronic stress in asthma attacks in children. Lancet 2000;356:982-7.
14. Marshall GD. Neuroendocrine mechanisms of immune dysregulation: applications to allergy and asthma. Ann Allergy Asthma Immunol 2004;93(suppl):S11-7.
15. McEwen BS. Protective and damaging effects of stress mediators. N Engl J Med 1998;338:171-9.
16. Dickerson SS, Kemeny ME. Acute stressors and cortisol responses: a theoretical integration and synthesis of laboratory research. Psychol Bull 2004;130:355-91.
17. Miller GE, Chen E, Zhou ES. If it goes up, must it come down? Chronic stress and the hypothalamic-pituitary-adrenocortical axis in humans. Psychol Bull 2007;133:25-45.
18. Sapolsky RM, Romero LM, Munck AU. How do glucocorticoids influence stress responses? Integrating permissive, suppressive, stimulatory, and preparative actions. Endocr Rev 2000;21:55-89.
19. Webster JI, Tonelli L, Sternberg EM. Neuroendocrine regulation of immunity. Annu Rev Immunol 2002;20:125-63.
20. Liu LY, Coe CL, Swenson CA, Kelly EA, Kita H, Busse WW. School examinations enhance airway inflammation to antigen challenge. Am J Respir Crit Care Med 2002;165:1062-7.
21. Rosenkranz MA, Busse WW, Johnstone T, Swenson CA, Crisafi GM, Jackson MM, et al. Neural circuitry underlying the interaction between emotion and asthma symptom exacerbation. Proc Natl Acad Sci U S A 2005;102:13319-24.
22. Miller GE, Cohen S, Ritchey AK. Chronic psychological stress and the regulation of pro-inflammatory cytokines: a glucocorticoid resistance model. Health Psychol 2002;21:531-41.
23. Chen E, Miller GE. Stress and inflammation in exacerbations of asthma. Brain Behav Immun 2007;21:993-9.
24. Miller GE, Chen E. Life stress and diminished expression of genes encoding glucocorticoid receptor and beta2-adrenergic receptor in children with asthma. Proc Natl Acad Sci U S A 2006;103:5496-501.
25. Harter S. Manual for the social support scale for children. Denver (CO): University of Colorado; 1985.
26. von Weiss RT, Rapoff MA, Varni JW, Lindsley CB, Olson NY, Madson KL, et al. Daily hassles and social support as predictors of adjustment in children with pediatric rheumatic disease. J Pediatr Psychol 2002;27:155-65.
27. Chen E, Hanson MD, Paterson LQ, Griffin MJ, Walker HA, Miller GE. Socioeconomic status and inflammatory processes in childhood asthma: the role of psychological stress. J Allergy Clin Immunol 2006;117:1014-20.
28. Wright RJ, Finn P, Contreras JP, Cohen S, Wright RO, Staudenmayer J, et al. Chronic caregiver stress and IgE expression, allergen-induced proliferation, and cytokine profiles in a birth cohort predisposed to atopy. J Allergy Clin Immunol 2004;113:1051-7.
29. The Childhood Asthma Management Program (CAMP): design, rationale, and methods. Control Clin Trial 1999;20:91-120.
30. Aiken LS, West SG. Multiple regression: testing and interpreting interactions. London: Sage Publications; 1991.
31. Bacharier LB, Strunk RC, Mauger D, White D, Lemanske RFJ, Sorkness CA. Classifying asthma severity in children: mismatch between symptoms, medication use, and lung function. Am J Respir Crit Care Med 2004;170:426-32.
32. Holtzman MJ. Where are the gaps in asthma research? A counter-perspective. J Allergy Clin Immunol 2003;111:244-7.
33. Holtzman MJ, Agapov E, Kim E, Kim JI, Morton JD. Developing the epithelial, viral, and allergic paradigm for asthma: Giles F. Filley lecture. Chest 2003; 123(suppl):377S-84S.
34. Konstantinos AP, Sheridan JF. Stress and influenza viral infection: modulation of proinflammatory cytokine responses in the lung. Respir Physiol 2001;128:71-7.

35. Cohen S, Doyle WJ, Skoner DP. Psychological stress, cytokine production, and severity of upper respiratory illness. Psychosom Med 1999;61:175-80.
36. Sandel M, Wright RJ. When home is where the stress is: expanding the dimensions of housing that influence asthma morbidity. Arch Dis Child 2006;91:942-8.
37. Wolf JM, Miller GE, Chen E. Parent psychological states predict changes in inflammatory markers in children with asthma and healthy children. Brain Behav Immun 2008;22:433-41.
38. Kaugars AS, Klinnert MD, Bender BG. Family influences on pediatric asthma. J Pediatr Psychol 2004;29:475-91.
39. Buske-Kirschbaum A, von Auer K, Krieger S, Weis S, Rauh W, Hellhammer D. Blunted cortisol responses to psychosocial stress in asthmatic children: a general feature of atopic disease? Psychosom Med 2003;65:806-10.
40. Chida Y, Sudo N, Sonoda J, Hiramoto T, Kubo C. Early-life psychological stress exacerbates adult mouse asthma via the hypothalamus-pituitary-adrenal axis. Am J Respir Crit Care Med 2007;175:316-22.
41. Barnes PJ. Corticosteroid resistance in airway disease. Proc Am Thorac Soc 2004; 1:264-8.
42. Leung DY, Bloom JW. Update on glucocorticoid action and resistance. J Allergy Clin Immunol 2003;111:3-22.
43. Miller GE, Chen E, Sze J, Marin T, Arevalo JMG, Doll R, et al. A genomic fingerprint of chronic stress in humans: blunted glucocorticoid and increased NF-κB signaling. Biol Psychiatry 2008;64:266-72.
44. Cole SW, Hawkley LC, Arevalo JM, Sung CS, Rose RM, Cacioppo JT. Social regulation of gene expression: inflammation and the human transcriptional response to loneliness. Genome Biol 2007;8:r89.
45. Goleva E, Hauk PJ, Hall CF, Liu AH, Riches DW, Martin RJ, et al. Corticosteroid-resistant asthma is associated with classical antimicrobial activation of airway macrophages. J Allergy Clin Immunol 2008;122:550-9.
46. Klinnert MD, Price MR, Liu AH, Robinson JL. Unraveling the ecology of risks for early childhood asthma among ethnically diverse families in the southwest. Am J Public Health 2002;92:792-8.
47. Marshall GD, Agarwall SK, Lloyd C, Cohen L, Henninger EM, Morris GM. Cytokine dysregulation associated with exam stress in healthy medical students. Brain Behav Immun 1998;12:297-307.