3 Psychological Stress and Its Relationship to Cytokines and Inflammatory Diseases

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STRESS, TH1/TH2 CYTOKINES, AND INFLAMMATORY DISEASE

The role of psychological stressors in health and disease has been a dominant topic of research in health psychology and psychoneuroimmunology. Much of the early research in this field focused on the immunosuppressive consequences of stress on the immune system.[1,2] However, the past decade has seen the evolution of theories regarding stress and the immune system. The theories account for the different conceptualizations of stress (acute versus chronic) and diverse immune parameters (complements and cytokines). Consequently, they present a more complex view of the relationship between stress and immunity that moves away from seeing stress as solely immunosuppressive and sees it as immunomodulatory as well.[3–6]

The goal of this chapter is to discuss the relationship between psychological stress and the group of cytokines known as Th1 and Th2. We will present some basic biological information that will discuss the structures and functions of Th1 and Th2 cytokines and their interactions with hormones such as glucocorticoids. We will review studies that assessed specific types of stressors and the differential effects of these stressors on Th1 and Th2 cytokines and discuss the implications of these relationships for inflammatory diseases. In particular, we will describe the role of these cytokines in asthma and rheumatoid arthritis and demonstrate how shifts or dysregulations of these cytokines during stress affect these diseases. Finally, we will discuss some future research directions with regard to stress, Th1/Th2 cytokines, and inflammatory diseases.

TH1/TH2 CYTOKINES: BRIEF IMMUNOLOGICAL BACKGROUND

Cytokines are protein molecules secreted by white blood cells to regulate the immune response. They have a wide range of biological functions that include attracting cells to sites of injury and infection, activating and suppressing various cellular functions, and inducing proliferation and differentiation. Th1 and Th2 cytokines gain their names from the fact that they are secreted by T helper (Th) cells to serve different functions during an immune response. Th1 cytokines generally regulate cell-mediated immunity (responses to intracellular pathogens that involve activation of cells such as cyotoxic T cells and natural killer cells), and include molecules such as interleukin-2 (IL-2), tumor necrosis factor alpha (TNF-α), and interferon-gamma (IFN-γ). In contrast, Th2 cytokines play an important role in regulating humoral immunity (responses to extracellular pathogens that involve activation of B cells and antibody production), and include molecules such as IL-4, IL-5, IL-9, IL-10, and IL-13.[7–10]

Although researchers sometimes refer to white blood cells as Th1 or Th2, it is important to remember that this distinction is functional and not morphological, that is, cells are grouped in this way because of the cytokines they generally (but not always) secrete, not because they have observable structural differences. In fact, researchers have had little success in documenting morphological differences between cells that have Th1 versus Th2 cytokine secretion profiles.

The role of the above cytokines also extends beyond the scope of Th1- and Th2-related immune functions. However, based on our interest in implications for

inflammatory diseases noted in this chapter, we will restrict our discussion to the roles of Th1 and Th2 cytokines.

Recent research has shown that both psychological and biological (glucocorticoid) components of stressful experiences can influence the expression of Th1 and Th2 cytokines.[3,9,11] In turn, Th1 and Th2 cytokines play a role in the pathophysiology of inflammatory diseases such as asthma and rheumatoid arthritis. Individuals with asthma have marked predominances of Th2 cytokine profiles. Patients with rheumatoid arthritis are shifted toward a Th1 cytokine profile.[3,4,12–15] Focusing on Th1 and Th2 cytokines will allow us to discuss the implications of psychological stress on the exacerbation and progression of these diseases.

Differentiation and Cross-Regulation

Th1 and Th2 cells are derived from the same precursors: Th0 cells. These naïve Th0 cells are undifferentiated CD4+ helper T cells that become polarized and develop into Th1 or Th2 cells via cytokines and stress hormones that are present at the time of differentiation (in addition to other immune components such as antigen-presenting cells, that will not be discussed in this chapter).[9] Th0 cells require the presence of IL-12, a cytokine produced by activated monocytes or other antigen-presenting cells, to polarize to Th1 cells.[3,9,11] In contrast, IL-4 must be present for Th2 polarization.[9]

Interestingly, Th1 and Th2 cells do not exist independently; they cross-regulate or counterbalance each other via their respective cytokine production mechanisms.[9] For example, IFN-γ suppresses the secretion of IL-4, the cytokine responsible for the differentiation of Th0 cells to Th2.[9] In addition, IL-4 and IL-10 inhibit the secretion of IFN-γ and IL-12, the cytokine responsible for differentiation to Th1.[9] It is important to note that not all Th1 and Th2 cytokines regulate Th0 cell development. For example, IL-13 (a Th2 cytokine) does not drive the differentiation of Th0 cytokines to Th2, as its IL-4 Th2 companion is known to do.[10]

Measurement of Th1 and Th2 Cytokines in Stress Studies

Before reviewing stress and Th1/Th2 cytokine studies, it is important to comment on the measurement of these cytokines in human psychological stress studies. The Th1 and Th2 cytokines discussed in the following sections are generally difficult to detect in basal concentrations because they degrade quickly in tissue and do not spill over into peripheral blood (where immune processes are typically assessed in human subjects) in large quantities.[16] Thus, an alternative methodological approach to investigating the role of stress in cytokines is the stimulation of human lymphocytes *in vitro* with a mitogen (i.e., lipopolysaccharide [LPS] or phytohemagglutinin [PHA]).

Over the course of a 24- to 48-hour incubation period, lymphocytes will then secrete cytokines in response to the mitogen, and researchers can measure levels of mitogen-stimulated cytokines in culture supernatant. When interpreting research in this area, it is critical to remember that important conceptual and methodological differences exist in these approaches to cytokine measurement. With measures of basal cytokine in circulation, researchers ask whether stress influences the expression

of cytokines in peripheral blood at a particular moment in time. With stimulated-cytokine measures, the question is whether stress influences the ability of white blood cells to produce cytokines when they are challenged *in vitro*.

Apart from asking different types of questions about stress and immunity, these procedures also reveal differing strengths and weaknesses related to internal and external validity. A person's basal concentration of cytokine can be influenced by many factors — an ongoing infection, exposure to allergens, a recent injury or surgery — but in most cases it is impossible to definitively identify the stimulus. Hence, basal measures are subject to multiple influences that a researcher cannot control, and this introduces the possibility of alternative explanations for any links between stressors and cytokines. Much less ambiguity surrounds cytokines produced following mitogenic stimulation. The culturing conditions are standardized across subjects and tightly controlled, and any differences in cytokine production can be attributed to individual differences such as stressor exposure. That said, the true meaning of basal measures is clear: a high level of a given cytokine means an immune response is ongoing which, in the case of asthma or arthritis, could worsen symptoms. With mitogen-stimulated cytokine production, cells are taken from their natural environment and placed in culture with (typically) pharmacologic doses of a mitogen. It is unclear how well these artificial conditions simulate what occurs in a patient during real-life exposure to a pathogen or allergen.

PSYCHOLOGICAL STRESS AND TH1/TH2 CYTOKINES

In earlier decades, stress was believed to exert globally suppressive effects on the immune system including reduced humoral immunity, reduced proliferation of lymphocytes, and reduced functioning (cytotoxicity) of naturally killer cells.[1,17,18] This diminished functioning of the immune system was thought to account for high rates of infectious and neoplastic disease in chronically stressed individuals.[19,20]

The immunosuppression model has been critiqued more recently because it cannot explain why stress exacerbates medical conditions that involve activation of the immune response (e.g., asthma, arthritis, cardiac disease). Also, increasing numbers of studies have illustrated that stress may produce *shifts* in cytokine levels or immunomodulation specifically in regard to Th1 and Th2 cytokines rather than suppression.[4,13,14] Th1 and Th2 cytokines have been shown to shift toward a Th2 response in the presence of certain stressors in healthy individuals,[4,14] that is, psychological stressors may result in suppression of one group of cytokines (Th1) and enhancement of another (Th2). Thus, stress may modulate this axis of cytokines instead of universally suppressing it.

In addition, psychological research has grown increasingly sophisticated in its understanding of stress. While diverse stressors were tested earlier, there is a growing consensus that it is important to distinguish the characteristics of stressors, such as duration, frequency, and severity.[21] Furthermore, not all stressors will affect Th1 and Th2 cytokines in the same way; it is important to understand what types of stressors produce what types of effects on these cytokines. In the sections below, we will define different types of psychological stressors and review empirical studies that describe their impacts on Th1 and Th2 cytokines.

Brief Naturalistic Stressors and Th1/Th2 Cytokines

Acute stressors are negative events that are time-limited in duration, perception, and the responses they elicit.[16] One category of acute stressors is the group of brief naturalistic stressors that occur in the real world (as opposed to acute stressors induced in a laboratory) and are time-limited. Among humans, brief naturalistic stressors are often studied because they provide more ecologically valid stress paradigms that can reveal whether immune changes occur in response to stressors encountered in daily life.[16] Examples include academic stress and brief hospital stays.[4,22]

Overall, the literature illustrates that in healthy individuals, the stress associated with taking academic exams causes a shift toward the Th2 response, as seen by a decrease in Th1 cytokines and a corresponding increase in Th2 cytokines.[4,13,14] In the Marshall et al. study of medical students, taking an exam was associated with a significant increase in IL-10 production and a slight decrease in IFN-γ production.[4] They calculated the IFN-γ :IL-10 ratio before and after the exam period in order to assess shifts across the Th1/Th2 axis, and concluded that the stress of exams resulted in a shift along the axis toward a Th2 response as the IFN-γ :IL-10 ratio decreased. In addition, greater reports of daily hassles were negatively correlated with the IFN-γ: IL-10 ratio, illustrating that psychological self-report measures of stress also are associated with the Th1/Th2 shift toward a Th2 response.

Other studies using the exam stress paradigm reported decreased IFN-γ and IL-2 levels during exam periods, illustrating that Th1 cytokines decrease as a result of exam stress.[14,23] In the Paik et al.[23] and Kang and Fox[14] studies, blood samples were obtained from subjects on the day of or during the week of exams; samples used in the Marshall et al.[4] study were obtained several days after the exam period. These differences in timing may explain why Marshall[4] found small but non-significant differences in Th1 cytokines and Paik [23] and Kang and Fox[14] found significant decreases in Th1 cytokines.

A recent review of the stress and immunity literature concluded, after meta-analyses of over 300 studies, that brief naturalistic stressors exert a reliable effect on cytokine production that involves a shift away from Th1 (cellular) immunity, as shown by a decrease in Th1 cytokines, and toward Th2 (humoral) immunity, as shown by an increase in Th2 cytokines.[5]

The consequences of this shift away from a Th1 cytokine response during brief naturalistic stressors may be harmful based on the potential susceptibility to viruses or bacteria to which the Th1 or cellular arm of the immune system responds. If an individual experiences this type of naturalistic stressor and is exposed to a pathogen, it could be more difficult for his or her immune system to resist these pathogens if the cytokines that help coordinate the response are diminished.

Segerstom and Miller[5] also found in their meta-analysis that functional (as opposed to numerical) measures of the immune response such as natural killer cell cytotoxicity and T cell proliferative responses were decreased in the presence of brief naturalistic stress. Th1 cytokines play a vital role in mediating these responses, which aid in the defense and eradication of viruses and other pathogens. This illustrates additional evidence for a link between stress and a Th1/Th2 cytokine shift. In addition, the shift *toward* Th2 cytokines in the presence of brief naturalistic stressors is interesting to consider in regard to inflammatory diseases such as rheumatoid arthritis and asthma,

which have altered cytokine levels that shift toward Th1 and Th2 cytokine profiles, respectively. A shift toward an enhanced humoral response to allergens during a brief naturalistic stressor could promote airway inflammation and obstruction that may affect individuals with asthma. We will discuss these implications later in this chapter.

Chronic Stress and Th1/Th2 Cytokines

In contrast to brief naturalistic stressors, chronic stressors take place over an extended period of time, often with unclear endpoints, and elicit prolonged psychological and biological responses.[5] Chronic stressors in humans are typically studied in naturalistic settings and include stressors such as caring for a chronically ill family member.[16] The literature has shown that chronic unrelenting stress is associated with declines in Th1 and Th2 responses as well as declines in other immune parameters such as natural and specific immune responses.[5] For example, the chronic stressor of serving as a caregiver or having a mother with breast cancer was associated with decreased production of Th1 cytokines including IL-2, IFN-g, and IL-12[17,24,25] as well as decreased production of Th2 cytokines such as IL-4.[26] Thus, chronic stress appears to fit the traditional immunosuppressive theory.

However, not all studies are consistent with this pattern. For example, Glaser et al.[24] found that caregivers expressed greater percentages of IL-10+/CD8+ peripheral blood leukocytes compared to control subjects not experiencing chronic stress. This suggests a shift toward a Th2 cytokine response under chronic stress. They did not find differences in Th1 cytokine expression between caregivers and controls.

Studies investigating the effects of chronic stress have more often examined Th1 rather than Th2 cytokines, and overall reveal a reliable effect of chronic stress decreasing Th1 responses.[5] In contrast, effects on Th2 cytokines may be more uncertain, although a recent meta-analysis concluded that chronic stress reliably decreases Th2 responses when one includes measures of humoral immunity other than cytokine production (e.g., antibodies to vaccination).[5] These types of immunosuppressive effects have important implications for disease susceptibility, including potentially increased risk for infectious and neoplastic diseases under chronic stress.[19,20] At the same time, however, they suggest that diseases characterized by exacerbated inflammatory responses would benefit from the immunosuppressive effects of chronic stress, but this result has not been observed clinically. This suggests that the relationship between chronic stress and Th1/Th2 cytokines in humans is not as straightforward as might initially appear. To help explain associations among chronic stress and inflammatory diseases, we next consider the role of glucocorticoids.

GLUCOCORTICOIDS, TH1/TH2 CYTOKINES, AND PSYCHOLOGICAL STRESS

Glucocorticoids and Th1/Th2 Cytokines

Stress hormones, specifically glucocorticoids, influence the polarization of naïve Th0 cells into Th1 and Th2 cells. Glucocorticoids (which take the form of cortisol in humans) are hormones secreted by the hypothalamic–pituitary–adrenal (HPA)

axis, often after exposure to stressors. Glucocorticoids exert inhibitory effects on Th1 cytokines and enhancing effects on Th2 cytokines.[9,11,27,28] The relationship between glucocorticoids and Th1/Th2 cytokine production and differentiation adds an important layer to the relationships of psychological stress and cytokines.

The literature illustrates that cortisol levels increase when an individual experiences stressful situations where his or her coping resources are not able to counteract the demands of the environment.[28,29] However, another body of literature finds decreased cortisol levels and blunted cortisol responsiveness when individuals experience chronic stressors.[30–32]

The role of glucocorticoids in stress is complex. This section will review the relationship of stress to hyper- versus hypo-cortisolism profiles and discuss reasons for some discrepancies in previous research. We will also discuss biological evidence regarding stress hormones and the shift toward Th2 cytokines and studies illustrating a preliminary link of glucocorticoids, Th1/Th2 cytokines, and human psychological stress. Finally, we will discuss the notion of glucocorticoid resistance in chronic stress and its implications for shifts or dysregulation along the Th1/Th2 axis.

Glucocorticoids and Th1/Th2 Cytokines: Biological Link

Basic immunology research in the areas of stress hormones and Th1/Th2 cytokines reveals that glucocorticoids act on lymphocytes in order to induce the production of Th2 cytokines and decrease the production of Th1 cytokine precursors and consequently Th1 cytokines.[3,9,11,27,33–35] Specifically, glucocorticoids work by suppressing the production of IL-12 by antigen-presenting cells (recall that IL-12 is necessary for Th1 cell development), and by down-regulating IL-12 receptor expression on T-cells and NK cells — cells that help produce IFN-γ when stimulated by IL-12.[11,36,37]

Glucocorticoids work by directly suppressing IL-12 production, preventing the presence of the cytokines necessary for Th1 development and reducing the ability of IL-12 to stimulate the production of other Th1 cytokines such as IFN-γ. In addition, glucocorticoids induce both IL-4 and IL-10 production.[3,9] Thus, in physiological concentrations, glucocorticoids may cause a shift from a Th1 immune response pattern to Th2 via alteration of cytokine production. Agarwal and Marshall[38] empirically demonstrated the ability of glucocorticoids to shift cytokine levels across the Th1/Th2 axis toward a Th2 response by using dexamethasone, a synthetic glucocorticoid, to model cortisol stress responses. They added exogenous dexamethasone (to mimic the release of hormones during the stress response) to human peripheral blood mononuclear cells (PBMCs) and assessed the alterations in Th1/Th2 cytokine levels (specifically IL-12, IFN-γ, IL-10, and IL-4). They found that dexamethasone decreased Th1 cytokines and increased Th2 cytokines, demonstrating a shift toward Th2 cytokines in response to glucocorticoids.

We next discuss the link among stressors, glucocorticoids, and Th1/Th2 cytokines in ecologically valid human stress paradigms.

Brief Naturalistic Stressors, Glucocorticoids, and Th1/Th2 Cytokines

Preliminary Links

As mentioned above, the literature regarding brief naturalistic stress and the Th1/Th2 axis of cytokines supports a shift toward Th2 cytokines. This stressor–cytokine shift is well established, but the role of glucocorticoids in this shift has not been adequately explored. The literature demonstrates that acute laboratory stressors reliably produce increases in cortisol levels.[39] The greatest effects were found with stressors that relate to social evaluation. In addition, numerous other acute stressors such as academic examinations,[40] public speaking,[41] parachute jumping,[42] hostage imprisonment, and public speaking combined with mental arithmetic in a laboratory[45] have been found to stimulate the HPA axis and produce increases in cortisol.

Few human studies have measured glucocorticoids (cortisol) together with Th1/Th2 cytokines in response to stress. Marshall et al. found no difference in plasma cortisol when comparing 2-day post-exam levels to pre-exam (3 weeks preceding exam) levels.[4] However, they found a shift toward Th2 (IL-10) cytokines and a decrease in Th1 (IFN-γ) cytokines resulting in a decreased IFN-γ:IL-10 ratio as a result of the exam stress. A study by Liu et al.[46] focusing on children with asthma also found no increase in cortisol levels during a school examination, although they found increases in IL-5 levels and decreases in IFN-γ levels.

It appears surprising that no increases in cortisol levels were noted in these two studies because cortisol would be expected to facilitate a Th1-to-Th2 shift. However, the timing of cortisol measures in both studies may explain why no changes were detected. Previous studies illustrated that cortisol reaches it peak in circulation 20 to 40 minutes after the onset of an acute laboratory stressor.[29] In basic biological studies, simply culturing naïve (Th0) cells with glucocorticoids can drive Th1/Th2 cell and cytokine differentiation.[27] When using brief naturalistic stressors, however, the optimal time to measure cortisol responses to stress is more ambiguous because the durations and intensities of naturalistic stressors vary. In the studies cited, temporary changes in cortisol may not have been captured based on the timing of assessments. For example, the studies measured cortisol several days after exams started and assessed cortisol concurrently with cytokines.[4,46] Changes in cortisol may have been more apparent if measures of cortisol were taken during or immediately after the exam. In addition, collecting cortisol measures prior to measuring Th1/Th2 cytokines would have allowed researchers to better assess whether cortisol drives the Th1/Th2 shift.

Other stress hormones such as catecholamines have been shown to cause a shift toward Th2 cytokines at the levels of both antigen-presenting cells and Th1 cells.[3] These hormones may be more responsive during times of brief naturalistic stress and consequently may help drive the shift along the Th1/Th2 axis.

Ultimately, the few studies of brief naturalistic stressors, glucocorticoids, and Th1/Th2 cytokines suggest that additional investigations involving repeated measures to clarify the timing of cortisol responses to naturalistic stressors are important. They will allow researchers to correlate changes in cortisol levels with changes in Th1 and Th2 cytokines in the context of psychological stress paradigms.

Chronic Stress, Glucocorticoids, and Th1/Th2 Cytokines

Preliminary Links

The literature regarding chronic stress, glucocorticoids and the Th1/Th2 axis of cytokines is also in its preliminary stages. An increasing body of literature reports that in the face of chronic stress, individuals exhibit decreased or blunted cortisol levels.[31,32,47] However, some researchers found increased cortisol levels in the face of chronic stress.[17,32,48] With respect to cortisol and Th1/Th2 cytokines, Cohen et al.[17] found that cortisol levels were negatively associated with IL-2 levels among individuals whose mothers had breast cancer. This study illustrates a direct relationship between increased glucocorticoids and decreased Th1 cytokine levels under conditions of chronic stress.

Some researchers proposed that chronic stress alters the ability of cortisol to regulate the immune system. For example, Miller et al.[47] found that among adults experiencing chronic stress, immune cells exposed to dexamethasone (a synthetic cortisol) produced higher levels of IL-6 (an integral cytokine in pro-inflammatory responses) compared to the cells of adults not facing chronic stressors. They proposed a model of *glucocorticoid resistance* to explain this finding. Specifically, the model argues that increased cortisol secretion in the presence of chronic unrelenting stress forces the immune system to adapt by down-regulating its glucocorticoid receptors, thus leading to an inability of immune cells to respond effectively to glucocorticoid signals. This results in an inability to shut down certain immune responses that in turn elevates levels of cytokines such as IL-6.[47]

The glucocorticoid resistance model may help explain some of the inconsistent patterns found for cortisol levels and chronic stress. The core idea of glucocorticoid resistance is that the presence of chronic stress results in elevated cortisol levels. Cells then down-regulate their receptors for glucocorticoids to accommodate the elevated levels of cortisol. It is not central to the concept of glucocorticoid resistance that the cortisol levels remain elevated *continuously*, but rather that they remain elevated long enough to alter receptor expression. Indeed, research has shown that sometimes cortisol levels rebound below normal to recover from prolonged periods of elevation.[31] Thus, in humans experiencing chronic stress, elevated cortisol levels may result in glucocorticoid resistance; however, over time, cortisol levels may rebound below normal, resulting in a hypoactive stress system. Thus the different patterns of cortisol with respect to chronic stress may stem in part from differences in the timing of cortisol collection relative to the duration of the chronic stressor.

Consistent with this notion, Bauer et al.[49] found that chronic caregiver stress was associated with decreased lymphocyte sensitivity to glucocorticoids as well as increased salivary cortisol levels. Although this study does not directly support the biological link between increased glucocorticoids and decreased sensitivity to glucocorticoids, it does imply the potential for dysfunction along the Th1/Th2 axis related to cortisol. This idea that altered stress hormones may cause Th1 or Th2 cytokines to remain unregulated may have important implications for inflammatory diseases defined by Th1 or Th2 cytokine predominance — the topic for our next section.

INFLAMMATORY DISEASE AND TH1/TH2 CYTOKINES

Stress has been shown to exacerbate inflammatory diseases such as asthma and rheumatoid arthritis, resulting in increased inflammation and symptomology.[53] However, the mechanism by which stress results in the general worsening of these two conditions is challenging to understand. As noted, asthma is marked by a predominance of a Th2 cytokine profile and RA is marked by a Th1 cytokine profile.[3,12–14] Both diseases are exacerbated by stress. This suggests that a single uniform model of stress, cytokines, and inflammatory disease may not explain both types of diseases.

This section covers the complexities of the interactions of stress and these diseases. We will begin with a discussion of asthma, including cytokine profiles associated with the disease and the impact of stress on immune indicators and clinical symptoms. We will then discuss RA and the role of Th1/Th2 cytokines. The role of altered glucocorticoid functioning in this disease and its implications for Th1/Th2 cytokine expression will also be discussed.

Asthma and Th1/Th2 Cytokines

Asthma is an immune-mediated inflammatory disease characterized by (1) airway obstruction, (2) airway inflammation, and (3) increased responsiveness of the airway to stimuli.[54] Researchers have hypothesized that certain cytokines are important for the orchestration of cellular events related to airway inflammation and hyperresponsiveness.[55] For example, Th2 helper cells secrete cytokines (e.g., IL-4, IL-5, and IL-13) that recruit inflammatory cells and release mediators that result in allergic inflammation, smooth muscle contraction, and mucus production.[3,13,14,54–56] Specifically, Th2 cell secretion of IL-4 and IL-13 induces B cells to switch to producing IgE antibodies.[57]

IgE is responsible for allergic responses and the up-regulation of eosinophil adhesion molecules, leading to obstructed airways and mucus production.[58] The longer lasting inflammatory response involves recruitment of eosinophils to the airways, which also promotes airway inflammation and obstruction. Eosinophil count, in turn, has been associated with symptoms and severity levels of asthma.[59,60] Th2 cell secretion of the IL-5 cytokine has been found to increase eosinophil production. Some researchers have argued that the inflammatory response in asthma involves a Th2 mechanism (IL-4, IL-5, and IL-13 cytokines).[55,61,62]

Research has demonstrated that patients with asthma differ from healthy individuals in their cytokine profiles. Asthma patients have cells that produce higher levels of cytokines such as IL-4 and IL-5 compared to healthy individuals[63–65] and greater expression of mRNA for IL-4 and IL-5.[66–68] Following allergen challenge, levels of IL-4 and IL-13 cytokines increase in patients with asthma.[69]

Stress and Asthma

The psychological role of stress in asthma was cited long before scientists uncovered the biological and immunological pathways linking stress and the disease. In the 19th century, asthma was believed to be a "neurotic affection" instigated solely by psychological stress.[54] Research in the past decades reveals a more sophisticated approach to explaining how psychosocial stress "gets inside" the body to impact the

expression of asthma symptoms. As reviewed earlier, certain types of psychological stress are related to the suppression of cellular (Th1) immunity and heightened humoral (Th2) immunity in healthy people.[5] If stress is also related among individuals with asthma to this type of shift in Th1/Th2 cytokine profiles, it may indicate one pathway to worsening clinical symptomatology in asthma.[70]

Acute Stress and Asthma

Studies have shown links between brief naturalistic stressors and asthma. For example, in a study by Sandberg and colleagues,[71] children between 6 and 13 years of age recorded asthma symptoms and life stressors over an 18-month period using daily diary and interview assessments. Experiencing an acute life stressor was related to increased risk of asthma exacerbations 4 to 6 weeks after the occurrence of the event.[70,71] In daily diary studies of patients with asthma, acute life stressors were associated with same-day lower peak flow rates and greater self-reports of asthma symptoms.[72] The number of asthma exacerbations induced by colds was found to be higher in asthmatic adults who had high numbers of negative life events and low social support.[73] Finally, an intervention involving disclosure of stressful life experiences improved pulmonary function months later in a sample of patients with asthma.[74]

Studies of the impacts of brief naturalistic stressors in patients with asthma revealed altered immune profiles during times of stress. In a study of the responses of 20 college students with asthma to antigen challenges during times of low stress (mid-semester) and high stress (final exam period), results revealed that during high stress, students' eosinophil and IL-5 production increased, consistent with a Th1/Th2 cytokine shift.[46] In another sample of patients with asthma, taking a school exam was associated with greater stimulated Th2 cytokine (IL-5) production in adolescents with asthma compared to healthy control adolescents.[75] IL-5 levels in adolescents with asthma remained elevated even 2 to 3 weeks after examinations compared to control adolescents.[75]

Kang and colleagues[70] reported that the impact of exam stress on immunity was reduced when students reported having high social support. More specifically, students with asthma who had high social support when they were under stress showed smaller reductions in natural killer cell cytotoxicity compared to students with asthma who lacked social support.

On a neuroendocrine level, in individuals with inflammatory diseases such as asthma, the HPA axis is thought to be dysregulated. Subjects with asthma have been described as having hypocortisolistic profiles or blunted HPA axes.[6] Stress may also contribute to this blunted cortisol profile. For example, in response to an acute laboratory stressor (public speaking), children with asthma displayed lower cortisol responses compared to healthy children.[76] The same patterns were found for children with similar inflammatory conditions (atopic dermatitis).[77]

Chronic Stress and Asthma

With respect to chronic stress, studies of children with asthma revealed that chronic stress alters the clinical profile of asthma. For example, among children with asthma who were experiencing chronic stress, an acute life event produced an increased risk

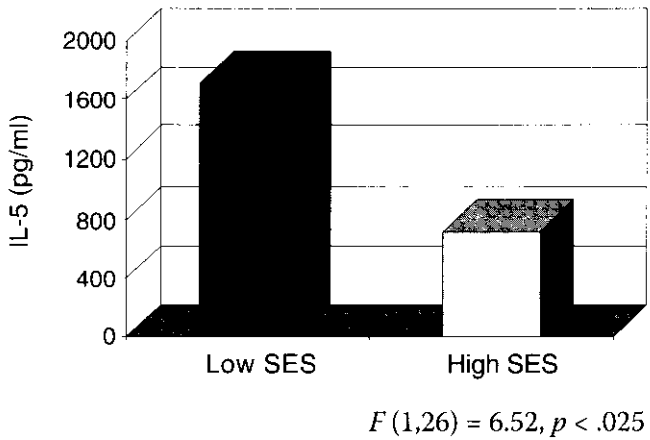

FIGURE 3.1 Asthmatic adolescents from low socioeconomic status neighborhoods had higher stimulated production of the IL-5 Th2 cytokine compared to asthmatic adolescents from high socioeconomic status neighborhoods. (From Chen E et al. (2003). *Psychom. Med.* 65: 984–992. With permission.)

of an asthma attack more quickly (within 2 weeks) compared to children with asthma who did not experience chronic stress.[78] With respect to links to immune measures, adolescents with asthma who came from low socio-economic status (SES) backgrounds had higher levels of stimulated Th2 cytokine IL-5 compared to high SES adolescents with asthma.[79] In addition, chronic stress was found to mediate the relationship between living in a low SES neighborhood and heightened production of IL-5 among adolescents with asthma (see Figure 3.1).

In a study of infants 6 to 18 months old, high levels of chronic stress among caregivers were associated with altered IgE and cytokine expression in their infants, consistent with the patterns of Th2 immunity found in people with asthma.[80] The infants of caregivers who reported greater psychological stress as measured by the Perceived Stress Scale had higher levels of allergen and mitogen-stimulated TNF-α and lower levels of IFN-γ than infants from low-stress households. The number of IgE antibodies was also greater in children whose caregivers reported higher levels of psychological stress. These findings suggest that stress experienced in early life alters Th1/Th2 cytokine profiles toward a dominant Th2 immune response, which in turn may predispose these children to developing chronic inflammatory diseases such as asthma later in life.

Earlier we reviewed evidence documenting that chronic stress had immunosuppressive effects. However, these studies were all conducted in physically healthy individuals undergoing chronic stressor (e.g., serving as a caregiver for a chronically ill family member). In contrast, among individuals with asthma who already have dysregulated immune profiles, it is possible that chronic stress pushes the immune system further toward a Th2 cytokine imbalance, elevating risk for exacerbations of asthma.

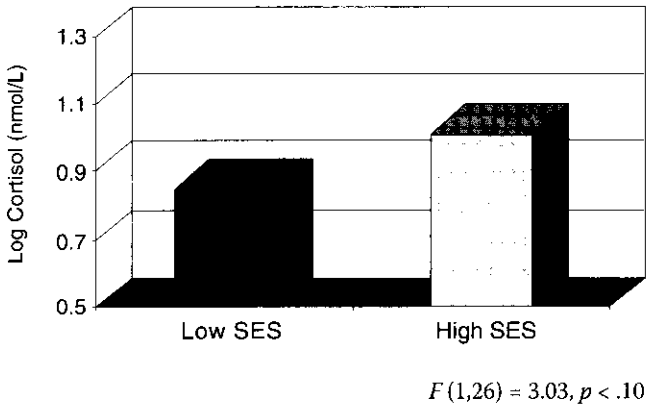

FIGURE 3.2 Asthmatic adolescents from low socioeconomic status neighborhoods had marginally lower morning cortisol levels than asthmatic adolescents who came from higher socioeconomic status neighborhoods. (From Chen E et al. (2003). *Psychom. Med.* 65: 984–992. With permission.)

On the neuroendocrine level, chronic stress also contributes to a blunted cortisol profile. For example, among women with asthma, low serum cortisol concentrations were found in those who had high levels of life stress and low levels of social coping resources.[81] Adolescents with asthma who came from low SES neighborhoods (conceptualized as high stress) had marginally lower morning cortisol values than adolescents with asthma who came from high SES neighborhoods[79] (see Figure 3.2). These blunted cortisol profiles under high chronic stress may allow inflammatory processes to flourish unchecked, resulting in exacerbations of asthma. Overall, both acute and chronic stressors have been associated with heightened production of Th2 cytokines and blunted cortisol profiles in patients with asthma. This suggests that real-life acute and chronic stressors may shift the Th1/Th2 axis of cytokines and glucocorticoid production in a manner that could lead to heightened and unchecked production of Th2 cytokines in the face of exposure to an antigen, resulting in increased airway obstruction and inflammation for patients with asthma.

Rheumatoid Arthritis and Th1/Th2 Cytokines

Rheumatoid arthritis (RA) is a chronic inflammatory disorder of unknown etiology.[82] The tissues around one or more joints trigger an inflammatory response that affects joint mobility and can cause increased pain and difficulty in daily life. It is well established in the literature that the balance of Th1/Th2 cytokines is skewed toward a Th1 response in patients with RA.[3,12,34,83] Specifically, RA involves excesses of IL-12 and TNF-α, but IL-10 production is deficient.[34] Psychological stress has been associated with exacerbations of RA.[84] Daily diary studies revealed that weeks of high stress were associated with more clinician-rated disease activity than weeks of low stress among patients with RA.[85] Increases in the number of stressors experienced from week to week were associated with increases in disease activity.[86] Finally, when

patients with RA reported high levels of social support and coping behaviors during times of chronic stress or major life events, RA symptoms did not change, suggesting that while stress can exacerbate disease severity, coping and social support may mitigate its effects on RA outcomes.[87]

Research on the effects of stress on Th1/Th2 cytokines in patients with RA is limited. Some studies have shown that subgroups of patients with RA who experience significant increases in interpersonal stress also show increases in certain immune measures such as soluble IL-2 receptor levels.[53,85] Other studies reported blunting of Th2 stress responses and Th1 in RA patients, that is, healthy subjects showed increases in both Th2 (IL-10) and Th1 (IFN-g) cytokine production in response to acute laboratory stressors, while these changes were not evident in patients with RA.[88]

Some research relates to cortisol and RA. One study investigated the effects of an acute laboratory stressor on cortisol in RA patients[12] who exhibited significantly smaller cortisol responses to acute laboratory stressors when compared to healthy control subjects without RA.[12] This study illustrated that individuals with RA have decreased cortisol responses to acute stress, and is consistent with previous literature that describes a hypoactive stress system in terms of basal cortisol profiles in patients with RA.[3,6] However, it should be noted that one study found that RA and healthy patients showed similar cortisol responses to experiences of daily stress events.[89]

The literature on stress and RA in humans does not provide much direct evidence about the implications of a hypoactive stress system in RA on the Th1/Th2 axis of cytokines. However, it is possible that the low levels of glucocorticoids found in RA patients may mean a compromised ability to shift the Th1/Th2 axis toward a Th2 profile under stress (the typical effect of glucocorticoids). This could explain why stress has typically been associated with a worsening of symptoms in RA patients rather than the improvement that would be expected if stress shifted the Th1/Th2 axis toward a Th2 profile.

Some studies involved the use of exogenous glucocorticoids such as dexamethasone to challenge the immune response and act as a biological model for stress.[82,83] DeAntonio et al.[83] performed a glucocorticoid sensitivity assay with PBMCs from RA patients to determine the responsiveness of cytokines to glucocorticoids. They found that IFN-γ and TNF-α levels of RA patients remained elevated after administration of dexamethasone when compared to healthy controls. In addition, higher levels of dexamethasone were needed to inhibit TNF-α cytokines in the RA patients. These findings allude to the decreased ability of RA patients to respond to glucocorticoids. Other studies also reported decreased expression of glucocorticoid receptors in cells of RA patients and consequent reduced sensitivity to circulating glucocorticoids.[11,89,90]

Immune systems of RA patients seem to lack responsiveness to glucocorticoids, and as a result it may be difficult for the immune systems of RA patients to downregulate Th1 cytokines. Thus, when the immune systems in RA patients are challenged by stress, there is a blunted cortisol response as well as glucocorticoid resistance on the part of immune cells, both of which could result in an inability to shift away from the disease Th1 cytokine profile toward a Th2 cytokine response during stress.

Some evidence indicates that psychological stress in RA is characterized by blunted glucocorticoid reactivity. In addition, the decreased glucocorticoid receptor expression and glucocorticoid sensitivity in RA may explain the inability to induce Th2 cytokine production and suppress the damaging elevation in Th1 cytokines when stressors are present. The discussion of Th1/Th2 cytokines and stress in RA is very preliminary, although the links between RA and glucocorticoid expression and functioning set the stage for speculation regarding RA and Th1/Th2 cytokines during stress. Future studies that focus on stress-related alterations in both Th1 and Th2 cytokines in the face of glucocorticoid resistance in RA patients are necessary to achieve a better understanding of the mechanisms involved in exacerbations of RA.

CONCLUSIONS AND FUTURE DIRECTIONS

In the past decade, clinical research on the role of psychological stress on the expression of Th1/Th2 cytokines has moved from conceptualizing the effects of stress as immunosuppressive to considering the immunomodulatory effects of stress. Th1 and Th2 cytokines orchestrate different immune pathways to fight pathogens: Th1 cytokines coordinate cellular immune responses and Th2 cytokines coordinate humoral immune responses. Under stress, one type of immunity may be enhanced and another suppressed, resulting in immunomodulatory (rather than globally immunosuppressive) effects.

The effects of stress on Th1/Th2 cytokines depend on the type of stressor investigated. Psychological stress can be characterized according to the duration and resolution of the stressful event. Brief naturalistic stressors occur during daily life and are time-limited stressors (e.g., academic examinations). Studies have shown that this type of psychological stress elicits predominantly Th2 immune responses. In contrast, chronic stress (e.g., caring for a loved one with dementia) is a prolonged experience often with an unclear endpoint. Studies have shown that this type of stress suppresses both Th1 and Th2 responses.

Glucocorticoids, hormones released during times of psychological stress, inhibit the polarization of Th1 cytokines and enhance Th2 cytokine production. Cortisol levels are affected by stress; however the relationship among stress, hormones, and cytokines is complex and differs by the type of stressor measured. Under conditions of acute stress, glucocorticoids increase, perhaps causing a shift toward Th2 immunity. Studies of chronic stress sometimes reported increases and at other times decreases in cortisol. It is possible that glucocorticoid levels depend on time since onset of a chronic stressor; for example, glucocorticoid levels may be high at the beginning of a chronic stressor, but may then rebound below normal levels as the stressor becomes prolonged. This dysregulation of stress hormones may then impair the cross-regulatory functioning of the Th1/Th2 axis, with implications for inflammatory diseases.

Cytokine profiles specific to asthma and rheumatoid arthritis were considered in this chapter as representative of dominant Th2- and Th1-mediated diseases, respectively. Among patients with asthma, stress has been associated with clinical exacerbations of the disease and as also associated with elevations in Th2 cytokines and low levels of cortisol, suggesting immune and neuroendocrine pathways from stress to asthma. Among patients with rheumatoid arthritis, levels of Th1 cytokines were elevated, although these elevations were not definitively linked to stress.

Patients with RA were also found to have blunted cortisol responses to stress and decreased sensitivity to glucocorticoids, suggesting that alterations to the HPA axis may be partially responsible for elevated Th1 cytokines in RA.

A number of recommendations should be considered in determining future directions for research on the relationship of stress, Th1/Th2 cytokines, and inflammatory diseases. First, it is important to understand the timing of when stressors affect both cytokines and cortisol. Future studies should involve repeated assessments after the onset of a stressor to determine when cytokines peak (or shifts occur) and determine when cortisol profiles become altered. Understanding these temporal patterns may help reconcile inconsistencies in previous research studies that used different timing parameters for assessing cytokines and cortisol. These types of assessments are important for both brief naturalistic stressors and chronic stressors. With respect to chronic stressors, it is also important to assess the effects of the duration of the stressor on cytokine and cortisol profiles. Because stressors persist over time, cell receptors may become up-regulated or down-regulated in response to low or high levels of certain cytokines and cortisol. This may in turn shift cytokine and cortisol profiles over the long term. It is also important for future studies to address the interrelationships of Th1/Th2 cytokines and cortisol in studies of psychological stress in humans. Understanding the effects of cortisol on Th1 and Th2 cytokines in response to both brief naturalistic and chronic stressors will help researchers develop a more accurate overall model of how cortisol regulates the Th1/Th2 axis in the faces of different types of stressors.

Additional studies of patient populations are needed to clarify the biological pathways between stress and disease exacerbation. Few studies of patients with RA focus on how different types of stressors affect Th1 versus Th2 cytokines. In addition, few studies of asthma patients target the role of glucocorticoid resistance in the relationship between stress and Th1/Th2 cytokines. Finally, studies that investigate the links from stress to cytokine and cortisol profiles and from cytokine and cortisol profiles to clinical indicators of disease in patients with inflammatory conditions such as asthma and RA are needed.

Intriguing evidence concerns potential pathways from stress to inflammatory diseases operating through Th1/Th2 cytokines and cortisol. Future research directly addressing these issues in patient populations will help researchers develop biologically plausible alternatives to the immunosuppression model to reveal how psychological stress affects inflammatory conditions.

ACKNOWLEDGMENTS

This research was supported by the Canadian Institutes for Health Research and the Human Early Learning Partnership. The authors would like to thank Gregory Miller for helpful comments on this chapter.

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