C57BL/6 Mice Are Resistant to Acute Restraint Modulation of Cutaneous Hypersensitivity

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C57BL/6 mice, in contrast to BALB/c mice, display minimal behavioral changes in response to environmental stressors and are considered relatively stress-resistant. We have shown that application of acute restraint prior to chemical challenge enhanced cutaneous hypersensitivity (CHS) in BALB/c mice and that this enhanced response is partially glucocorticoid dependent. Due to strain differences in the immune response and in the response to environmental stressors, we hypothesized that acute restraint would not enhance CHS in the less stress-sensitive C57BL/6 mice. We sensitized and challenged C57BL/6 mice with the contact sensitizer, 2, 4-dinitrofluorobenzene (DNFB) in the presence and absence of restraint. Acute restraint, applied prior to chemical challenge, significantly increased serum corticosterone, but to concentrations approximately 60% of those reported for BALB/c mice. Neither restraint nor the exogenous administration of corticosterone enhanced chemical-induced ear swelling in C57BL/6 mice. Pharmacological interruption of the hypothalamic pituitary adrenal axis (HPAA) with the glucocorticoid type II receptor antagonist, RU486, did not alter the development of CHS, however, adrenalectomized (ADX) mice exhibited decreased ear swelling, a measurement that was increased further by restraint. Combined application of acute restraint and corticosterone prior to chemical challenge significantly enhanced the ear swelling response in C57BL/6 wild-type mice. These data confirm that C57BL/6 mice have a blunted corticosterone response to restraint and that acute restraint does not modulate cutaneous hypersensitivity. Furthermore, our data demonstrate that stress-resistance is not conferred exclusively through the glucocorticoid pathways.

Key Words: stress; corticosterone; RU486; skin; sensitization.

The skin, due to its external anatomical location, represents a unique immunological interface for interactions with occupational and environmental exposures and also provides an organ system in which to evaluate the interplay between the central nervous system and the peripheral immune response. Over recent years, evidence has accumulated to suggest that stress plays an important role in several types of chronic health problems including modulation of immunologic diseases (Dobbs et al., 1993; Kodama et al., 1999; Zhang et al., 1998). These data suggest that combined exposure to a workplace stressor and a sensitizing chemical may alter the time course and magnitude of the allergic response; however, the molecular interaction between the stress response and the cutaneous immune response is incompletely understood.

Psychological stressors activate the hypothalamic pituitary adrenal axis (HPAA), which in turn, stimulates the release of glucocorticoids and catecholamines from the adrenal gland (Tomaszewksa and Przekop, 1997). Glucocorticoids are major mediators of the stress response and modulate many signaling events in the immune response. Previous studies have shown that inflammatory products, such as the cytokines IL-1 and tumor necrosis factor-α (TNF-α), can activate the HPAA and stimulate the release of neurohormones. Glucocorticoids, such as corticosterone, modulate antigen presentation, cytokine production, T-cell expansion, and natural killer cell activity (Beliso et al., 1982; Bonneau et al., 1997; Chrousos and Gold, 1992; Maes et al., 1998; Snyder and Unanue, 1982; Steer et al., 1998; Wiegers and Reul, 1998).

Acute restraint of mice, which has many physiological similarities to emotional stress in humans, is often used to examine the influence of stress on the murine immune system (Sheridan et al., 1994; Dhabhar et al., 1997, 1999, 2000, 2001; Zhang et al., 2000). However, different strains of rodents respond differently to the effects of restraint (Dhabhar et al., 1995a,b; Sternberg et al., 1992). BALB/c mice appear to be highly responsive to stressors. They produce high levels of plasma adrenocorticotropic releasing hormone (ACTH) and corticotropin releasing hormone (CRH), and they exhibit more anxiety, as determined by behavioral disturbances than do DBA, C3H, and C57 mice (Anisman et al., 1998; Thompson, 1953). Shanks et al. (1990) demonstrated similar basal corticosterone levels in 6 strains of mice; however, in response to a stressor, the magnitude and the rate of clearance of corticosterone differed significantly between the strains. C57BL/6 mice have been reported to be relatively stress resistant and although they have basal corticosterone levels similar to BALB/c mice, they produce lower concentrations of ACTH in response to acute stressors (Anisman et al., 1998; Shanks et al., 1990). We have
previously shown that, contrary to anti-inflammatory effects of chronic stress, acute restraint prior to chemical challenge enhances the chemical-induced ear swelling response and pro-inflammatory cytokine production in the stress susceptible BALB/c mice, and that these changes are partially corticosterone dependent (Flint et al., 2001). We hypothesize that in contrast to our observations in BALB/c mice, restraint stress would not alter the DNFB-induced ear swelling response in C57BL/6 mice because of their blunted HPAA response to an acute stressor.

In this study, we evaluated acute restraint modulation of 2,4-dinitrofluorobenzene (DNFB)-induced cutaneous hypersensitivity in stress-resistant C57BL/6 mice. We measured restraint-induced changes in serum corticosterone and ear swelling, a well-established measure of CHS. To determine if changes in the concentration of serum corticosterone influenced our outcome measures, C57BL/6 mice were treated with exogenous corticosterone or the glucocorticoid receptor antagonist, RU486, in the presence or absence of restraint.

MATERIALS AND METHODS

Mice. Young adult male C57BL/6 mice weighing 20–25 g were purchased from Jackson Laboratories (Bar Harbor, ME). All animal protocols were approved by the NIOSH Animal Care and Use Committee. The animal room was maintained on a 12-h light/dark cycle, and lights went on at 0600 h and off at 1800 h. All animals were given food and tap water ad libitum according to ALAC approved guidelines.

Adrenalectomized (ADX) C57BL/6 mice received 30 μg/ml of corticosterone in their drinking water during transportation and were housed individually. Upon arrival, ADX mice were rested for 5 days following removal of corticosterone from the water and administered 0.9% saline and sucrose in their drinking water throughout the course of the experiment.

Induction of allergic contact dermatitis (ACD). Prior to the experiment, animals were weighed, numbered, and shaved on the back. On days 1 and 2 of the experiment 100 μl of 0.5% 2, 4-dinitrofluorobenzene (DNFB; Sigma-Aldrich, St. Louis, MO), diluted in a vehicle of 4:1 acetone:olive oil (AOO) was applied slowly to the back skin with a micropipette. On day 5, baseline ear thickness was measured for the right and left pinnae. On day 6, the right pinnae were challenged with 50 μl of 0.25% DNFB and the left pinnae were treated with AOO. For application of restraint prior to challenge, mice were restrained for 2 h on day 6. The thickness of the right and left ear pinnae were measured 24, 48, and 72 h after challenge using a digital micrometer.

Manipulation of the HPAA

Restraint. Each mouse was placed in an adequately ventilated 50 ml conical plastic tube (Corning Inc., Corning, NY) for 15 min to 2 h as specified in each experiment. Mice were not physically squeezed and felt no pain. They could rotate from a supine to prone position, but not turn head to tail. Restraint was applied at 1000 h for all experimental manipulations. Nonrestrained mice were left undisturbed in their home cages.

Corticosterone administration. A pilot study was performed to confirm the dose of corticosterone necessary to produce the required serum concentration. Corticosterone (Sigma-Aldrich, St. Louis, MO) was dissolved in polyethylene glycol 400 (PEG), and each mouse received one sc injection of corticosterone (6 mg/kg in 0.01 ml volume in PEG) or PEG only at 1000 h. Mice were evaluated 0, 15, 30, 60, and 120 min after injection. To determine the serum concentration of corticosterone following injection and restraint, mice received an injection of corticosterone at 0800 h and restraint was applied at 1000 h for 0, 15, 30, 60, and 120 min. The effect of exogenous corticosterone on restraint modulation of ACD was evaluated within 4 h following administration, well within the peak response of the drug (Peeters et al., 1992).

RU486 treatment. RU486 is a potent glucocorticoid type II receptor antagonist that can also block the progesterone receptor. Under conditions of our studies and using male mice, we interpreted the effects of RU486 in terms of the type II glucocorticoid receptor antagonism. RU486 peaks in the serum 1 h after injection and has an elimination half life of 86 h (Foldesi et al., 1996). Based on a pilot study in our laboratory, RU486 (Sigma-Aldrich, St. Louis, MO) was dissolved in polyethylene glycol 400 (PEG; Sigma-Aldrich, St. Louis, MO), and each mouse received one sc injection of RU486 (25 mg/kg in PEG; 0.01 ml/g body weight) or PEG 1 h prior to restraint. The effect of RU486 administration on restraint modulation of ACD was evaluated 3 h following administration, well within the peak response of the drug.

Adrenalectomy. Bilaterally ADX C57BL/6 mice were purchased from Taconic (Germantown, NY). This surgical procedure significantly decreases the endogenous source of corticosterone and epinephrine. Auxiliary nodes can produce corticosterone 14–21 days post surgery, therefore all experiments were conducted within 14 days after surgery. Selected mice received 6 mg/kg of corticosterone in PEG or PEG alone 2 h prior to restraint or 4 h prior to challenge.

Corticosterone analysis. Mice were sacrificed immediately following the 2 h restraint period, and blood was obtained by cardiac puncture and collected into plasma separator tubes containing lithium heparin (Becton-Dickinson & Co., Franklin Lakes, NJ). Serum was separated by centrifugation. Fifty μl samples were assayed in duplicate for corticosterone content using an anti-rat corticosterone-coated tube and 121-I-corticosterone tracer protocol (Coat-A-Count RIA kit, DPC Inc., Los Angeles, CA). The RIA was performed according to the manufacturer’s protocol, and samples were analyzed by gamma scintillation and duplicates were averaged. Because serum corticosterone concentrations can vary in mice due to handling techniques, laboratory baseline values were determined. The baseline range at noon in C57BL/6 mice was between 18–32 ng/ml.

Statistical analysis. Data are presented as the mean ± SEM for each experimental group. For the corticosterone data analysis, statistically significant differences (p < 0.05) between restrained and nonrestrained groups were determined by the Student’s t-test.

Ear swelling data analysis was conducted using the SAS software program. Statistical analysis was performed on the raw data. Descriptive statistics such as means and standard deviations were calculated and compared using PROC MIXED, and adjusted for the covariate, initial ear thickness. The analysis combined 2 experiments done in different weeks. Each experiment was assumed to be a random effect that resulted in the interaction term of experiment by treatment being the appropriate error term for testing treatment effects. When analyzing repeated measures data, the REPEATED option was used to model the within subjects covariance structure. The best covariance structure was chosen by using 2 model-fit criteria, Akaike’s Information Criterion (AIC) and Schwarz’ Bayesian Criterion (SBC). The covariance structure that produced the largest value for these criteria was considered the best model-fit. The LSMEANS option was used to calculate means adjusted for unequal sample sizes among treatment groups.

RESULTS

The Effects of Restraint Stress on Baseline Serum Corticosterone Levels

Two-hour restraint is an acute, mild stressor that activates the HPAA, and changes in serum corticosterone provide indirect measure of this activation. To verify that our restraint procedure activated the HPAA in C57BL/6 mice, we first determined the concentration of serum corticosterone in re-

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strained and nonrestrained mice. Two-hour restraint significantly increased serum corticosterone from 31 ± 10.4 ng/ml to 582 ± 15.6 ng/ml, a measurement approximately 60% of that obtained for BALB/c mice under the same experimental manipulation in the same experiment. We previously reported that restraint increased serum corticosterone levels to 800–900 ng/ml in BALB/c mice and that restraint prior to challenge increased DNFB-induced ear swelling and the number of cutaneous inflammatory cells 24 and 48 h after challenge (Flint et al., 2001). These responses were partially corticosterone-dependent.

To extend these observations, we next asked if administration of exogenous corticosterone to C57BL/6 mice would produce serum corticosterone levels similar to the BALB/c mice and promote an enhanced CHS response. We evaluated the administration of exogenous corticosterone at 1000 h in nonrestrained mice at 0, 15, 30, 60, and 120 min following injection in order to verify the time course for the increase of the drug in the serum. Mice that received 6 mg/kg of corticosterone demonstrated a significantly increased serum concentration of corticosterone at 15 min, 1285 ± 122 ng/ml that remained elevated at 956 ± 126 ng/ml at 120 min (Table 1). The combined effect of exogenous corticosterone and restraint produced a similar time course, with no further increase in serum corticosterone attributable to the addition of restraint (Table 2). Mice in this study received the corticosterone injection at 0800 h and restraint was applied for 0, 15, 30, 60, and 120 min, beginning at 1000 h. Serum corticosterone levels in this experiment were not statistically different from the mice that received the injection only, and the t = 0 time point for the injected and restrained mice occurred 2 h after injection and was equivalent in time after injection to the 120 min time point in Table 1.

We also verified the serum corticosterone levels in the ADX C57BL/6 mice. Basal serum corticosterone measured 9.3 ± 2.4 ng/ml; injection with the vehicle control increased that concentration to 82.6 ± 40.5 ng/ml, and corticosterone injection produced a serum concentration of 1398 ± 111 ng/ml. These data verify that 2 h restraint stress enhanced serum corticosterone in intact and ADX C57BL/6 mice and that administration of 6 mg/kg corticosterone by sc injection significantly elevates serum corticosterone to exceed the relative range (800–900 ng/ml) observed in BALB/c mice.

**The Effect of Restraint on Ear Swelling in C57BL/6 Mice Treated with DNFB**

Chemical-induced ear swelling is characterized by edema, erythema, and influx of inflammatory cells throughout the epidermis and dermis that can be measured 24, 48, and 72 h postchallenge. To determine if C57BL/6 mice demonstrate the restraint-enhanced ear swelling response observed in BALB/c mice, C57BL/6 mice were dosed on the back with 0.5% DNFB on days 1 and 2 and dosed onto the ear with 0.25% DNFB on day 6. Restraint was applied prior to chemical challenge. Control mice were sensitized and challenged only. Results are expressed as mean increase percentage of ear swelling compared to baseline ± SEM. Data are representative of 3 independent experiments.

**TABLE 1**

<table>
<thead>
<tr>
<th>Elapsed time (min)</th>
<th>Serum corticosterone (ng/ml)</th>
</tr>
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<tbody>
<tr>
<td>0</td>
<td>21.2 ± 4.5</td>
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<tr>
<td>15</td>
<td>1285 ± 122</td>
</tr>
<tr>
<td>30</td>
<td>1308 ± 72</td>
</tr>
<tr>
<td>60</td>
<td>1214 ± 18</td>
</tr>
<tr>
<td>120</td>
<td>956 ± 126</td>
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</tbody>
</table>

*Note.* Elapsed time following corticosterone injection. Results are expressed as mean ± SEM.

**TABLE 2**

<table>
<thead>
<tr>
<th>Length of restraint (min)</th>
<th>Serum corticosterone (ng/ml)</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>1123 ± 193</td>
</tr>
<tr>
<td>15</td>
<td>1303 ± 74</td>
</tr>
<tr>
<td>30</td>
<td>1317 ± 78</td>
</tr>
<tr>
<td>60</td>
<td>1144 ± 43</td>
</tr>
<tr>
<td>120</td>
<td>841 ± 49</td>
</tr>
</tbody>
</table>

*Note.* Length of restraint (min) following corticosterone injection. Results are expressed as mean ± SEM.

**FIG. 1.** Restraint does not enhance ear swelling in C57BL/6 mice. Groups of mice (n = 5) were topically dosed onto the flank with 0.5% DNFB on days 1 and 2 and dosed onto the ear with 0.25% DNFB on day 6. Restraint was applied prior to chemical challenge. Control mice were sensitized and challenged only. Results are expressed as mean increase percentage of ear swelling compared to baseline ± SEM. Data are representative of 3 independent experiments.
that reached a maximum of 40% at 72 h. Restraint applied prior to challenge in C57BL/6 mice did not change the response at any time point. Ear swelling at 48 and 72 h were 45 and 40% respectively. These data show that, in contrast to our observations with BALB/c mice, the cutaneous immune response to chemical in C57BL/6 mice is insensitive to modulation by acute restraint stress administered prior to chemical challenge.

Because restraint failed to modulate ear swelling in C57BL/6 mice, we hypothesized that the cutaneous immune response is insensitive to the increases in serum corticosterone produced by this strain of mice. To determine if the failure to respond as the BALB/c mice was due to insufficient production of corticosterone, we (1) investigated the effect of exogenous corticosterone in both restrained and nonrestrained mice, (2) blocked the type II glucocorticoid receptor with RU486, and (3) utilized ADX C57BL/6 mice that produce greatly reduced levels of corticosterone and epinephrine and added back corticosterone.

The Effect of Corticosterone Administration on Ear Swelling in C57BL/6 Mice Treated with DNFB

To determine the effect of exogenous corticosterone on DNFB-induced ear swelling, C57BL/6 wild-type mice received a single injection of corticosterone, or the vehicle, PEG, 2 h prior to restraint. Additional mice were treated with corticosterone or PEG and left in their home cages. Although neither restraint nor corticosterone injection alone increased the DNFB-induced ear swelling response, the combination of restraint and corticosterone significantly increased ear swelling at all time points (Fig. 2). Administration of PEG to either restrained or nonrestrained mice did not significantly alter DNFB-induced ear swelling, indicating that the injection-induced increase in serum corticosterone, 80–90 ng/ml, as measured for both wild type and ADX mice was insufficient to effect change in the ear thickness measurements (data not shown).

The Effect of a Glucocorticoid Receptor Antagonist, RU486, on Ear Swelling in Mice Treated with DNFB

To explore further the role of corticosterone in restraint modulation of contact hypersensitivity, we tested the effect of RU486 on the development of CHS in C57BL/6 wild-type mice (Fig. 3). Two receptors for glucocorticoids have been characterized, the high affinity type I receptor and the type II receptor that has a 3–5 fold lower affinity for corticosterone (Reul and de Kloet, 1985). Under basal conditions, corticosterone binds to the type I receptor, and upon saturation of the type I receptor, corticosterone binds the lower affinity type II receptor. Therefore, in our experimental paradigm, RU486 would block the systemic effects of a stress-induced increase in, or exogenously administered, corticosterone that were mediated through the type II glucocorticoid receptor (Flint et al., 2001; Ratka et al., 1989). In this study, mice were treated with a single application of RU486 or the vehicle, PEG, 1 h prior to restraint. Another group was restrained, and a third group was left in their home cages. Although a significant decrease in ear swelling was observed in 3 mice at 24 and 48 h, we observed no statistically significant differences in ear swelling in duplicate experiments for restrained mice and restrained mice treated with RU486 for any time points (n = 8 mice). At 24 h, a 12–20% increase in DNFB-induced ear swelling was observed, the response peaked at 28–32% at 48 h and declined to 24% at 72 h. The DNFB-induced ear swelling response was not altered in PEG-treated and restrained mice (data not shown). These data demonstrate that although these mice have elevated concentrations of corticosterone, blocking the low affinity type II receptor has no significant effect on the ear swelling response.

The Effect of Adrenalectomy on Ear Swelling

To evaluate an interaction between restraint and chemical-induced ear swelling in the absence of corticosterone and
Epinephrine, C57BL/6 ADX mice received topical applications of DNFB and corticosterone injection (6 mg/kg). Nonrestrained C57BL/6 ADX mice displayed increased ear swelling measurements of 20% at 24 h, 26% at 48 h, and 18% at 72 h (Fig. 4), a diminished response compared to C57BL/6 wild-type mice (Fig. 1). Although restraint did not modulate significantly the ear swelling response at any time point, we observed a decrease in the mean ear thickness when restraint and corticosterone were applied to mice separately and a restoration to nonrestrained levels when restraint and exogenous corticosterone were administered together. Although the injection procedure constitutes a mild stressor, as evidenced by the increase in serum corticosterone in PEG-injected ADX mice (Table 3), vehicle injection had no significant effects on the ear swelling response in restrained and nonrestrained ADX mice, suggesting that this increase was not physiologically significant in our experimental paradigm. These data show that, in the absence of a full HPAA activation there is a less intense ear swelling response to DNFB, and this response is not altered by restraint.

**DISCUSSION**

In the present study we demonstrated that, in contrast to our observations in BALB/c mice, restraint activation of HPAA prior to chemical challenge fails to enhance the ear swelling response in C57BL/6 mice. We have further demonstrated that exogenous administration of corticosterone in the absence of restraint has no effect on the ear swelling response, however, the combination of restraint and administration of corticosterone significantly enhances ear swelling.

Following initial application of chemical to the skin, the release of proinflammatory cytokines triggers LC migration from the epidermis to the draining lymph nodes where they activate CD4$^+$ and CD8$^+$ lymphocytes (Kimber, 1994). In response to cutaneous challenge with the same chemical, these activated leukocytes return to the skin to mount an antigen specific cell-mediated immune response. Glucocorticoids modulate the contact hypersensitivity response at many levels. For example, corticosterone can affect trafficking of lymphocytes from the peripheral blood to the lymph nodes and to sites of active inflammation (Davenpeck et al., 1998; Dhabhar et al., 1996; Schleimer, 1993), vascular permeability and edema, or directly influence proinflammatory cytokine production (Flint et al., 2000). These modulatory events are frequently accomplished by changes in the concentration of intercellular signaling molecules or in the pattern of receptor expression.

<table>
<thead>
<tr>
<th>TABLE 3</th>
<th>Mean Concentrations of Corticosterone in ADX Mouse Sera</th>
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<tbody>
<tr>
<td>Serum corticosterone (ng/ml)</td>
<td></td>
</tr>
<tr>
<td>Restrained mice</td>
<td>9.3 ± 2.4</td>
</tr>
<tr>
<td>Restrained mice treated with PEG</td>
<td>82.6 ± 40.5</td>
</tr>
<tr>
<td>Restrained mice treated with corticosterone</td>
<td>1398 ± 111</td>
</tr>
</tbody>
</table>

**FIG. 3.** The effect of RU486 applied prior to challenge on ear swelling C57BL/6 mice. Control groups of mice (n = 4) were sensitized onto the flank with 0.5% DNFB on days 1 and 2 and challenged onto the ear with 0.25% DNFB. Mice were injected sc with 25 mg/kg of RU486 in PEG (0.01 ml/g body weight), or PEG alone, 1 h prior to restraint or 3 h prior to challenge. Nonrestrained groups were left alone in their home cages until challenged onto the ears. Results are expressed as mean increase percent of ear swelling compared to baseline ± SEM. Data are representative of 2 independent experiments.

**FIG. 4.** The effect of ADX on ear swelling in C57BL/6 mice. Groups of mice (n = 3) were sensitized onto the flank with 0.5% DNFB on days 1 and 2 and challenged onto the ear with 0.25% DNFB on day 6. Mice received 6 mg/kg of corticosterone in PEG or PEG alone, 4 h prior challenge. Results are expressed as mean percentage of ear swelling compared to baseline ± SEM.
We have previously shown that restraint before chemical challenge significantly enhanced serum corticosterone and DNFB-induced ear swelling in BALB/c mice and that corticosterone was partially responsible for the enhanced ear swelling (Flint et al., 2001). In addition, other studies have shown that restraint stress enhances these parameters in rats and B6,129 mice, and Dhabhar and colleagues confirmed the additive effects of glucocorticoids and epinephrine in the enhanced response in rats (Dhabhar et al., 1996). In this study we have demonstrated that, in spite of similar basal levels of serum corticosterone, C57BL/6 mice produce significantly lower levels of corticosterone in response to 2 h restraint than do BALB/c mice. In our laboratory, BALB/c mice typically produce 800–900 ng/ml corticosterone in response to restraint whereas C57BL/6 mice routinely show an elevation of 450–500 ng/ml (Flint et al., 2001).

We also demonstrated that, in restrained mice, administration of corticosterone enhances DNFB-induced ear swelling and that surgical interruption of the HPAA diminished ear swelling, an effect that cannot be overcome by administration of exogenous corticosterone. Responses to stressors are initiated by the hypothalamus and translated by the HPAA and autonomic nervous system. Products from both systems, corticosteroids and catecholamines respectively, are able to modulate the activity of immune effector cells to restore homeostasis in the mouse. Studies have shown that strains of mice differ in terms of adrenal response and that acute stress does not significantly alter levels of pituitary CRH and plasma ACTH in C57BL/6 mice, a possible explanation for the lower levels of serum corticosterone we observed (Anisman et al., 1998). Interestingly, administration of either restraint or exogenous corticosterone to ADX mice depressed the mean ear swelling response. The adrenalectomy was performed prior to chemical sensitization, and we have shown previously that restraint applied to a naive mouse immediately before chemical sensitization suppresses development of contact hypersensitivity (Flint et al., 2001). It is likely that immune suppression following a major surgical procedure such as adrenalectomy will have an extended time course. In addition to severely limiting corticosteroid production, adrenalectomy also interrupts epinephrine production. Others have previously shown that epinephrine contributes to the enhanced ear swelling in ADX mice (Dhabhar and McEwen, 1999). These findings highlight the importance of catecholamines in the stress response and the multifactorial link between the stress response and the cutaneous immune response.

Many studies have documented that corticosterone binds first to the high affinity type I receptor and only after saturation of the type I receptor does corticosterone bind the lower affinity type II receptor. Because RU486, a type II receptor antagonist, had no significant effects on ear swelling and increased corticosterone did not modulate cutaneous hypersensitivity in C57BL/6 mice, it is possible that there are strain dependent limitations in surface expression of the type II corticosterone receptor or changes in the receptor affinity. Consistent with this theory, earlier work suggested that corticosterone binding capacity in the hippocampus is lower in C57BL/6 mice than BALB/c mice, a condition that may extend to other organs in this mouse (Patacchioli et al., 1990). These data suggest that strain dependent differences in the HPAA underlie acute stress modulation of cutaneous hypersensitivity.

Taken together, our data demonstrate that C57BL/6 mice produce moderate levels of corticosterone in response to 2 h restraint and that increases in the concentration of serum corticosterone alone do not produce the enhanced ear swelling response observed in BALB/c mice. These studies support our hypothesis that acute restraint stress applied before chemical challenge has no effect on ear swelling in stress resistant C57BL/6 mice and suggest that strain-specific differences in the stress response impact the magnitude and direction of the cutaneous immune response to chemical. Additional studies are required to determine if, in humans, the combination of workplace stress and chemical exposure may alter the exposure-disease paradigm and indicate the need for additional considerations when establishing protective measures.

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REFERENCES


