population crashes (7). Reducing the parasite burden reduces the variance in the population growth rate and produced an apparent reduction in the decline of the treated populations (Fig. 2). This experiment illustrated that parasitic nematodes were necessary for the cyclic declines in abundance that were observed in grouse populations. In both populations that were treated twice (Fig. 2C) and in one of the populations that was treated once (Fig. 2B), the effect of the treatment was apparent in comparison with the controls, although the results are less clear in the remaining population, which was treated just once. We suspect this was because the keeper treated a relatively low proportion of the grouse population (∼15%). Even with these results, the findings were still significant and demonstrate that parasites played a key role in causing population cycles.

To determine the effectiveness of the treatment, we calculated the proportion of the population that should be treated in order to prevent a population crash. We addressed this problem with a modified form of the general macroparasite model (5) that incorporates the experimental protocols of direct oral treatment (8) (Table 1). Individuals in the model were classified as either untreated (with natural levels of infection) or treated (with no parasites). Treatment of a proportion (p) of the population was triggered in the model whenever the growth rate of the parasite population increased (becomes positive). The worms in the treated grouse suffered an increased mortality rate, so their life expectancy was <1 week, whereas the remaining untreated birds (1 – p) continued to release infective stages into the environment, which infected both treated and untreated hosts. Numerical solutions of the model’s dynamics showed that treatment of >20% of the hosts was sufficient to prevent the cyclic crashes in host density (Fig. 3) and provided a good explanation for all the results of the experiment.

The results from this study show that population cycles in red grouse are the result of a single trophic interaction between a parasite and its host. Combined with the modified macroparasite model, these results show that parasites were both sufficient and necessary in causing cycles in these populations. They also show that intrinsic mechanisms do not need to be evoked as a cause of cyclic fluctuations in grouse abundance (9). Previous studies have undertaken detailed experiments at a lower spatial scale. For example, a factorial manipulation of the food and predators of snowshoe hares on 1-km² plots indicated that at least three trophic levels of interaction are involved in producing cycles (3). Nevertheless, to the best of our knowledge, this is the first time that manipulations of a mechanism in a cyclic species have demonstrated the cause of population cycles on a large scale.

Recent decades have brought dramatic increases in the prevalence and severity of allergic asthma. In the United States, 15 million people are currently thought to suffer from the disorder (1). Allergic asthma is characterized by airway hyperresponsiveness (AHR) to a variety of specific and nonspecific stimuli, chronic pulmonary eosinophilia, elevated serum immunoglobulin E (IgE), and excessive airway mucus production (2). The pathophysiology of asthma is thought to be mediated by CD4+ T lymphocytes producing a type 2 cytokine profile: (i) CD4+ T cells are necessary for the induction of allergic asthma in murine models; (ii) CD4+ T cells producing type 2 cytokines undergo expansion in these models and in patients with allergic asthma; and (iii) the amount of type 2 cytokines is increased in the airways tissues of asthmatics and animal models (3–5). The circumsitual evidence for the importance of IL-4 and IL-5, which are the paradigmatic type 2 cytokines, has been compelling (6–8). However, although an antibody-mediated blockade of IL-4 during allergen sensitization ablates the development of allergic asthma, a similar blockade of IL-4 before or during an antigen challenge inhibits neither allergic in-
examined the role of IL-13 in allergic asthma. A well-characterized murine model of allergic asthma was used, in which allergen exposure results in AHR, pulmonary eosinophilia, increases in antigen-specific serum IgE amounts, and increases in airway epithelial mucus content (12). Male A/J mice were immunized intraperitoneally and were subsequently challenged intratracheally with soluble ovalbumin (OVA); the allergic phenotype was assessed 4 days after the antigen challenge (13). Blockade of IL-13 was performed by the systemic administration of a soluble IL-13Rα2-IgG fusion protein (sIL-13Rα2-Fc), which specifically binds to and neutralizes IL-13, 24 hours before subsequent intratracheal allergen challenges (14). Antigen challenge of allergen-immunized mice resulted in significant increases in airway responsiveness to acetylcholine (15) (Fig. 1A). Blockade of IL-13 resulted in a complete reversal of such allergen-induced AHR; thus, IL-13 is necessary for the expression of AHR in this model. The ability of IL-13 ablation to reverse AHR after the full development of the phenotype of allergic asthma contrasts with the inability of IL-4 ablation to accomplish such a reversal. The mechanism underlying the effectiveness of IL-4Rα blockade in reversing allergen-induced AHR (12) may be the inhibition of IL-13–mediated processes, which is consistent with the fact that Stat6 activation is downstream of IL-4Rα–mediated signaling for both cytokines. IL-13 is probably the primary CD4+ T cell–derived factor responsible for allergen-induced AHR.

To evaluate the candidate mechanisms underlying IL-13–dependent expression of AHR, we characterized known allergic effector cascades. Eosinophils have been implicated as primary effector cells in asthma and asthmatic AHR (16), but the inhibition of IL-13 before repeat antigen provocation did not significantly affect allergen-induced pulmonary eosinophilia (17) (Fig. 1B). To assess the relevance of IgE-mediated pathways, we measured OVA-specific serum IgE (18).
OVA-specific IgE was observed in OVA-sensitized and OVA-challenged mice, whereas no antigen-specific antibody was detected in phosphate-buffered saline (PBS)-immunized and PBS-challenged mice (Fig. 1C). Blockade of IL-13 did not alter OVA-specific IgE concentrations—a lack of suppression that is likely due to the fact that the IL-13 blockade occurred after initial antigen priming and antibody formation. Nonetheless, these results show that AHR is not dependent on IgE production in this model, which is consistent with a report that allergic AHR blocks normally in IgE-deficient mice (19).

In congruence with the pathology of human asthma, allergic asthma in murine models is associated with a substantial increase in the mucus content of the airway epithelium (7, 12). Mucus hypersecretion is particularly profound in autopsied specimens from patients who die of acute asthma attacks (20). Blockade of IL-13 reverses allergen-induced increases in mucus-containing cells in the airways (Fig. 2), demonstrating that allergen-induced increases in airway mucus content are dependent on IL-13. IL-4 has also been implicated in this process, because IL-4 transgenic mice display goblet-cell hyperplasia in the absence of antigen sensitization (7).

However, the transfer of Tg2 clones from IL-4–deficient mice into murine airways induces mucus overproduction (21), which suggests that the immunoregulatory role of IL-4 should be carefully differentiated from its role as an effector molecule.

If IL-13 is necessary for the expression of allergic AHR, is it sufficient to induce it? The daily administration of recombinant IL-13 (rIL-13) to the airways of naïve (unimmunized) mice induced AHR, demonstrating that increases in IL-13 activity were sufficient to induce AHR (Fig. 3A) (22). AHR developed within 72 hours from the start of rIL-13 administration. A significant influx of eosinophils into bronchoalveolar lavage (BAL) fluid was observed soon after rIL-13 administration; however, pulmonary eosinophilia was not observed at the time of expression of AHR (Fig. 3B). Although the importance of the time course of eosinophil influx remains unclear, it suggests that IL-13 alone may be sufficient to initiate eosinophilic infiltration of the airways, perhaps through its ability to up-regulate chemokine expression (23).

Airway administration of rIL-13 also resulted in a time-dependent increase in total serum IgE (Fig. 3C) (24), which is in line with the ability of IL-13 to regulate IgE synthesis (25). Increases in serum IgE were independent of any immunization with allergen; these findings are consistent with the observation that the human asthmatic phenotype correlates better with total, rather than allergen-specific, serum IgE concentrations (26). As predicted from our IL-13 inhibition studies, the administration of rIL-13 induced an increase in airway mucus production (Fig. 3D) (27).

Although IL-13 thus appears capable of inducing the entire allergic asthmatic phenotype, the results of the IL-13 blockade experiments clearly show that IL-13–dependent AHR occurs by mechanisms that are independent of IgE and eosinophil in this model. The exact mechanism or mechanisms by which IL-13 induces AHR are currently unknown. The delayed time course for AHR induction suggests that IL-13 does not directly cause airway smooth muscle constriction. Reasonable hypotheses include direct time-dependent alterations in smooth muscle function (IL-13 receptors have yet to be demonstrated on airway smooth muscle) and indirect effects that are achieved through mediators released by surrounding cells. Although recent studies have suggested a possible role for sensory neuron–derived tachykinins in AHR, preliminary studies in our laboratory do not support a role for these neuropeptides in IL-13–induced AHR (28).

Our data demonstrate a critical role for IL-13 in the expression of murine asthma and suggest that, although IL-4 may be of immunoregulatory importance, IL-4 is not a prime effector molecule. These findings may be relevant to human asthma. Overexpression of IL-4 is predominantly found in the airways of allergic asthmatics, whereas significant elevations in IL-13 expression are found in the airways of patients with both allergic and nonallergic asthma (4, 29). Human asthma has been linked to a region of chromosome 5q, which contains the genes for both IL-4 and IL-13 (30). Although polymorphisms in the IL-13 gene have yet to be examined, polymorphisms in the IL-4 gene are well-described (31). No significant correlations between such polymorphisms and the asthmatic phenotype have been found; however, a gain-of-function mutation in IL-4Rx was recently shown to be associated with asthma (32). These insights into the immunopathogenesis of allergic asthma should provide direction for the development of therapeutics for this increasingly prevalent disease.
The pathogenesis of asthma reflects, in part, the activity of T cell cytokines. Murine models support participation of interleukin-4 (IL-4) and the IL-4 receptor in asthma. Selective neutralization of IL-13, a cytokine related to IL-4 that also binds to the α chain of the IL-4 receptor, ameliorated the asthma phenotype, including airway hyperresponsiveness, eosinophil recruitment, and mucous production. Administration of either IL-13 or IL-4 conferred an asthma-like phenotype to nonimmunized T cell–deficient mice by an IL-4 receptor α chain–dependent pathway. This pathway may underlie the genetic associations of asthma with both the human 5q31 locus and the IL-4 receptor.

Allergic asthma is a complex disorder characterized by local and systemic allergic inflammation and reversible airway obstruction. Asthma symptoms, especially shortness of breath, are primarily related to airway obstruction, and death is almost invariably due to asphyxiation (4). Increased airway responsiveness to provocative stimuli, termed airway hyperresponsiveness (AHR), and mucus hypersecretion by goblet cells are two of the principal causes of airway obstruction observed in asthma patients (2). Data from animal models consistently reveal a critical role for T(H)2 (T helper 2) cells and IL-4 in the development of asthma, with both the human 5q31 locus and the IL-4 receptor.

We revealed a phenotype in BALB/c mice deficient in either IL-4 or the IL-4 receptor α chain (IL-4Rα) (9). After intranasal challenge with the antigen ovalbumin (OVA), BALB/c mice developed a stereotypical asthma phenotype characterized by eosinophil influx of the airways, goblet cell metaplasia with mucus overproduction, and an increase in AHR as revealed by enhanced sensitivity to acetylcholine challenge (6, 7). IL-4 and IL-4Rα–deficient mice showed incremental attenuation of each of these asthma indices (Fig. 1, C through E) (10). Thus, in agreement with prior studies (5–7), IL-4 contributes to the asthma phenotype, but these data suggest an independently greater contribution by IL-4Rα.

IL-13 is a cytokine closely related to IL-4 that binds to IL-4Rα and is also expressed by T(H)2 cells from asthma patients (11). To assess whether IL-13 might contribute to the asthma phenotype in BALB/c mice deficient in either IL-4 or the IL-4 receptor α chain (IL-4Rα) (9), we used an intranasal challenge with the antigen ovalbumin (OVA).

Fig. 1. PAS-stained histologic sections of murine lungs. Arrowheads point to goblet cells within the respiratory epithelium. (A) Wild-type mice were primed with OVA and challenged with PBS intranasally. (B) Wild-type mice were administered IL-13 intranasally. (C) IL-4–deficient and (D) IL-4Rα–deficient mice were primed with OVA and challenged with OVA intranasally. Wild-type mice were primed with OVA and challenged intranasally with (E) OVA and human Fc control protein or with (F) OVA and IL-13R-Fc. Note the marked reduction in goblet cells in (D) and (F).
Editor's Summary

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