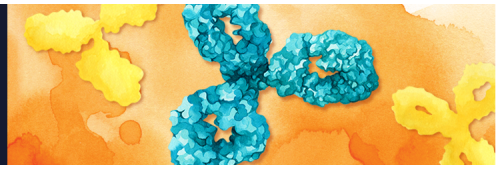


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Histamine Potently Suppresses Human IL-12 and Stimulates IL-10 Production via H2 Receptors

Iliia J. Elenkov,^{1*} Elizabeth Webster,[†] Dimitris A. Papanicolaou,[†] Thomas A. Fleisher,[‡] George P. Chrousos,[†] and Ronald L. Wilder^{*}

IL-12 and IL-10, respectively, stimulate Th1 and Th2 immune responses. The development of some allergic reactions, infections, and tumors are associated with excessive histamine production and a shift toward Th2 responses. Here we address the possibility that this association is causally linked, at least in part, to modulation of IL-12 and IL-10 production by histamine. We report that histamine dose-dependently inhibited the secretion of human IL-12 (p70) and increased the production of IL-10 in LPS-stimulated whole blood cultures. These effects of histamine were antagonized by cimetidine, an H2 receptor antagonist, but not by selective H1 and H3 receptor blockers, and were mimicked by an H2 receptor agonist. The effects of histamine on IL-12 and IL-10 secretion were independent of endogenous secretion of IL-10 or exogenous addition of IL-12, while Ro 20-1724, a phosphodiesterase inhibitor, potentiated the effects of histamine on IL-12 and IL-10 production, implicating cAMP in its actions. Similar modulatory effects of histamine on IL-12 and IL-10 production, which were reversed by the H2 antagonist cimetidine, were observed in PBMC and isolated monocytes stimulated by *Staphylococcus aureus* Cowan strain 1 and LPS, respectively. Thus, histamine, via stimulation of H2 receptors on peripheral monocytes and subsequent elevation of cAMP, suppresses IL-12 and stimulates IL-10 secretion, changes that may result in a shift of Th1/Th2 balance toward Th2-dominance. This may represent a novel mechanism by which excessive secretion of histamine potentiates Th2-mediated allergic reactions and contributes to the development of certain infections and tumors normally eliminated by Th1-dependent immune mechanisms. *The Journal of Immunology*, 1998, 161: 2586–2593.

Interleukin-12, a heterodimeric cytokine produced mainly by monocytes/macrophages, is a central inducer of cell-mediated immunity by promoting the development, proliferation and function of Th1 cells (1). Th1 cells promote the activation and function of NK, T-cytotoxic (Tc)² cells, and monocytes/macrophages, which are the principal effectors of cellular immunity (1–2). Excessive production of IL-12 may be involved in the pathogenesis of autoimmunity. For example, a recent study indicated that stimulation of IL-12 secretion by microbial products was the crucial factor for the proliferation and differentiation of pathogenic autoreactive Th1 effector cells in experimental allergic encephalomyelitis (3). IL-12 also induces tissue- or organ-specific inflammatory responses resulting in insulin-dependent diabetes mellitus in NOD mice (4) and type II collagen-induced arthritis in DBA/1 mice (5). Conversely, IL-12 deficiency has been documented in tuberculosis and HIV infections (6), and low levels of IL-12 have been associated with tumor growth, as opposed to tumor regression observed with administration of IL-12 delivered in situ or systemically (7).

In contrast to IL-12, IL-10 inhibits several macrophage functions. Together with IL-4, this cytokine stimulates the development, proliferation, and function of Ab-producing B cells and promotes IgE production (8). IL-10 also prevents Ag-specific T cell proliferation, inhibits the production of the principal proinflammatory cytokines IL-12, TNF- α , and IFN- γ , and prevents endotoxin shock in mice (reviewed in Ref. 9). In humans, initial clinical trials demonstrated that IL-10 administration ameliorated inflammatory symptoms associated with endotoxemia (10), inflammatory bowel disease (11), and rheumatoid arthritis (12). Overexpression of IL-10 seems to play an inappropriate immunosuppressive role, allowing increased malignant tumor growth, as seen in melanoma (13) and in systemic diseases with excessive production of Abs, such as the immune complex-related manifestations of lupus erythematosus (14).

Histamine, a recognized regulator of inflammation, is also known for its role in promoting allergic reactions, gastric acid secretion, and tumorigenesis. As a reflection of its importance, histamine-blocking agents, worldwide, are the largest selling drug group today. Atopic states, associated with excessive histamine production, involve a shift from Th1 to Th2 responses and increased IgE production. On the other hand, chronic gastritis with progression into peptic ulcer disease is also associated with increased histamine production in the gastric mucosa. Chronic gastritis is largely caused by infection with *Helicobacter pylori*, against which Th1 responses appear to have a protective role (15). Finally, suppression of Th1 responses accompanies the development of diverse tumors, while, in some tissues, histamine itself appears to promote tumorigenesis (16). Interestingly, recent evidence indicates that the use of H2 receptor antagonists increases survival of patients with gastric and colorectal cancer (17, 18).

Thus, the question arises: Does histamine directly induce or promote a Th2 shift? Theoretically, histamine, or any other mediator,

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² Abbreviations used in this paper: Tc, T-cytotoxic; NOD, nonobese diabetic; SAC, *Staphylococcus aureus* Cowan strain 1; ND₅₀, 50% neutralizing dose.

could alter the Th1/Th2 balance at the level of APC, Th1 and Th2 cells, or directly on effector cells. Since IL-12 and IL-10, as noted above, have important regulatory influences on Th1 and Th2 functions, we speculated that histamine could modulate IL-12 and IL-10 production and, thus, Th1/Th2 balance. Here we report that histamine, via an H2 receptor-mediated process, potentially suppressed the production of human IL-12, while it enhanced the secretion of IL-10. This may represent a mechanism through which histamine skews Th1/Th2 cytokine balance toward Th2-type dominance.

Materials and Methods

Drugs and reagents

Flow cytometric evaluation of manipulated cell preparations was performed using directly conjugated mAbs to the following surface Ags: CD45, CD14, CD3, CD20, CD16/56 (Becton Dickinson Immunocytometry Systems (BDIS), San Jose, CA). LPS from *Escherichia coli*, serotype K-235 (Sigma, St. Louis, MO), was dissolved in distilled water; the mixture was sonicated for at least 3 min in a sonicating bath, and aliquots were stored at -20°C until use. After thawing, appropriate dilutions were made in RPMI 1640. *Staphylococcus aureus* Cowan strain 1 (SAC) was obtained from Calbiochem-Behring, (La Jolla, CA). Histamine, dimaprit, cimetidine, pyrilamine maleate (mepyramine), thioperamine maleate, and Ro 20-1724 were purchased from Research Biochemicals (Natick, MA). Anti-human IL-10 Abs and recombinant human IL-12 were obtained from R&D Systems (Minneapolis, MN).

Blood donors

Thirty-five healthy male and female volunteers between 20 and 40 years old participated in this study, which was approved by an institutional review board of the National Institutes of Health. Volunteers abstained from using any drug, including antihistamines, cyclooxygenase inhibitors, and hormones during the week before the study.

Whole blood cultures

The whole blood assay was established recently as a suitable *ex vivo* model by which to study cytokine production under conditions in which many of the physiologically relevant cellular interactions and natural microenvironments remain intact (19). In addition, this method offers the opportunity to induce and detect cytokine production in small blood samples and to study multiple samples under different conditions, employing the blood from the same donor, and, thereby, eliminating interdonor variability as a confounding factor. In brief, blood was drawn into sodium heparin-containing sterile blood collecting tubes (Vacutainer, Becton Dickinson, Lincoln Park, NJ). Blood was transferred to 50 ml Falcon tubes, diluted 1:5 with RPMI 1640 (supplemented with 1% glutamine and gentamicin, 50 $\mu\text{g}/\text{ml}$) with no added exogenous serum, and aliquoted (1.0 ml) into 24-well cell culture plates (Costar, Cambridge, MA). To induce cytokine production, bacterial LPS was added at 1 $\mu\text{g}/\text{ml}$ final concentration, and the samples were incubated in 5% CO_2 at 37°C for 18 h. Histamine and the H2 receptor agonist dimaprit were added 10 min before LPS. Histamine antagonists and the phosphodiesterase inhibitor Ro 20-1724 were added to the wells 10 min before histamine. Anti-IL-10-neutralizing Abs or recombinant human IL-12 were added simultaneously with LPS. After incubation, the blood was centrifuged, and the supernatant plasma was collected and stored in polypropylene tubes at -70°C until assayed.

PBMC, PBL, and monocyte isolation and culture

Human peripheral blood samples were obtained by leukapheresis of volunteers at the National Institutes of Health Department of Transfusion Medicine. PBMC were isolated by density gradient centrifugation using Ficoll-Hypaque (Histopaque, Sigma). To obtain monocyte-enriched preparations, total PBMC were depleted of T and B lymphocytes using magnetic beads (Dyna, Lake Success, NY) coated with mAb specific for the CD2 (Pan T cells) and CD19 (Pan B cells) surface Ags. Total PBMC at 10 to 15×10^6 cells/ml were incubated (two times, with >10 beads per target cell) and gently mixed with the magnetic beads at 4°C for 30 min. The bead-cell complexes were eliminated using magnetic separation. The monocyte-enriched preparations contained 73 to 89% monocytes ($n = 4$), as determined by flow cytometry (FACScan, BDIS) with monocyte-specific FITC-conjugated anti-CD14. Lymphocyte contamination ranged from 11 to 27% and consisted of 60 to 75% NK cells, 20 to 35% T cells, and $\leq 3\%$ B cells. Lymphocytes (PBL) were obtained by depleting monocytes

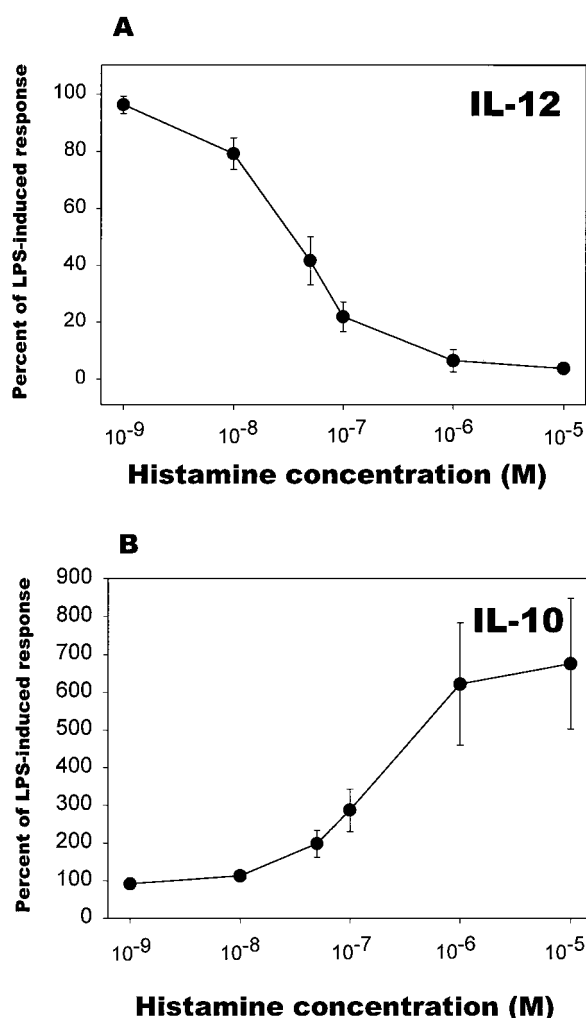


FIGURE 1. Effect of histamine on LPS-induced IL-12 (A) and IL-10 (B) production in human whole blood from six normal volunteers. Increasing concentrations of histamine were added as indicated. Data are expressed as the mean \pm SE. Mean LPS-induced IL-12 (p70) production was 82.3 ± 14.1 ; IL-10 was 210.0 ± 80.1 pg/ml.

from total PBMC using magnetic beads coated with a primary mAb specific for CD14. A single step incubation and elimination was performed as above. The PBL preparations contained $\leq 0.01\%$ monocytes (3 of 4 had 0%), as determined by flow cytometry.

PBMC were cultured at 1×10^6 cells/ml, while PBL and monocytes were cultured at 5×10^5 cells/ml. All cultures were set up in RPMI 1640 medium supplemented with 15% FCS, 1% glutamine, gentamicin 50 $\mu\text{g}/\text{ml}$ at a final volume of 0.5 ml. PBMC were activated with SAC at a final dilution of 1:200, while PBL and monocytes were stimulated with LPS (1 $\mu\text{g}/\text{ml}$). All cultures were incubated in 5% CO_2 at 37°C for 18 h.

Cytokine assays

IL-12 (p70 and p40) and IL-10 were measured using ELISAs employing the multiple Ab sandwich principle (Quantikine, R&D Systems). These assays specifically detect human IL-12 p70 (the biologically active heterodimer), the 40-kDa (p40) subunit of IL-12 and human IL-10, respectively. IL-12 p70 ELISA recognizes specifically the IL-12 heterodimer without cross-reactivity with the individual subunits of the dimer (p35 and p40). The detection limits of the IL-12 p40, IL-12 p70, and the high sensitivity (HS) IL-12 p70 ELISAs were 15.0, 5.0, and 0.5 pg/ml, respectively, while for the IL-10 ELISA it was 2 pg/ml. Plates were read by a microplate reader (Model 550, Bio-Rad, Richmond, CA), and absorbency was transformed to cytokine concentration (pg/ml) using a standard curve computed by Microplate Manager III, Macintosh Data Analysis Software (Bio-Rad).

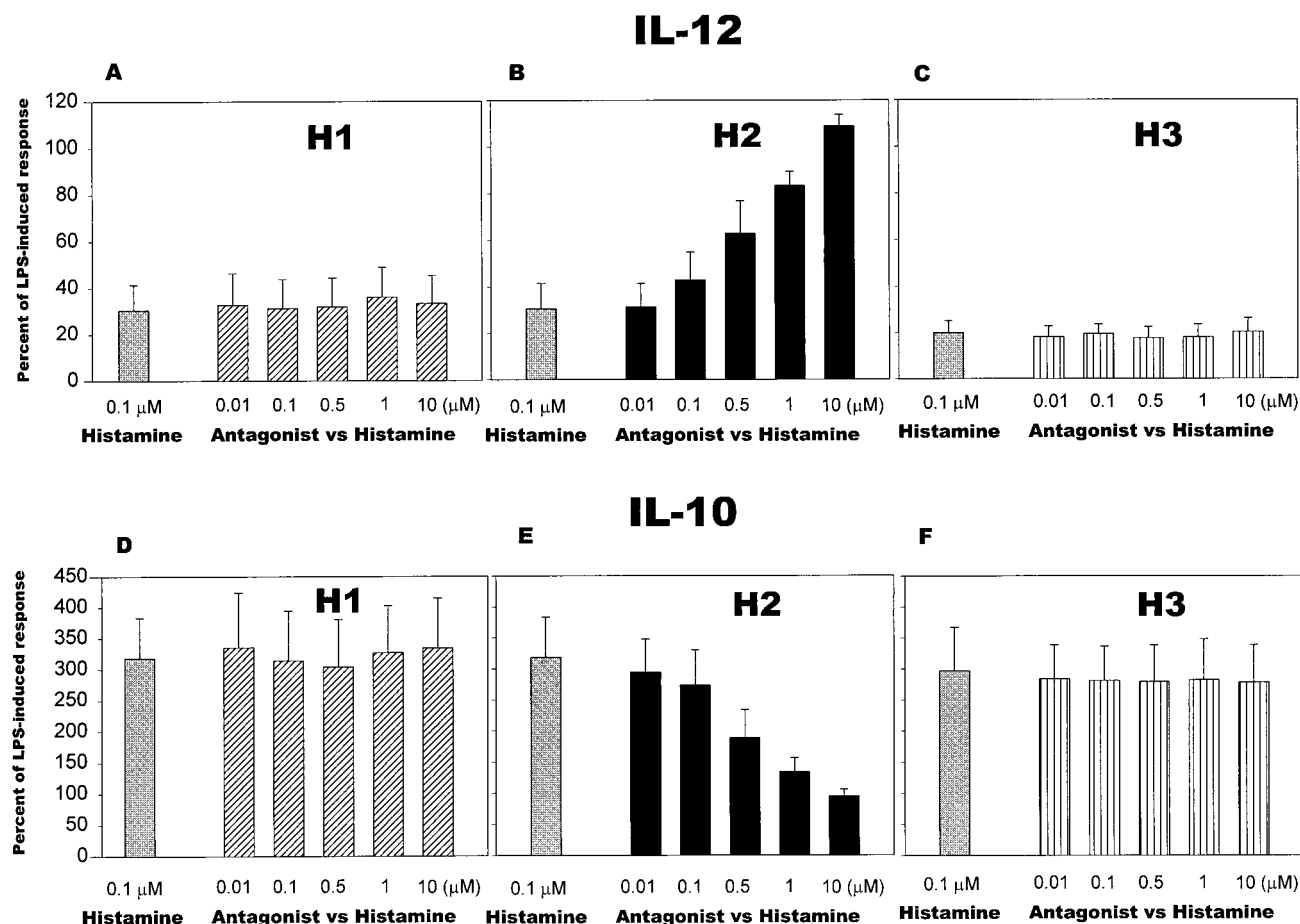


FIGURE 2. Cimetidine, an H2 receptor blocking agent, antagonizes the effect of histamine. Increasing concentrations of pyrilamine maleate, an H1 receptor antagonist (A and D); cimetidine (B and E); and thioperamine maleate, an H3 receptor antagonist (C and F), were added to whole blood samples as described. Data are expressed as the mean \pm SE of 10 normal volunteers (five for H1 and H2 receptor antagonists and five for H3 receptor antagonist). Mean LPS-induced IL-12 (p70) production was 60.7 ± 20.9 ; IL-10 was 143.0 ± 59.2 pg/ml (for H1 and H2 receptor antagonist experiments). IL-12 (p70) was 37.8 ± 12.7 , and IL-10 was 179.4 ± 76.2 pg/ml for H3 receptor antagonist experiments.

Results

Histamine suppresses LPS-induced IL-12 and potentiates IL-10 production

Addition of increasing concentrations of histamine to the whole blood culture, resulted in a dose-dependent inhibition of LPS-induced IL-12 p70 production (Fig. 1A). Histamine at 10^{-5} M concentration resulted in $>95\%$ inhibition of IL-12 production. Conversely, increasing concentrations of histamine caused a dose-dependent increase of LPS-induced IL-10 production (Fig. 1B), yielding a substantial increase of IL-10 production at 10^{-7} M (186% increase) and 10^{-6} M (521% increase).

The H2 receptor antagonist, cimetidine, completely blocks the effect of histamine on IL-12 and IL-10 production

The addition of increasing concentrations of the H1 receptor antagonist, pyrilamine maleate (mepyramine), and the H3 receptor antagonist, thioperamine maleate, failed to prevent the effect of histamine (0.1μ M) on both IL-12 and IL-10 secretion (Fig. 2, A, C, D, and F). However, both the inhibitory and the stimulatory effects of histamine (0.1μ M) on IL-12 and IL-10, respectively, were antagonized by increasing concentrations of cimetidine, an H2 receptor-blocking agent, leading to a complete blockade of the effect at 10μ M concentration of cimetidine (Fig. 2, B and E). These results suggest that the effects of histamine on these cytokines were mediated by the H2 receptor. Increasing doses of the

H2 antagonist cimetidine, identical to those used to block the effect of histamine, without added histamine did not affect LPS-induced IL-12 p70 or IL-10 production ($n = 4$; data not shown).

The H2 receptor agonist, dimaprit, mimics the effect of histamine

To demonstrate the involvement of H2 receptors in the histamine effect more definitively, we used dimaprit, an H2 receptor agonist in the subsequent experiments. As evident from Figure 3, A and B, the addition of this drug induced dose-dependent inhibition and potentiation of LPS-induced IL-12 and IL-10 production, respectively. Thus, the effect of this drug fully mimicked the effect of histamine on the production of these cytokines.

Histamine alone has no effect on basal IL-12 and IL-10 production

In subsequent experiments, we tested the possibility that histamine itself, without LPS, is able to induce or reduce IL-12 or IL-10 production. Addition of increasing doses of histamine (10^{-9} – 10^{-5} M), without LPS, failed to affect basal IL-12 production. The level of IL-12 at this concentration range of histamine was <0.5 pg/ml, similar to the negative control (no histamine), which was 0.5 ± 0.1 pg/ml ($n = 4$). Similarly, increasing doses of histamine (10^{-9} – 10^{-5} M), without LPS, failed to affect IL-10 production. The level of IL-10 at this concentration range of histamine was between 2.0

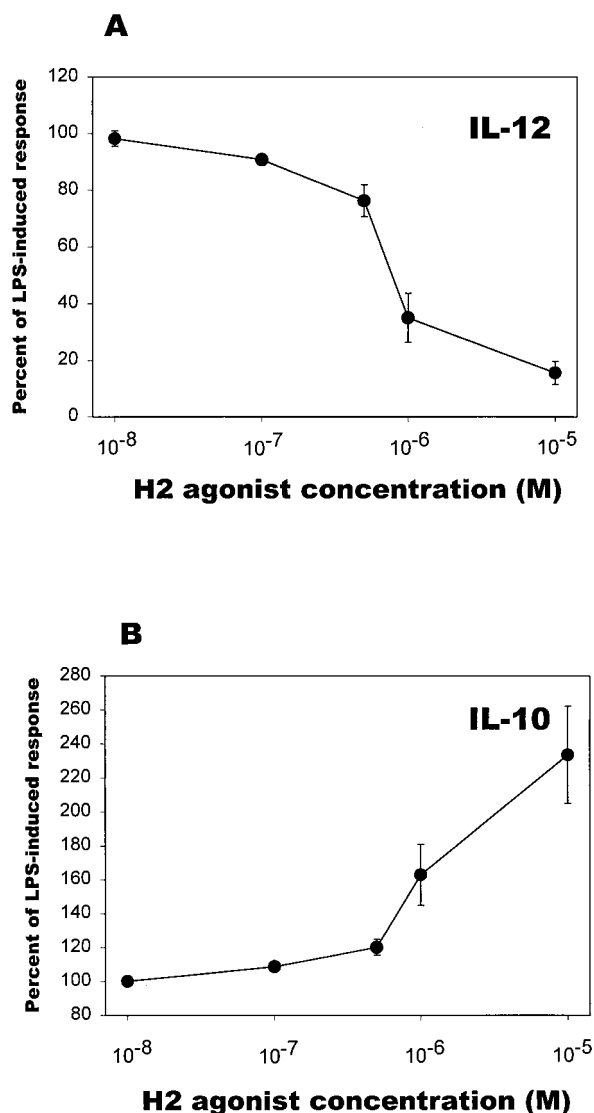


FIGURE 3. Effect of dimaprit, an H2 receptor agonist on LPS-induced IL-12 (A) and IL-10 (B) production in human whole blood from six normal volunteers. Increasing concentrations of dimaprit were added as indicated. Data are expressed as the mean \pm SE. Mean LPS-induced IL-12 (p70) production was 21.7 ± 3.8 ; IL-10 was 200.8 ± 49.2 pg/ml.

and 2.8 pg/ml, similar to the negative control (no histamine), which was 3.5 ± 1.1 pg/ml ($n = 4$).

The effect of histamine is independent of endogenous IL-10 production and is not affected by administration of exogenous IL-12

IL-10 has been described to inhibit the secretion of proinflammatory cytokines from monocytes/macrophages. This prompted us to investigate the possibility that the inhibitory effect of histamine on IL-12 production was caused by induction of endogenous IL-10 secretion. Conversely, since IL-12 inhibits IL-10 production, we decided to test whether administration of exogenous IL-12 might interfere with the effect of histamine on IL-10 production. For this purpose, we investigated the effect of adding neutralizing Abs to IL-10 or exogenous IL-12 to whole blood cultures stimulated with LPS in the presence of histamine. The concentration of anti-IL-10 Abs used in our experiments was 10 μ g/ml. This concentration, according to the manufacturer's instructions, gives a 50% neutralizing dose (ND₅₀) in the presence of 5 ng/ml of rhIL-10. The mean

LPS-induced IL-10 levels in our experiments were 300 ± 81.3 pg/ml (Fig. 4A). Thus, the concentrations of endogenously produced IL-10 were about 16-fold less than the concentration at which anti-IL-10 Abs express ND₅₀. Thus, we concluded that the concentration of neutralizing anti-IL-10 Abs used in these experiments was sufficient to eliminate endogenously produced IL-10. As shown in Figure 4A, the inhibition of IL-12 induced by histamine (0.1 μ M) could not be prevented by anti-IL-10 Abs. The LPS-induced levels of IL-12 p70 in our whole blood system usually ranged between 20 and 100 pg/ml. For this reason, we employed 50 and 200 pg/ml of exogenous IL-12 in our experiments. As shown in Figure 4B, the exogenously administered IL-12 did not reverse the histamine-induced enhancement of IL-10 production.

cAMP potentiates the effect of histamine

In the preceding experiments, we observed that the effect of histamine was mediated by H2 receptors. In many tissues, including immune cells, the stimulation of H2 receptors results in increased cAMP production. Phosphodiesterase is an enzyme involved in the breakdown of cAMP. Inhibition of this enzyme results in increased levels of cAMP. We investigated the effect of Ro 20-1724, a specific phosphodiesterase inhibitor. In our assay, Ro 20-1724 itself (1 μ M), without histamine, induced only a slight, nonsignificant inhibition or potentiation of LPS-induced IL-12 and IL-10 production. However, the addition of Ro 20-1724 to cultures containing histamine induced twofold potentiation of the inhibitory effect on IL-12 and the enhancing effect on IL-10 production (see Fig. 5A and B). These observations suggest the involvement of cAMP production in these immunoregulatory effects by histamine.

The effect of histamine on IL-12 and IL-10 production is not stimulus-specific

To prove that the effects of histamine are not limited to LPS stimulation, PBMC were stimulated with SAC, in the presence or absence of increasing concentrations of histamine. As shown in Figure 6A and B, histamine potently suppressed IL-12 (p70) production in a concentration-dependent manner, while it potentiated the production of IL-10. Similar to the whole blood assay, the effects of histamine on SAC-induced IL-12 and IL-10 production were blocked by the H2 receptor antagonist cimetidine, implicating once again the role of H2 receptors.

Monocytes are target cells for the effect of histamine on IL-12 and IL-10 production

As shown in Figure 7A and C, PBL, free of monocytes, produced negligible levels of IL-12 (p40) and no detectable levels of IL-10 in response to LPS. While PBMC produced significantly less IL-12 and IL-10 than enriched monocytes, coculture of PBMC with monocytes resulted in intermediate production of these two cytokines. These results indicate that monocytes are the primary source of IL-12 and IL-10 in the LPS-stimulated whole blood assay. This is in accordance with previous results of others (20). To verify that peripheral blood monocytes were directly affected by histamine, enriched human monocytes were stimulated with LPS in the presence or absence of increasing concentrations of histamine. As shown in Figure 7, B and D, histamine, as in the case of LPS-stimulated whole blood or SAC-stimulated PBMC, substantially inhibited LPS-induced IL-12 (p40) production in a concentration-dependent manner, while it potentiated the production of IL-10 from human monocytes. Similar to the findings with whole blood and PBMC, the effect of histamine on LPS-induced IL-12 and IL-10 production from monocytes was blocked by the H2 receptor

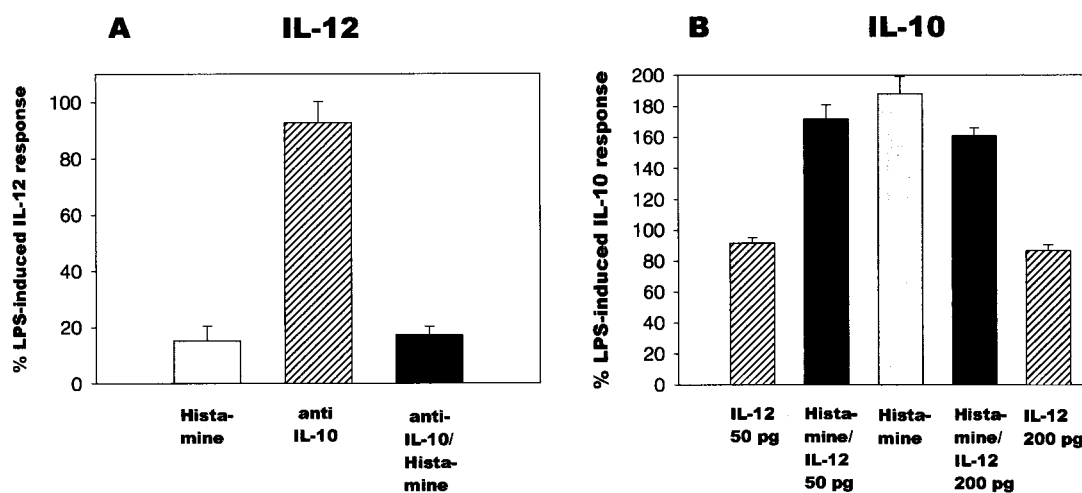


FIGURE 4. Effect of neutralizing anti-IL-10 Ab on IL-12 secretion (A) and exogenous recombinant human IL-12 on IL-10 secretion (B) in the presence and absence of histamine were determined in LPS-stimulated whole blood from six and five normal volunteers, respectively. Data are expressed as the mean \pm SE. Mean LPS-induced IL-12 (p70) production was 24.8 ± 8.4 (Fig. 4A); IL-10 was 250.1 ± 55.8 pg/ml (Fig. 4B).

antagonist cimetidine, thus implicating the critical role of H2 receptors on monocytes as mediating the effect of histamine in human peripheral whole blood or PBMC. Since isolated, unprimed blood monocytes produce little IL-12 p70 when stimulated with LPS or SAC, the effect of histamine on p70 could not be reliably evaluated in these experiments.

Discussion

Since IL-12 and IL-10 play important and contrasting roles in regulating immune responses, there is much interest in the factors that control their production. Focus has been on the regulation exerted by cytokines from within the immune system. Thus, IL-12 production is suppressed by IL-10 and IL-4, while IL-10 production is inhibited by IL-12 and IFN- γ . Recent evidence, however, indicates that factors other than cytokines have important regulatory influences on IL-12 and IL-10 production. These factors include the inflammatory mediators PGE₂ (20) and nitric oxide (21), and the two major stress hormones, glucocorticoids and catecholamines (22).

Here we report that histamine mediates dose-dependent inhibition of human bioactive IL-12 and stimulation of IL-10 production in whole blood cultures stimulated with LPS. Cimetidine, an H2 receptor antagonist, blocked the effects of histamine on IL-12 and IL-10 production, while H1 and H3 receptor antagonists failed to alter the effects of histamine. These data suggest that histamine is modulating the production of these two cytokines via the H2 receptor. This conclusion is further substantiated by the finding that dimaprit, an H2 receptor agonist, mimicked dose-dependently the effect of histamine. In agreement with previous studies (20), we provide evidence that monocytes are the main IL-12- and IL-10-producing cells in LPS-stimulated human peripheral blood. Human monocytes and monocytic lines express H2 receptors, and stimulation of these receptors by histamine or dimaprit results in increased cAMP formation (23). In addition, we demonstrated that, in monocytes, the effects of histamine on IL-12 and IL-10 production were similar to those in whole blood and also were blocked by the H2 receptor antagonist cimetidine, while Ro 20-1724, a

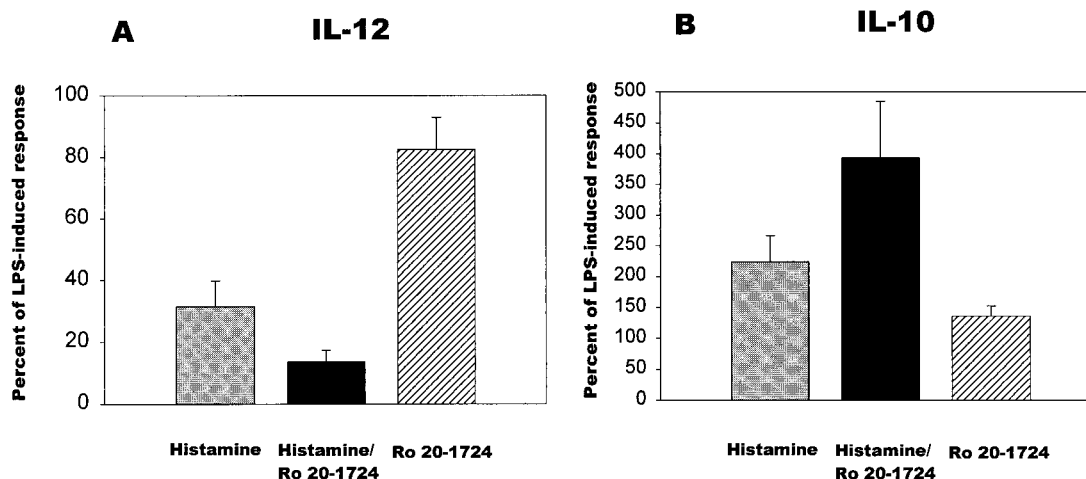
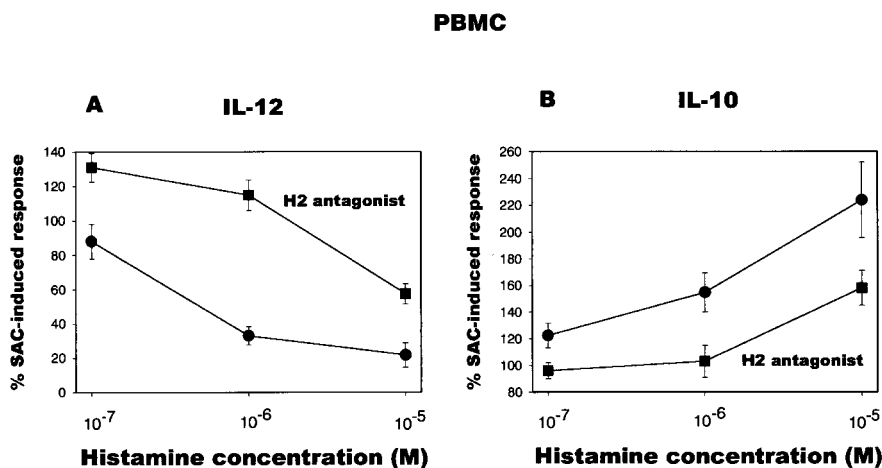


FIGURE 5. Effect of Ro 20-1724 on LPS-induced IL-12 (A) and IL-10 (B) secretion in whole blood from six normal volunteers. Ro 20-1724, histamine, and histamine plus Ro 20-1724 were added as described. Mean LPS-induced IL-12 (p70) production was 38.9 ± 7.1 ; IL-10 was 258.0 ± 45.1 pg/ml.

FIGURE 6. Effect of histamine on SAC-induced IL-12 (A) and IL-10 (B) production in human PBMC from four normal volunteers. Increasing concentrations of histamine were added as indicated. Data are expressed as the mean \pm SE. Mean LPS-induced IL-12 (p70) production was 23.3 ± 6.7 ; IL-10 was 182.4 ± 90.2 pg/ml.



phosphodiesterase inhibitor, potentiated the effects of histamine on both IL-12 and IL-10 secretion. Thus, H2 receptors on monocytes appear to mediate the effects of histamine on IL-12

and IL-10 cytokine production in peripheral blood via an increase of intracellular cAMP. This is in accordance with recent data showing that increased cAMP induced by other mediators,

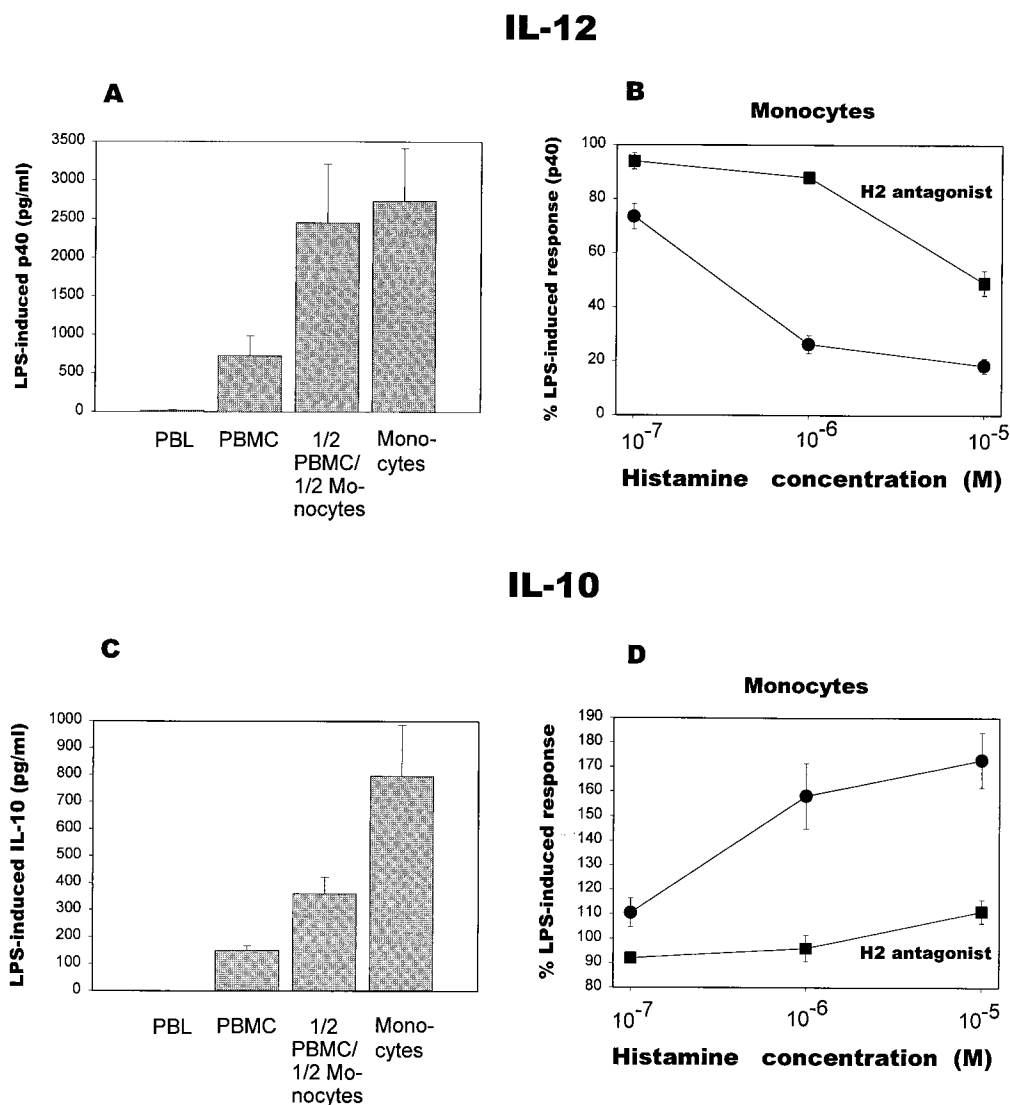


FIGURE 7. LPS-induced IL-12 p40 (A) and IL-10 (B) production from different cell compartments of human peripheral blood. Effect of histamine on LPS-induced IL-12 p40 (C) and IL-10 (D) production in human monocytes. Mean LPS-induced IL-12 p40 production was 2736.0 ± 680.2 ; IL-10 was 795.6 ± 189.7 pg/ml. Data (A-D) from four experiments are expressed as the mean \pm SE.

such as PGE₂ and catecholamines, have similar differential effects on IL-12 and IL-10 production (20, 22).

Monocytes/macrophages and other phagocytic cells are components of innate or natural immune mechanisms, while lymphocytes are components of acquired (adaptive) or specific immune mechanisms. Innate and specific immunity, however, are not autonomous. Cells of the innate immune system can determine which Ags the acquired immune system responds to and the nature of that response (24). This can be mediated through cytokines released by monocytes/macrophages that play a critical role in influencing the Th1/Th2 pattern that in turn dictates the type of immune response generated. Thus, IL-12 is a central inducer of Th1 differentiation and serves as a bridge between the innate and specific immunity (25), whereas IL-10 antagonizes the activities of IL-12. The Th1/Th2 pattern is often regarded as a balance between Th1/Th2 cell cytokine activities, but our observations suggest that conditions related to increased local or systemic levels of histamine may also contribute to Th1/Th2 balance. In particular, Th2-associated humoral immunity is potentiated. Our data are consistent with previous studies showing that histamine via H2 receptors inhibits TNF- α production from monocytes and mast cells (26–27), while it potentiates IL-6 production from endothelial cells (28). This view is further substantiated by a recent study demonstrating that histamine, also via H2 receptors, inhibits IFN- γ production by Th1-like cells but has no effect on IL-4 production from Th2 clones (29). Moreover, histamine directly enhances the production of human IgE from B cells (30), while it inhibits T cell cytotoxicity. Thus, available data suggest that histamine induces a Th2 shift at the level of monocytes, at the level of Th cells, and directly on effector cells. Interestingly, in the 1980s Rocklin and coworkers published several studies showing that histamine induced suppressor-cell activity and suppressor factor that were dependent of the presence of monocytes (31). We can speculate that this factor might have been IL-10. In summary, our data, considered in the context of these other studies, strongly suggest that histamine, apart from exerting potent effector functions in inflammation and allergy (mainly via H1 receptors), may have important immunoregulatory functions via H2 receptors expressed on immune cells.

The pattern of modulation exerted by histamine on IL-12 and IL-10 production might also be relevant to understanding host resistance to various infectious agents, where the selection of Th1- vs Th2- responses plays an important role. For example, recent evidence indicates that Th1 responses are important in the defense against infection with *H. pylori* (15). Now it is clear that this infection is the most common cause of chronic gastritis that in some cases progresses to peptic ulcer disease. An increased local concentration of histamine, induced by inflammatory or stress-related mediators, may play a pathogenic role in these conditions. It is tempting to speculate that a Th2 shift, induced by local excessive histamine production, may represent an additional pathogenic factor that participates in the development or extension of *H. pylori* infection. Our results also raise the question as to whether H2 receptor antagonists, widely used to prevent histamine-induced acid secretion by preventing the effect of histamine on cytokine-producing resident gastric monocytes/macrophages, may also favorably alter the host response to *H. pylori* infection and prevent progression into peptic ulcer disease, by restoring local Th1 responses.

It was recently reported that treatment with cimetidine correlated with increased survival in patients with gastric and colorectal cancer (17, 18). Histamine has been suggested to be a receptor-dependent “growth factor” in some colon, gastric, breast, and mel-

anoma cell lines. Also, high concentrations of histamine have been measured within colorectal and breast cancer tissues, and large numbers of mast cells have been identified within tumor tissues (16). The above mentioned beneficial effect of cimetidine on survival from cancer remains poorly understood (18). Our results suggest that, by preventing the effect of histamine on IL-12 and IL-10 production, cimetidine may contribute to a restoration of Th1 responses and a better defense against these particular tumors.

Atopic reactions are characterized by dominant Th2 responses. Our results suggest that histamine might participate in a positive feedback loop, whereby allergen/Ag-IgE-induced release of histamine directly modulates IL-12 and IL-10 production that promotes and sustains a shift to IgE production.

In conclusion, we have demonstrated that histamine alters substantially the pattern of monocyte IL-12/IL-10 production and that this effect can be blocked by an H2 antagonist. Our results suggest that H2 receptor antagonists might be candidates for pharmacologic enhancement of Th1 functions in certain types of infections and tumors and for inhibiting a Th2 shift in certain allergic reactions.

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