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Immunology

Mouse splenic and bone marrow cell populations that express high-affinity Fc_ε receptors and produce interleukin 4 are highly enriched in basophils

(mast cell/histamine/immunoglobulin E)

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ABSTRACT **Splenic and bone marrow cells from normal mice, and from mice that have been polyclonally activated by injection of anti-IgD antibody, contain cells that produce interleukin 4 (IL-4) in response to crosslinkage of Fc_ε receptors ($Fc_\varepsilon R$) or $Fc_\gamma R$ or to ionomycin. Isolated $Fc_\varepsilon R^+$ cells have recently been shown to contain all of the IL-4-producing capacity of the nonlymphoid compartment of spleen and bone marrow. Here, purified $Fc_\varepsilon R^+$ cells are shown to be enriched in cells that contain histamine and express alcian blue-positive cytoplasmic granules. By electron microscopy, the vast majority of cytoplasmic granule-containing cells are basophils; they constitute ≈25% and ≈50%, respectively, of $Fc_\varepsilon R^+$ spleen and bone marrow cells from anti-IgD-injected mice. The $Fc_\varepsilon R^-$ populations contain cells that form colonies typical of mast cells. The $Fc_\varepsilon R^+$ populations also contain cells that, upon culture with IL-3, form colonies of alcian blue-positive cells, but (in contrast to colonies derived from $Fc_\varepsilon R^-$ populations) the colonies are small, and all the cells die within 2–3 weeks. The $Fc_\varepsilon R^+$ cells synthesize histamine during a 60-hr culture with IL-3, while the $Fc_\varepsilon R^-$ cells do not. These results indicate that IL-4-producing $Fc_\varepsilon R^+$ cells are highly enriched in basophils.**

Interleukin 4 (IL-4) and a series of other cytokines are secreted by transformed and factor-dependent mast cell lines; the latter cells do so in response to crosslinkage of high-affinity Fc_ε receptors ($Fc_\varepsilon R$) or to treatment with calcium ionophores (1–5). These observations led us to question whether normal cells of mast cell or basophil lineage had the capacity to produce lymphokines and whether such production might have physiologic significance. Cells in the non-B, non-T cell compartment of spleen suspensions and in bone marrow produced IL-4 in response to crosslinkage of high-affinity $Fc_\varepsilon R$ (6). This effect was strikingly augmented by the presence of IL-3 and, in the presence of IL-3, crosslinkage of $Fc_\gamma RII$ also led to IL-4 production (7). Marked increases in the IL-4-producing potential of splenic non-B, non-T cells and bone marrow cells occurred in mice that had been infected with the helminthic parasite *Nippostrongylus brasiliensis* or had been injected with the polyclonal stimulant anti-IgD antibody (8). We have demonstrated that a small population of splenic non-B, non-T cells (1–2%) and of bone marrow cells (0.5–5%) express high-affinity $Fc_\varepsilon R$ (R.A.S., M.P., F.D.F., J. F. Urban, Jr., S.A.B., and W.E.P., unpublished work). These $Fc_\varepsilon R^+$ cells had essentially all of the IL-4-producing capacity of the non-B, non-T cell population in response to stimulation by crosslinkage of $Fc_\varepsilon R$ or $Fc_\gamma RII$ or by the calcium ionophore ionomycin. Among $Fc_\varepsilon R^+$ cells from spleen and bone marrow of mice that had been polyclonally activated by injection of anti-IgD, one-fifth were capable of secreting IL-4 following $Fc_\varepsilon R$ cross-linkage.

Here, we show that splenic and bone marrow cells bearing high-affinity $Fc_\varepsilon R$ are enriched in alcian blue-positive and histamine-containing cells. By electron microscopy, the vast majority of granulated cells in these preparations are basophils. These results raise the possibility that basophils are a source of IL-4.

MATERIALS AND METHODS

Animals, Immunization, and Cytokines. Virus-free BALB/c female mice, ages 8–12 weeks, were obtained from the Division of Cancer Therapy Animal Program, National Cancer Institute. Cells were obtained either from normal mice or from mice that had received an intravenous injection of 800 μg of monoclonal anti-IgD, 6 days earlier (8). Synthetic IL-3 was a gift of Ian Clark-Lewis (The Biomedical Research Centre, Vancouver) (9).

Splenic Non-B, Non-T Cells. These were prepared from spleen cell suspensions as described (6).

Cell Sorting. Bone marrow cells and splenic non-B, non-T cells ("unstained, unsorted" cells) were incubated sequentially with IgE, biotinylated anti-IgE (10), and streptavidin–phycoerythrin, to yield "stained, unsorted cells." These cells were subjected to fluorescence-activated cell sorting with an Epics 753 cell sorter (Coulter). Two populations were obtained by sorting. Cells that had clearly greater fluorescence than control (stained with streptavidin–phycoerythrin only) were designated $Fc_\varepsilon R^+$. A population of the phycoerythrin-negative cells (usually the 50% of the cells with the least phycoerythrin fluorescence) were called $Fc_\varepsilon R^-$.

Histamine Determinations. Histamine was measured by an enzymatic assay involving the conversion of histamine to [^{3}H]methylhistamine in the presence of *S*-adenosyl-L-[*methyl*-^{3}H]methionine and purified histamine *N*-methyltransferase (11, 12). Histamine content was assessed by boiling the cells for 45 min, then cooling on ice, centrifuging to remove cell debris, and measuring the content of the supernatant. Histamine production (13, 14) was assessed by

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Abbreviations: $Fc_\varepsilon R$ and $Fc_\gamma R$, receptor(s) for Fc portion of IgE and IgG, respectively; IL-3 and IL-4, interleukins 3 and 4.

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culturing the cells in serum-free RPMI 1640 containing synthetic IL-3 (250 ng/ml) for 60 hr, then centrifuging and measuring the histamine in the supernatant.

Alcian Blue Staining. Cells were centrifuged onto glass slides (cytocentrifuged), lightly heat-fixed with an alcohol lamp, stained for 45 min with 0.5% alcian blue at pH 1, and then counterstained for 15 min with 0.1% safranin in 1% acetic acid.

Electron Microscopy. Specimens were fixed and processed for transmission electron microscopic examination as previously described in detail (15–18). Thick (1 μm) sections were scanned after alkaline Giemsa staining by light microscopy at ×1000 to estimate the number of cells in these preparations that were basophils and/or mast cells. Thin sections of the same specimens then were examined at ×3000 in a Philips 400 transmission electron microscope. Cells exhibiting cytoplasmic granules and/or vacuoles were then photographed and printed at ×18,000. The photographed cells were identified by published criteria (17, 19) as basophils, basophilic myelocytes, eosinophils, eosinophilic myelocytes, neutrophils, neutrophilic myelocytes, monocytes/macrophages, plasma cells, "possible mast cell precursors," or "mononuclear cells (identity indeterminate)." "Possible mast cell precursors" were mononuclear cells with abundant cytoplasm that contained occasional cytoplasmic vacuoles. While such cells could not be identified with certainty as in the mast cell lineage, cells with similar ultrastructural features can be identified in primary cultures of bone marrow-derived, growth factor-dependent mouse mast cells or in cloned populations of growth factor-dependent or -independent mouse mast cells (ref. 20 and A.M.D. and S.J.G., unpublished data).

Mast Cell or Basophil Precursor Frequency. Cells were cultured for 10–14 days in 12–24 replicate wells, each with 200 μl of RPMI 1640 containing 10% fetal bovine serum and 20–25% WEHI-3 cell culture supernatant (21, 22). Each well was scored for the presence or absence of colonies that had the morphology of mast cell colonies (described in ref. 21). An estimate of their frequency was obtained by plotting ln(negative wells/total wells) against cell number and calculating the value where ln(negative wells/total wells) = −1. In other experiments, cells were plated in quadruplicate in 35 × 10-mm Nunc plates in 1 ml of alpha minimum essential medium containing 1.32% methylcellulose (1500 cP; 1 P = 0.1 Pa·sec), 10% supernatant of pokeweed mitogen (PWM)-stimulated spleen cells, 30% fetal bovine serum, 1% bovine serum albumin, 1 unit of recombinant human erythropoietin, 0.1 mM 2-mercaptoethanol, and penicillin/streptomycin (23). These cells were cultured for 7–10 days before total colonies and those with the morphology of mast cell colonies were counted. Cells from selected wells or colonies were stained to confirm that they contained alcian blue-positive granules. We call these colonies mast cell or basophil colonies because it is difficult to distinguish these two cell types by light microscopic examination of cytocentrifuged preparations.

RESULTS

Morphologic Characterization of $Fc_\varepsilon R^+$ Cells. To characterize $Fc_\varepsilon R^+$ cells and to compare them with cell populations lacking high-affinity $Fc_\varepsilon R$, we purified the cell populations and tested them for properties of cells of either the mast cell or the basophil lineage. Unsorted non-B, non-T cells from the spleens of normal and anti-IgD-injected mice had a frequency of 2–3% alcian blue-positive cells, whereas bone marrow of anti-IgD-injected mice contained ≈10% alcian blue-positive cells (Table 1). Since alcian blue staining at pH 1 identifies mast cell or basophil proteoglycans (24), and since expression of high-affinity $Fc_\varepsilon R$ is a property of cells of the mast cell and basophil lineages, the $Fc_\varepsilon R^+$ cells would be expected to be enriched in alcian blue-positive cells. $Fc_\varepsilon R^+$ cells were enriched 5- to 10-fold in alcian blue-positive cells, whereas the $Fc_\varepsilon R^-$ population was slightly depleted. The finding of alcian blue-positive cells in the $Fc_\varepsilon R^-$ population is consistent with the expression of granules but not $Fc_\varepsilon R$ in some immature mast cells and/or basophils (25). The alcian blue-positive cells from both the $Fc_\varepsilon R^+$ and $Fc_\varepsilon R^-$ populations of normal mice had only 1 or 2 granules per cell and those from anti-IgD-injected mice had 2–10 granules per cell [i.e., far fewer granules than those present in mature mast cells (19)]. Some of the alcian blue-positive cells had a polylobed nuclear morphology, suggesting that some or all of these cells were basophils.

Table 1. Alcian blue-positive cells among splenic non-B, non-T cells of normal or anti-IgD-injected mice or among bone marrow (BM) cells of anti-IgD-injected mice

Fraction	% alcian blue-positive cells		
	Normal spleen	Anti-IgD spleen*	Anti-IgD BM†
Unstained unsorted	2	2	10.3 ± 0.9
Stained unsorted	2	3	9.6 ± 0.9
$Fc_\varepsilon R^+$	10	32	50.1 ± 2.9
$Fc_\varepsilon R^-$	1	1	5.2 ± 0.9

*Average of two experiments.
†Mean ± SEM of four experiments.

Characterization of Cells by Electron Microscopy. Light microscopic examination of 1-μm sections of the specimens to be examined by electron microscopy indicated that ≈25% of the cells present in the $Fc_\varepsilon R^+$ populations of spleen cells derived from anti-IgD-injected mice contained alkaline Giemsa-stained granules, whereas the corresponding figure for $Fc_\varepsilon R^+$ cells derived from the bone marrow of such mice was ≈50%. These percentages are similar to those obtained by alcian blue staining of cytocentrifuged specimens (Table 1). By contrast, the $Fc_\varepsilon R^-$ populations from the spleen or bone marrow of anti-IgD-injected mice contained <5% cells with alkaline Giemsa-stained granules. By electron microscopy, we identified those cells which contained cytoplasmic granules and/or vacuoles. More than 90% of such cells in the $Fc_\varepsilon R^+$ populations could be identified as basophils or basophilic myelocytes. A typical basophil is shown in Fig. 1. Table 2 summarizes the morphologic characteristics of the granulated and/or vacuolated cells. Most splenic $Fc_\varepsilon R^+$ cells in the basophil lineage were mature basophils, whereas 31% of the cells in the basophil lineage in the $Fc_\varepsilon R^+$ bone marrow cells were basophilic myelocytes. All $Fc_\varepsilon R^+$ populations examined contained small numbers (3.5–4.1%) of cells with ultrastructural features suggesting that they might represent mast cell precursors. The $Fc_\varepsilon R^+$ populations also contained small numbers of cells in the eosinophil or neutrophil lineage. In addition to cells containing cytoplasmic granules and/or vacuoles, the $Fc_\varepsilon R^+$ populations also contained substantial numbers of mononuclear cells lacking ultrastructural features permitting their definite assignment to a particular lineage. Based on the overall cell frequencies obtained by light microscopy, these nongranulated, nonvacuolated mononuclear cells represented ≈75% and ≈50% of the cells from the spleens and bone marrow, respectively, of anti-IgD-immunized mice.

The $Fc_\varepsilon R^-$ populations contained predominantly neutrophils and eosinophils, with small numbers of eosinophilic myelocytes and monocytes/macrophages. Approximately 20–25% of the $Fc_\varepsilon R^-$ populations of spleen cells were mononuclear cells of uncertain lineage, whereas these cells were less frequent (<10% of the total cells) in $Fc_\varepsilon R^-$ cells derived from the bone marrow. No cells in the basophil lineage, and no possible mast cell precursors (as defined in refs. 17 and

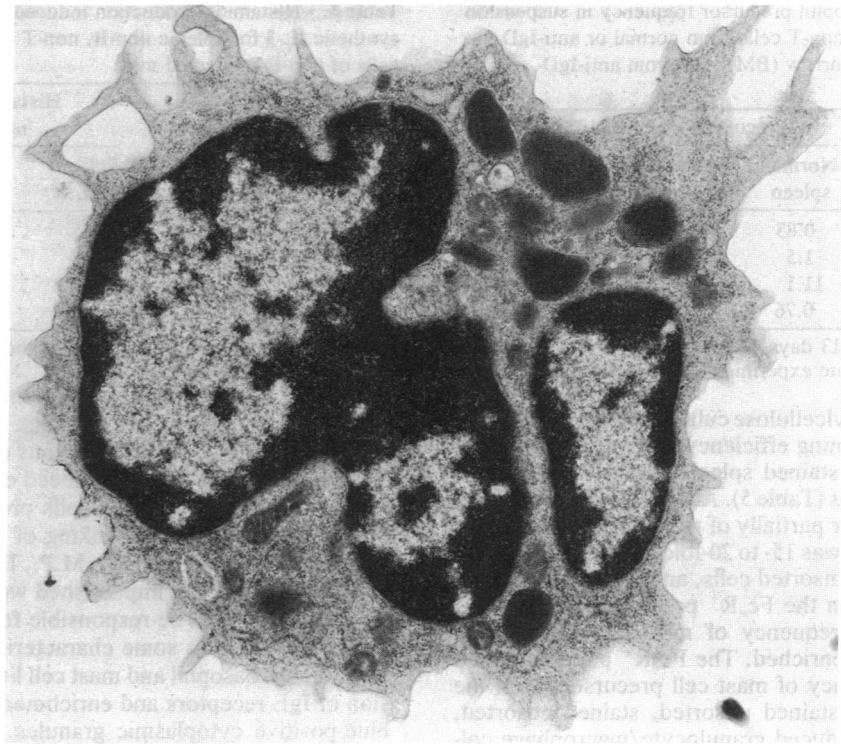

FIG. 1. Basophil in the $Fc_\varepsilon R^+$ cell population from spleens of anti-IgD-injected mice. This basophil has a polylobed nucleus with a condensed chromatin pattern, as well as irregular, broad surface processes and a small number of homogeneously electron-dense, membrane-bound cytoplasmic granules. (×17,600.)

19), were identified in one of the two $Fc_\varepsilon R^-$ spleen cell populations and in the $Fc_\varepsilon R^-$ bone marrow population. The other $Fc_\varepsilon R^-$ spleen cell population contained four basophils and a single possible mast cell precursor.

Histamine Content. The presence of histamine is a characteristic of mast cells and basophils, and histamine content per cell increases during mast cell maturation (20, 26). As the $Fc_\varepsilon R^+$ cells have only a few granules, their histamine content was expected to be low. Indeed, the content of $Fc_\varepsilon R^+$ cells from anti-IgD-injected mice was ~2 ng per 10^6 cells, 10-fold greater than that of the unseparated cells (Table 3), but 10- to 500-fold lower than in mouse mast cell lines or human basophils (20, 26–28).

Table 2. Identity of $Fc_\varepsilon R^+$ cells containing cytoplasmic granules and/or vacuoles and derived from the spleens or bone marrows of anti-IgD-injected mice

Cell type	No. of cells (%)		
	Spleen specimen 1	Spleen specimen 2	Bone marrow specimen 1
Basophil	63 (86.3)	25 (92.6)	108 (63.2)
Basophilic myelocyte	3 (4.1)	0 (0)	48 (28.1)
Eosinophil	1 (1.4)	0 (0)	1 (0.6)
Eosinophilic myelocyte	0 (0)	0 (0)	1 (0.6)
Neutrophil	3 (4.1)	1 (3.7)	5 (2.9)
Neutrophilic myelocyte	0 (0)	0 (0)	2 (1.2)
Possible mast cell percursor	3 (4.1)	1 (3.7)	6 (3.5)

Data are from three separate experiments. Only cells displaying cytoplasmic granules and/or vacuoles are listed. Nongranulated, nonvacuolated cells constituted ≈75% and ≈50%, respectively, of $Fc_\varepsilon R^+$ splenic non-B, non-T cells and $Fc_\varepsilon R^+$ bone marrow cells.

Mast Cell or Basophil Precursor Frequency. Compared with unsorted cells, the $Fc_\varepsilon R^+$ fraction of splenic non-B, non-T cells had a 4- to 10-fold higher capacity, in suspension cultures, to form colonies containing granulated alcian blue-positive cells (Table 4). The $Fc_\varepsilon R^+$ cells from bone marrow also were enriched in such colony-forming cells. The $Fc_\varepsilon R^-$ population was only slightly diminished in the frequency of cells forming alcian blue-positive colonies. Since the $Fc_\varepsilon R^-$ population represented ≥94% of the non-B, non-T cells or bone marrow cells from normal and from anti-IgD-injected mice (R.A.S., M.P., F.D.F., J. F. Urban, Jr., S.A.B., and W.E.P., unpublished work), the majority of precursors of colonies containing cells with alcian blue-positive granules, from either normal or anti-IgD-injected mice, lacked detectable $Fc_\varepsilon R$. Furthermore, following culture with IL-3, these $Fc_\varepsilon R^-$ precursors developed into fully granulated, $Fc_\varepsilon R^+$ cells, which continued to proliferate for at least 4 weeks and which produced IL-4 upon $Fc_\varepsilon R$ crosslinking (unpublished results). In contrast, the colonies derived from $Fc_\varepsilon R^+$ cells were small, grew poorly, and died in 2–3 weeks, and the cells were less heavily granulated than those derived from $Fc_\varepsilon R^-$ cells. Similar results were obtained in the presence of irradiated syngeneic spleen cells (22) (data not shown).

Table 3. Histamine content of splenic non-B, non-T and bone marrow cells of anti-IgD-injected mice

Fraction	Histamine, ng per 10^6 cells	
	Spleen	Bone marrow
Unstained unsorted	0.28	0.25
Stained unsorted	0.18	0.25
$Fc_\varepsilon R^+$	1.8	1.7
$Fc_\varepsilon R^-$	0.26	0.11

Data are from two separate experiments. The lower limits of detection of histamine content were 0.06 ng per 10^6 cells for unstained unsorted, stained unsorted, and $Fc_\varepsilon R^-$ cells and 0.6 ng per 10^6 cells for $Fc_\varepsilon R^+$ cells.

Table 4. Mast cell or basophil precursor frequency in suspension cultures of splenic non-B, non-T cells from normal or anti-IgD-injected mice or of bone marrow (BM) cells from anti-IgD-injected mice

Fraction	Precursors, no. per 10^4 cells		
	Normal spleen	Anti-IgD spleen	Anti-IgD BM
Unstained unsorted	0.83	1.1	ND
Stained unsorted	1.5	2.4	2.0
$Fc_\epsilon R^+$	11.1	9.1	6.7
$Fc_\epsilon R^-$	0.76	1.0	2.0

Cells were cultured for 13 days with 25% WEHI-3 supernatant. Data are from three separate experiments. ND, not determined.

We also utilized methylcellulose cultures, under conditions that yielded greater cloning efficiency than suspension cultures. Of unstained or stained splenic non-B, non-T cells, 0.25–1% formed colonies (Table 5). About 35% were colonies consisting exclusively or partially of mast cells or basophils. The number of colonies was 15- to 20-fold higher in the $Fc_\epsilon R^+$ population than in the unsorted cells, and >90% of the cells in colonies derived from the $Fc_\epsilon R^+$ population were alcian blue-positive, so the frequency of mast cell or basophil colonies was ≈50-fold enriched. The $Fc_\epsilon R^-$ population had a slightly lower frequency of mast cell precursors than the unsorted cells. The unstained unsorted, stained unsorted, and $Fc_\epsilon R^-$ cells all produced granulocyte/macrophage colonies, while the $Fc_\epsilon R^+$ cells did not. The bone marrow $Fc_\epsilon R^+$ cells did not produce granulocyte/macrophage colonies, but their degree of enrichment of "mast cell or basophil" colony-forming cells was not as great as for splenic $Fc_\epsilon R^+$ cells (data not shown).

Histamine Production. IL-3 is known to induce histidine decarboxylase, and thus rapid histamine synthesis and secretion, in populations of cells from mouse lymphoid organs (13, 14). These cells have been thought to be immature mast cells (22, 29, 30). Spleen and bone marrow cells produced large amounts of histamine (Table 6). Indeed, the levels of cell-associated histamine prior to culture with IL-3 were <1% of the levels of histamine produced in culture (compare Tables 3 and 6). Compared to stained unsorted cells, the $Fc_\epsilon R^+$ cells were 6- to 20-fold enriched in their capacity to synthesize histamine, whereas the $Fc_\epsilon R^-$ population lacked this capacity.

DISCUSSION

The existence of an $Fc_\epsilon R^+$ subset in cells derived from spleen or bone marrow was not anticipated, since no mature mast cells are identifiable in these preparations, and previous results (25, 31) have suggested that bone marrow cells, including immature mast cells, lack $Fc_\epsilon R$. Our results demonstrate that $Fc_\epsilon R^+$ subpopulations of splenic non-B, non-T cells and bone marrow cells indeed exist and are enriched in basophils. Since the $Fc_\epsilon R^+$ cells produce large amounts of IL-4 in response to crosslinking of $Fc_\epsilon RI$ or $Fc_\gamma RII$, or to calcium ionophores (R.A.S., M.P., F.D.F., J. F. Urban, Jr., S.A.B., and W.E.P., unpublished work), the results suggest that basophils may be responsible for IL-4 production.

Table 5. Hematopoietic colonies derived from methylcellulose cultures of splenic non-B, non-T cells of anti-IgD-injected mice

Fraction	No. of positive colonies per plate		
	100	1000	10,000
Unstained unsorted	0	10	40
Stained unsorted		5	25
$Fc_\epsilon R^+$	15	80	TNTC
$Fc_\epsilon R^-$		3	18

Hematopoietic colonies were counted after 8 days of culture with pokeweed mitogen-stimulated cell supernatant. Data are the means of quadruplicate plates seeded with 100, 1000, or 10,000 cells. The colonies consisted mainly of granulocyte/macrophage and mast cell or basophil colonies. For unstained, stained, and $Fc_\epsilon R^-$ cells, about 30–40% of the colonies were mast cell or basophil colonies; for $Fc_\epsilon R^+$ cells, >90% were mast cell or basophil colonies. TNTC, too numerous to count.

Table 6. Histamine production induced by 60 hr of culture with synthetic IL-3 for splenic non-B, non-T cells and bone marrow cells of anti-IgD-injected mice

Fraction	Histamine production, ng per 10^6 cells		
	Spleen exp. 1	Spleen exp. 2	Bone marrow
Unstained unsorted	105	17.7	81
Stained unsorted	38	10.2	39
$Fc_\epsilon R^+$	722	68	660
$Fc_\epsilon R^-$	3	3.6	2

Data are from three separate experiments.

$Fc_\epsilon R^+$ cells have some characteristics that are shared by cells of both basophil and mast cell lineage, including expression of IgE receptors and enrichment in cells bearing alcian blue-positive cytoplasmic granules. Also, these cells both store histamine and can be induced to synthesize and secrete histamine. Electron microscopy established that the vast majority of the granulated cells in the $Fc_\epsilon R^+$ population were basophils. The $Fc_\epsilon R^+$ cells formed colonies of alcian blue-positive cells, but these colonies were not typical of mast cells. In contrast to typical mast cell colonies derived from $Fc_\epsilon R^-$ cells, the colonies derived from $Fc_\epsilon R^+$ cells were smaller, died within 2–3 weeks, and were less heavily granulated. Thus the colonies derived from $Fc_\epsilon R^+$ cells may be basophil rather than mast cell colonies. Basophils are known to persist for a short time during culture of mouse bone marrow with IL-3-containing medium (17).

The frequency of IL-4-producing cells in the $Fc_\epsilon R^+$ population of spleen and bone marrow from anti-IgD-injected mice is ≈20% (R.A.S., M.P., F.D.F., J. F. Urban, Jr., S.A.B., and W.E.P., unpublished work). Since the $Fc_\epsilon R^+$ population is not homogeneous, the mature basophil, the basophil myelocyte, or the nongranulated cell(s) may account for IL-4 production. It is possible that some of the nongranulated, $Fc_\epsilon R^+$ cells are also in the basophil lineage, but the identity of the cells cannot be established by morphologic criteria. Similarly, the possibility that some of the nongranulated, $Fc_\epsilon R^+$ cells are in the mast cell lineage must be seriously considered.

Mouse basophils were described by light (32) and electron (17) microscopic criteria about 10 years ago. They are difficult to identify because, in contrast to human and guinea pig basophils, they contain few cytoplasmic granules (17, 19, 32). Nevertheless, the granules of mouse basophils are similar by ultrastructure to those of basophils of other species (17, 19). Several strains of mice, including the BALB/c mice used in these studies, are reported to have few or no circulating basophils under ordinary circumstances, but all strains developed 2–10% circulating basophils following immunization with heterologous serum (32). In our studies, the spleen and bone marrow of normal mice had small numbers of $Fc_\epsilon R^+$ cells, but $Fc_\epsilon R^+$ cells were markedly increased following injection of anti-IgD (R.A.S., M.P., F.D.F., J. F. Urban, Jr., S.A.B., and W.E.P., unpublished work), a stimulus known to induce IL-4 production *in vivo* and to increase serum IgE levels (8).

The $Fc_\varepsilon R^+$ cells contained not only all of the IL-4-producing capacity of the non-B, non-T cells, but also all of the histamine-synthesizing capacity. Histamine synthesis is characteristic principally of the mast cell or basophil lineages (33) and is almost certainly due to the induction of histidine decarboxylase activity (13, 14, 30). Some cells in the $Fc_\varepsilon R^-$ population responded to prolonged culture with IL-3 by developing into mast cells, which express IgE receptors and contain histamine. However, only the $Fc_\varepsilon R^+$ population underwent rapid IL-3-induced activation of histidine decarboxylase.

Human and guinea pig basophils are known to migrate into sites of inflammation, including responses to parasites, cutaneous basophil hypersensitivity reactions, contact dermatitis, and IgE-dependent late-phase inflammatory reactions (34–37). Only recently has convincing evidence been provided that rat and mouse basophils accumulate at cutaneous sites expressing immunologic responses (38–41). Human basophils are known to secrete histamine at late-phase inflammatory sites (42). Thus, mouse basophils would be excellent candidates to secrete IL-4 and other cytokines at inflammatory sites, and basophil-derived IL-4 could have a physiologically important role in IgE production and other immunologically mediated events.

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