Hepatocyte Growth Factor Is a Regulator of Monocyte-Macrophage Function

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Hepatocyte growth factor (HGF) is a paracrine factor produced by mesodermal tissues, capable of inducing an invasive behavior in neighboring epithelial and endothelial cells (1). This complex action results from the coordinate execution of the mitogenic and motogenic events that the factor can elicit in its target cells (for a review, see Ref. 2). HGF is mitogenic for epithelial and endothelial cells (3), and induces dissociation of epithelial sheets (4); it is able to increase motility and directional migration in dissociated cells (5, 6), to promote matrix invasion and degradation (7, 8), and to three-dimensionally organize invading cells in vitro (9). In vivo, HGF is a potent angiogenic factor (5, 10) and is involved in kidney and liver regeneration (11, 12).

HGF is secreted as a biologically inactive single-chain precursor of 92 kDa referred to as pro-HGF (13, 14), which binds the cell surface and/or the extracellular matrix via low affinity/high capacity binding sites formed by heparin-like glycosaminoglycans (8, 15). Pro-HGF also binds the MET receptor, but with low affinity and without triggering its kinase activity (13, 16). Extracellular proteolysis of pro-HGF generates a bioactive disulfide-linked heterodimer consisting of an α-chain of 60 kDa and a β-chain of 32 kDa (17, 18). The molecule shares structural similarities with factors of the blood clotting cascade, including four characteristic “kringle” domains within the α-chain, and a serine protease-like domain within the β-chain. However, HGF is devoid of any detectable proteolytic activity (19).

The HGF receptor is the transmembrane tyrosine kinase encoded by the MET protooncogene (8, 20). It is a 190-kDa heterodimer composed of a 50-kDa extracellular α-chain disulfide linked to a 145-kDa transmembrane β-chain. The latter consists of an extracellular domain involved in ligand binding, a transmembrane segment, and an intracellular domain containing the tyrosine kinase activity. Both the α and the β subunits derive from proteolysis of a single-chain precursor (21, 22).

The HGF receptor gene behaves as a delayed early gene. Its expression in epithelial cells is induced by treatment with phorbol esters, serum, and HGF itself (23). These observations are substantiated by the characterization of the MET promoter region, where responsive elements for phorbol-12-myristate-13-acetate (TPA) and serum are present (24).

As expected from the pleiotropic response that HGF elicits in its targets, intracellular signaling by the HGF receptor involves multiple pathways (25–27). HGF-induced receptor activation causes autophosphorylation of two tyrosines in the receptor tail, generating a nonconventional docking site capable of activating multiple cytoplasmic SH2-containing signal transducers (28).

Urokinase-type plasminogen activator (uPA) is an important pro-HGF convertase at the tissue level (13). Maturation of pro-HGF to the biologically active factor is a crucial limiting step in HGF signaling, and occurs through a tightly regulated proteolytic cleavage. Activation of pro-HGF by uPA has the uncommon feature of being a stoichiometric reaction (29); in other words, the amount of uPA is the limiting factor in the production of active HGF at the cell surface. This implies that pro-HGF activation can be locally tuned by modulating the amount of available uPA, and strongly supports the idea that HGF acts in a paracrine fashion.

An involvement of uPA in inflammation processes has been advocated, due to its role in tissue migration of mononuclear phagocytes, and to its ability of proteolytically activating or inactivating different proinflammatory cytokines (see Ref. 30, and references therein). We previously showed (29) that pro-HGF activation by uPA occurs in mononuclear cell cultures. We now report
that HGF is indeed active on monocytes, being able to induce migration and production of cytokines; moreover, monocyte activation enhances cell responsiveness to the factor by up-regulating the expression of the HGF receptor (the MET gene product) and the HGF convertase (uPA). Finally, we provide evidence for HGF production by activated monocytes, suggesting an autocrine stimulation of monocytes by HGF.

Materials and Methods

Human peripheral monocyte isolation

Mononuclear cells were obtained from healthy blood donors by Ficoll (Nycomed, Oslo, Norway) density gradient centrifugation. Mononuclear cells were washed three times in PBS and plated on petri dishes at a concentration of about 6 × 10^6 cells/cm^2 in endotoxin-free medium 199 (M199; Sigma, St. Louis, MO) containing 10% endotoxin-free bovine calf serum (HyClone, Logan, UT). After incubation for 1.5 h at 37°C, nonadherent cells were removed and monocyte monolayers were washed twice with PBS. For lymphokine-production assays, monocytes were cooled in PBS on ice for 30 min, carefully detached with a scraper and replated in 24-well plates at about 10^5 cells/cm^2.

Maintenance of endotoxin-free conditions

All tissue culture ware, if not of a sterile plastic disposable nature, was thoroughly washed and heated to 180°C for 3 h to destroy any contaminating endotoxin. Endotoxin-free M199 and bovine calf serum were obtained from Sigma and HyClone, respectively. All other reagents were prepared in pyrogen-free water.

Production and purification of human recombinant HGF

Full length HGF cDNA was cloned from human liver mRNA and expressed in Spodoptera frugiperda cells with the baculovirus expression system. Culture supernatant was collected and filtered, and recombinant pro-HGF was purified by affinity chromatography on a heparin-Sepharose fast protein liquid chromatography column, as described (13).

Flow cytometry analysis

Monocytes were detached and stained with polyclonal anti-HGF receptor antisera raised against extracellular epitopes of the MET sequence (31). Staining was carried on in PBS containing 2% BSA on ice for 30 min; cells were washed twice in PBS, then resuspended in the same buffer containing FITC-conjugated anti-rabbit Abs, according to the manufacturer’s recommendations (PharMingen, San Diego, CA). After washing three times in PBS, cells were analyzed on a FACScan flow cytometer (Becton Dickinson, Mountain View, CA).

MET probe and Northern blot analysis

Total cellular RNA was prepared from monolayer cultures, using the single-step method of extraction described by (32), and poly(A)^+ RNA was purified by oligo(dT)-coated magnetic beads according to the supplier directions (Promega, Madison, WI). For Northern blot analysis, mRNA was purified by oligo(dT)-coated magnetic beads according to the supplier method of extraction described by (32), and poly(A)^+ RNA was purified by oligo(dT)-coated magnetic beads according to the supplier directions (Promega, Madison, WI). For Northern blot analysis, mRNA was purified by oligo(dT)-coated magnetic beads according to the supplier directions (Promega, Madison, WI). For Northern blot analysis, mRNA was purified by oligo(dT)-coated magnetic beads according to the supplier directions (Promega, Madison, WI). For Northern blot analysis, mRNA was purified by oligo(dT)-coated magnetic beads according to the supplier directions (Promega, Madison, WI). For Northern blot analysis, mRNA was purified by oligo(dT)-coated magnetic beads according to the supplier directions (Promega, Madison, WI). For Northern blot analysis, mRNA was purified by oligo(dT)-coated magnetic beads according to the supplier directions (Promega, Madison, WI). For Northern blot analysis, mRNA was purified by oligo(dT)-coated magnetic beads according to the supplier directions (Promega, Madison, WI). For Northern blot analysis, mRNA was purified by oligo(dT)-coated magnetic beads according to the supplier directions (Promega, Madison, WI). For Northern blot analysis, mRNA was purified by oligo(dT)-coated magnetic beads according to the supplier directions (Promega, Madison, WI). For Northern blot analysis, mRNA was purified by oligo(dT)-coated magnetic beads according to the supplier directions (Promega, Madison, WI). For Northern blot analysis, mRNA was purified by oligo(dT)-coated magnetic beads according to the supplier directions (Promega, Madison, WI). For Northern blot analysis, mRNA was purified by oligo(dT)-coated magnetic beads according to the supplier directions (Promega, Madison, WI). For Northern blot analysis, mRNA was purified by oligo(dT)-coated magnetic beads according to the supplier directions (Promega, Madison, WI). For Northern blot analysis, mRNA was purified by oligo(dT)-coated magnetic beads according to the supplier directions (Promega, Madison, WI). For Northern blot analysis, mRNA was purified by oligo(dT)-coated magnetic beads according to the supplier directions (Promega, Madison, WI). For Northern blot analysis, mRNA was prepared in pyrogen-free water.

Western blotting

Cells were directly solubilized in boiling Laemmli buffer (33). Proteins, separated by SDS-PAGE, were transferred to nitrocellulose sheets and probed with Abs raised against a synthetic peptide corresponding to 19 C-terminal amino acids (from Ser^1372 to Ser^1390) of the MET sequence (34), as described elsewhere (21, 22, 35). Rabbit anti-mouse Ig conjugated to HRP and the enhanced chemiluminescence procedure (Amersham) were used.

Scatter assay

The presence of biologically active HGF in monocyte-conditioned media was assayed by measuring their scattering activity on Madin-Darby canine kidney (MDCK) cells, as described (4). Cells were seeded at low density on 96-well plates, in 2-fold serial dilutions of the samples. The scattering effect was monitored by light microscopy, and the titer was defined as the highest dilution capable of clearly dissociating MDCK cells.

Cytokine quantitation

The amount of IL-6, GM-CSF, and G-CSF in monocyte-conditioned media was determined by ELISA immunoassays (R&D Systems, Minneapolis, MN).

In vitro activation of HGF by uPA

Pro-HGF (1 μg) was radiolabeled with 1 mCi carrier-free NaI and 50 μg/ml IODO-GEN (Pierce, Rockford, IL) as described previously (8) to a specific activity of ~1.8 × 10^8 cpm/μg. Washed mononuclear cells were plated on 6-well plates at a concentration of about 6 × 10^5 cells/cm^2, and either immediately assayed for activation of pro-HGF, or incubated in endotoxin-free medium with 10% calf serum for 90 min at 37°C to allow adhesion, or activated by 100 ng/ml endotoxin for 6 h. Cells were then incubated in serum-free medium in the presence of known amounts of unlabeled pro-HGF and trace amounts of ^125I-labeled pro-HGF. After the indicated time, the culture supernatants and the cells were separately extracted with Laemmli buffer, and proteolytic cleavage of ^125I-labeled pro-HGF was monitored by reducing SDS-PAGE, autoradiography, and scanning of the autoradiogram.

Chemotaxis assay

Chemotaxis assays on human monocytes were performed with the Boyden chamber technique (36, 37). Polycarbonate filters (5 μm pore size; polyvinylpyrrolidone-free; Nucleopore, Pleasanton, CA) were coated with gelatin (Difco, 0.1% for 6 h at room temperature). HGF was seeded in the lower compartment of the chamber (50 μM/l in M199 containing 1% FCS), and 2 × 10^4 cells in M199 containing 1% FCS were then seeded in the upper compartment. After 8 h of incubation at 37°C, the upper surface of the filter was scraped with a rubber policeman. The filters were fixed and stained with Diff-Quick (Harleco, Philadelphia, PA), and 10 oil immersion fields were counted after coding samples. Platelet-activating factor (PAF; 1–0-octadecyl-2-acetyl-(R)-glycero-3-phospho-choline) and lyso-PAF (1–0-octadecyl-2-lyso-glycero-3-phosphocholine) were obtained from Bachem Feinkemikalien (Bubendorf, Switzerland).

Results

Expression of the HGF receptor in circulating and activated monocytes

We analyzed the expression of the HGF receptor in monocytes at different functional stages. Flow cytometry analysis on freshly isolated cells showed a low but detectable amount of the p190MET (Fig. 1a; Fig. 1c, isotype control). Adherent monocytes, cultured for 18 h in the absence of any activating agent, did not show a significant increase in the levels of HGF receptor expression (not shown), indicating that cell adhesion per se does not affect the HGF receptor expression level.

FIGURE 1. Flow cytometry analysis of human adherent (a) or endotoxin-activated (b) monocytes with anti-HGF receptor Abs (c, isotype control). Adherent monocytes express low but detectable levels of the HGF receptor, the expression of which is powerfully induced upon cell activation by endotoxin.
To investigate whether monocyte activation had any influence on the HGF receptor expression, we studied the HGF receptor mRNA and protein in monocytes treated with activating agents. Freshly prepared monocyte monolayers were incubated for 1–24 h in the presence of serum with or without 100 ng/ml endotoxin. Poly(A)^+ RNA was extracted from both nonactivated and activated monocytes, and analyzed in Northern blot using the full-size MET cDNA as a probe. The MET major transcripts were expressed at a barely detectable level in nonactivated monocytes, and increased 5- to 10-fold after an 18-h treatment with endotoxin (Fig. 2A).

Total cell extracts were probed in Western blot with mAbs directed against the HGF receptor. In lysates from endotoxin-activated monocytes, a band corresponding to the 145-kDa β-chain could be observed under reducing conditions, together with a 170-kDa band corresponding to the precursor (pr170^MET^; (21, 22). Truncated forms of the MET receptor could also be detected, as previously shown in other cell types (38). The receptor was detectable 4–6 h after addition of endotoxin to the culture and the induction was maximal after 12–24 h of stimulation (Fig. 2B).

The expression of the HGF receptor at the cell surface of endotoxin-activated monocytes was confirmed by flow cytometry analysis (Fig. 1B; Fig. 1C = isotype control). A pattern of induction similar to the one caused by endotoxin was seen upon stimulation with 10 ng/ml IL-1β, even though the overall level of expression was lower (data not shown).

Phorbol esters are known to induce the expression of the HGF receptor in epithelial cells (23) and are powerful monocyte activators. To study the effect of phorbol esters on the HGF receptor in monocytes, we used TPA as an activating agent. Lysates obtained from monocytes treated for 1–24 h with 10 μM TPA were probed in Western blot for the expression of the HGF receptor. Under reducing conditions, a single band was shown, comigrating with the 170-kDa receptor precursor. No mature β-chain could be detected, suggesting that TPA interferes with the HGF receptor maturation (Fig. 3A).

Phorbol esters are known to induce proteolysis of receptor tyrosine kinases (39). To investigate whether this was the case for the HGF receptor in TPA-activated monocytes, cells were treated with TPA for 1 h, extensively washed with PBS and extracted 1–24 h after TPA withdrawal. Under these conditions, the expression of both the precursor and the mature β-chain could be observed, with timing and relative amounts comparable to the endotoxin-treated monocytes (Fig. 3B). These data indicate that phorbol esters, differently from other more physiological activating agents, stimulate both the HGF receptor expression and its proteolytic degradation in monocytes.

Activated monocytes produce and release HGF in the culture medium

HGF production has been reported in alveolar macrophages and in HL-60 myeloid leukemia cells differentiated to a macrophage-like phenotype (40). To investigate whether primary circulating monocytes express HGF-specific transcripts, reverse transcriptase-PCR was performed on RNA preparations derived from adherent monocytes, using different couples of oligonucleotides specific for the HGF cDNA. HGF mRNA could be detected both in adherent and in endotoxin-activated monocytes (data not shown).

We studied the production of biologically active HGF by adherent and endotoxin-activated monocytes. Culture supernatants from nonactivated and endotoxin-activated monocytes were collected and probed for the ability to dissociate MDCK cell colonies (“scatter” assay). No scatter activity could be detected in media conditioned by nonactivated monocytes, while titers as high as 30 U/ml could be shown in media conditioned by endotoxin-treated monocytes (Table I).

Table I. HGF and IL-6 production by monocytes

<table>
<thead>
<tr>
<th>Stimulation</th>
<th>Control</th>
<th>HGF (100 U/ml)</th>
<th>LPS (100 ng/ml)</th>
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<tbody>
<tr>
<td>HGF (U/ml)</td>
<td>ND</td>
<td>3 ± 1</td>
<td>210 ± 17</td>
</tr>
<tr>
<td>IL-6 (pg/ml)</td>
<td>3 ± 1</td>
<td>210 ± 17</td>
<td>&gt;300</td>
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*Biologically active HGF was not detectable in culture media conditioned by nonstimulated monocytes, but was induced by treating the cells with endotoxin. Monocyte stimulation by 100 U/ml HGF resulted in the secretion of about 200 pg/ml immunoreactive IL-6. ND, Not detectable. Control, Medium conditioned by nonstimulated monocytes and supplemented with 100 ng/ml endotoxin after cell removal.
Activated monocytes efficiently process pro-HGF to the mature factor

We previously reported that inactive pro-HGF is physiologically converted to mature active HGF by specific proteolytic cleavage. uPA is a pro-HGF convertase (13, 29), whose expression is known to be induced in monocytes upon activation (41). As previously described (29), suspensions of freshly prepared mononuclear cells are unable to process 125I-labeled proHGF in serum-free conditions, while efficient proteolytic cleavage can be detected in adherent monocytes. This activity is most probably due to uPA. To investigate the possible increase of uPA-mediated pro-HGF activation in monocytes, we studied pro-HGF convertase activity in monocyte cultures treated with endotoxin. An enhancement of pro-HGF processing was observed in the conditioned medium of activated monocytes compared with adherent, nonactivated ones (Figs. 4A and 5A). To rule out a residual contamination from the serum used to induce monocyte adherence, all experiments were confirmed culturing cells in human heparinized plasma (data not shown).

We then determined the relative contribution of secreted vs cell-bound uPA in processing pro-HGF. When pro-HGF convertase activity of nonstimulated adherent monocytes was examined, efficient cleavage was shown in the conditioned medium; on the contrary, cell-association of cleaved HGF was strongly increased in the case of pretreatment of monocytes with endotoxin (Figs. 4B and 5B). This is most probably due to the concurrent increase of HGF receptor and uPA at the cell surface following monocyte activation.

HGF can trigger migration in PAF-primed monocytes

Monocytes are able to migrate in response to different chemotactic agents. HGF is a well-known motogenic factor, inducing motility and directional migration in epithelial and endothelial cells (1). The possible chemotactic activity of HGF on monocytes was investigated in Boyden chamber assays. Although freshly prepared monocytes did not show any detectable motogenic response to HGF, a clear effect could be measured when cells were pretreated with 10 nM (R)-PAF (Fig. 6). The effect was specific, because it was not triggered by lyso-PAF, a biologically inactive derivative of PAF (42). Whether this is due simply to the increased expression of the HGF receptor induced by PAF or to the acquisition of a more complex migration-competent phenotype following PAF priming, is not known.

HGF treatment up-regulates the level of GM-CSF, G-CSF and IL-6 transcripts in adherent monocytes

Monocytes play a key role in immune system intercellular signaling, being able to produce a variety of cytokines in response to appropriate activating stimuli. Cytokine production is crucial in...
monocyte function, and can be induced by activating agents such as endotoxin, IFN-γ and IL-1β (43). To investigate whether HGF is able to modulate monocyte secretory capabilities, we studied the effect of the factor on monocyte cytokine expression.

Freshly prepared monocyte monolayers were stimulated with highly purified recombinant HGF (50 U/ml), in the presence of serum. Poly(A)⁺ RNA preparations were obtained and probed in Northern blot using probes specific for various cytokines. Among the different cytokines analyzed, we could show an accumulation of IL-6, GM-CSF, and G-CSF transcripts. After 18 h of culture, in HGF-treated monocytes the levels of G-CSF and IL-6 transcripts were increased compared with controls, but considerably lower than in endotoxin-treated cells (Fig. 7); in the case of GM-CSF, on the contrary, HGF-stimulated cells yielded a higher amount of mRNA than endotoxin-treated ones (Fig. 7, middle panel). This is most probably due to a slower raise and a consequently delayed decrease in transcript level following HGF stimulation.

To verify whether secretion of functional cytokines followed the increase in mRNA levels, culture media conditioned by monocytes stimulated with highly purified HGF were tested with specific immunoassays. A detectable level of IL-6 could be shown by immunoenzymatic assays (Table I), while no G-CSF or GM-CSF production could be measured in HGF-stimulated monocytes. No other proinflammatory cytokine could be detected in the conditioned medium of HGF-treated monocytes.

We also investigated the possible role of HGF in the regulation of monocyte metabolic functions, by measuring its effect on oxidative burst. Cells were isolated from peripheral blood and resuspended in a luminol-containing buffer. Superoxide-dismutase activity was then monitored by a single photon counter. HGF stimulation did not significantly affect superoxide-dismutase activity (not shown).

Discussion

Monocytes play a central role in the immune system, being involved in both natural and acquired immunity. Although much is known about molecular events during monocyte activation, still a lot has to be learned about the mechanisms that regulate monocyte recruitment at different sites of the body and about their interactions with local tissue microenvironment. Monocytic cells display a wide phenotypical heterogeneity, ranging from circulating large mononuclear cells to tissue-resident macrophages at different stages of activation; however, at present, detailed information about the differential expression of stage-specific surface markers, adhesion molecules, and cytokine receptors is lacking.

We here show that HGF induces a complex and integrated biological response in monocytes, including up-regulation of the HGF receptor and pro-HGF convertase, secretion of HGF itself and other cytokines, as well as cell motility. The level of expression of the HGF receptor in nonactivated monocytes is barely detectable, and is greatly increased upon cell activation by endotoxin and IL-1β. Our findings are partially in disagreement with previous reports, where no HGF receptor expression could be detected in nonactivated monocytes (44, 45). Endotoxin-triggered intracellular signaling cascade includes hydrolysis of phosphoinositides, with subsequent generation of 1,2-diacylglycerol (DAG) and protein kinase C activation (46). Also IL-1β rapidly increases the level of intracellular DAG, although through a different kind of phospholipid hydrolysis (47).

TPA, another well known monocyte activator whose structure mimicks DAG, is able to induce the turnover of the receptor, showing an increase in the protein level after a short stimulation, even though a long-term treatment causes an apparent inhibition of maturation of the precursor. This effect is most probably due to TPA-induced proteolysis of the mature receptor, as previously described for other receptors (39). The expression and modulation of the HGF receptor in monocytic cells is also consistent with the structure of the MET gene promoter region, which contains TPA-responsive elements and a GATA-1 motif (24). The HGF receptor gene has been reported to be induced by TPA, serum, and HGF itself in epithelial cells, behaving as a delayed early gene (23). Our data do not rule out the possibility that the HGF receptor up-regulation could be at least in part indirect, mediated by autocrine secretion of cytokines by activated monocytes (e.g., IL-1β). Surprisingly enough, it must be noted that HGF stimulation of monocytes does not induce a detectable increase in the receptor level (not shown).

Monocyte activation can be defined as the acquisition of competence to execute complex functions, involving migration, endocytosis, Ag presentation, production of O₂⁻, and secretion of cytokines. It responds to the need of starting monocyte phagocytic and immunomodulating functions, and governing further responsiveness to exogenous signals. Our data show that monocyte activated phenotype includes the expression of the HGF receptor and of pro-HGF convertase. In addition, activated monocytes produce detectable amounts of biologically active HGF, which is, therefore, likely to act in an autocrine/paracrine fashion on monocytic cells. It must be noted that many important factors involved in the regulation of monocyte development and functions (e.g., IFNs, PGs leukotrienes, etc.; for a review, see Ref. 43) are secreted by monocytes themselves, and, therefore, act via negative and positive feedback loops.

In vitro, HGF stimulation induces IL-6 production in monocytes, and, therefore, is involved in the control of monocyte immunomodulatory functions. G-CSF and GM-CSF transcript levels are also increased, but the protein product is not detectable in the culture medium after stimulation by HGF alone.

The motogenic activity of HGF on responsive cells has been well documented (4). However, the factor is not able to induce migration of circulating monocytes, unless they are pretreated with PAF, indicating that HGF alone is not sufficient to trigger monocyte migration, but requires a costimulation by a primer factor.

![FIGURE 7](http://www.jimmunol.org/)
Biologically active HGF is produced at the cell surface by uPA-mediated proteolytic cleavage of pro-HGF. We previously showed (29) that pro-HGF activation is a tightly controlled reaction, which occurs both in monocytic cell lines and in primary monocytes. Notably, only adherent monocytes can efficiently process pro-HGF, while nonadherent ones cannot. We now report that monocyte activation further increases pro-HGF convertase activity in monocytic cell cultures, and that under these conditions, processed HGF is found mainly associated to the cell surface, most probably due to the concurrent increase in the number of HGF binding sites at the cell surface, namely the HGF receptor and uPA molecules. The regulation of uPA-mediated pro-HGF activation is complex and likely to involve both up-regulation of uPA itself and down-regulation of its inhibitors (PAIs; for a review, see Ref. 30). As we described elsewhere (48), HGF is able to induce proliferation and differentiation of erythroid precursors in vitro, and its receptor is expressed by primordial hematopoietic cells in mouse embryo. This effect on hemopoietic progenitors is direct, because they do express the HGF receptor. The fact that monocyte/macrophage respond to HGF suggests that the effect of the factor on hemopoiesis may also be in part indirect, mediated by its modulation of mono- cyte secretory activity. It must be remarked, in addition, that the HGF receptor is expressed by endothelial cells, monocytes, and erythroid progenitors. These three populations share a common ontogenic origin. In fact, embryonic hematopoietic structures, blood islands, and vessel primordia, give rise to both blood cell progenitors and endothelial cells (49). Moreover, monocytes ap- pear late in ontogenesis, early hemopoiesis being only erythroid, and phagocytic functions in early stages are accomplished by endo- thelial cells. The fact that these functionally and ontogenetically related cell populations express the HGF receptor and respond to the factor indicates that HGF could be one of the factors controlling their reciprocal interactions.

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References


