CONNECTIVE TISSUE PROTEINS AND PHAGOCYTIC CELL FUNCTION

Laminin Enhances Complement and Fc-mediated Phagocytosis by Cultured Human Macrophages

By JOHN F. BOHNSACK, HYNDA K. KLEINMAN,* TSUNEO TAKAHASHI,‡ JOHN J. O'SHEA, AND ERIC J. BROWN

*From the Laboratory of Clinical Investigation, National Institute of Allergy and Infectious Diseases, and the *Laboratory of Developmental Biology and Anomalies, National Institute of Dental Research, National Institutes of Health, Bethesda, Maryland 20205; and the ‡Cryobiology Unit, American Red Cross, Bethesda, Maryland 20014*

Ingestion of opsonized foreign particles by phagocytic cells is a critical event in host defense against invading microorganisms, and factors that influence the rate and extent of phagocytosis are of particular interest. Fibronectin, a molecule found both in plasma and as a component of extracellular matrix, binds to phagocytic cells and enhances opsonin-mediated phagocytosis by monocytes, macrophages, and activated polymorphonuclear leukocytes (1–3). This phagocytosis enhancement by fibronectin is a direct effect of the molecule on the phagocyte and occurs for both IgG- and complement-opsonized targets. Because of this, we have suggested that the phagocyte-fibronectin interaction may be important for optimal phagocytosis in areas of inflammation (4). This hypothesis raises the possibility that interaction with other extracellular matrix components may also modulate phagocyte function. In this report, we examine the effect of the matrix glycoprotein, laminin, on the phagocytic efficiency of human monocyte-derived macrophages.

Laminin, a connective tissue glycoprotein with a molecular weight of 1,000,000, was originally isolated from a murine tumor (5). Laminin has subsequently been found to be a major component of all mammalian basement membranes (6). In vitro, laminin binds to other basement membrane components, notably type IV collagen and basement membrane proteoglycan (7), and mediates epithelial cell adhesion to artificial substrates (8). Unlike fibronectin, which also binds to collagen, proteoglycans, and cells (9), laminin is not normally detectable in the circulation. Recently (10), a laminin receptor on macrophages has been described, but very little is known about the effects of the interaction of laminin with phagocytic cells. We have tested whether macrophage-laminin interactions might, like interaction with fibronectin, modulate macrophage phagocytic function. In this work, we demonstrate that highly purified laminin enhances phagocytosis of IgG- or complement-opsonized erythrocytes by human

Address reprint requests to J. F. Bohnsack, Laboratory of Clinical Investigation, National Institute of Allergy and Infectious Diseases, National Institutes of Health, Building 10, Room 11N-214, Bethesda, MD 20205.

culture–derived macrophages. Macrophages exposed briefly to solid phase laminin increased their phagocytosis of both complement-coated erythrocytes and erythrocytes coated with suboptimal quantities of IgG. This occurred because of a direct effect of laminin on the macrophage, without any association of laminin with the opsonized erythrocyte. This phagocytosis-enhancing effect of laminin may be mediated via CR1, CR3, or the IgG Fc receptor. The results of the laminin-macrophage interaction are analogous to the previously described (1–4) effects of fibronectin on human monocytes, polymorphonuclear leukocytes, and macrophages, but are not due to contamination of laminin by fibronectin. Thus, an interaction of laminin with human macrophages, most likely through a receptor-mediated mechanism, markedly enhanced the phagocytic function of these cells. It is likely that multiple interactions with the extracellular matrix profoundly affect macrophage function in the extravascular compartment and particularly in areas of inflammation.

Materials and Methods

Buffers and Media. Veronal-buffered saline (VBS),[1] VBS to which 0.1% gelatin was added (GVBS), 0.060 μM isosmotic dextrose GVBS containing 1.0 mM Mg^{++} and 0.15 mM Ca^{++} ($DGVBS^{++}$), VBS containing 10 mM EDTA (EDTA-GVBS), and phosphate-buffered saline at pH 7.4 (PBS) were prepared as described (11).

RPMI 1640 medium was purchased from Quality Biological (Gaithersburg, MD). L-glutamine and gentamicin were purchased from M. A. Bioproducts (Walkersville, MD). Hepes and fetal calf serum were purchased from Gibco Laboratories (Grand Island, NY).

Reagents. Laminin was prepared from murine EHS cell line tumor, as previously described (12), and was free from contaminating protein as determined by sodium dodecyl sulfate–polyacrylamide gel electrophoresis (SDS-PAGE) (12). Fibronectin (1) and C4 (13) were purified from EDTA-treated human plasma. C4 was labeled with tritium using sodium borohydride (14) to a specific activity of 2.3×10^6 cpm/μg. Rabbit IgG and IgM anti-Forssman antigen (11) and IgG antilaminin (12) were prepared as previously described. $F(ab')_2$ fragments of rabbit IgG were prepared by the method of Madsen and Rodkey (15), adjusted to an A_{280} of 8.0, and dialyzed into PBS, pH 7.4. $F(ab')_2$ were free of Fc fragments as judged by double immunodiffusion with commercial antiserum (Miles Laboratories, Elkhart, IN). The IgG fraction of mouse ascites containing IgG1 monoclonal antibody 3D9 to human CR1 was prepared as previously described (16). HG11, a mouse IgG1 antibody to human IgG prepared with the same SP2/0 fusion partner as 3D9, was used as a control.

Phagocytic Targets. Opsonized sheep erythrocytes (E) bearing IgG anti-Forssman antibody (EA_{IgG}) or C4b (EAC4b) were prepared as previously described (17). Briefly, for preparation of EAC4b, E were sensitized with IgM anti-Forssman (EA_{IgM}) and guinea pig C1 (Cordis Laboratories, Miami, FL) (EAC1). These EAC1 were then incubated with varying concentrations of C4 for 60 min at 37°C, then washed in EDTA-GVBS, and finally washed and suspended in $DGVBS^{++}$ (EAC4b). These cells bore C4b as their only opsonin; their binding to monocytes and macrophages was completely inhibited by monoclonal 3D9 antibody to CR1. Quantitation of C4b uptake onto E was made in some experiments by substituting an equally hemolytic concentration of ^{3}H-C4 for unlabeled C4 and determining cell-associated radioactivity. Nonspecific binding of ^{3}H-C4, assessed by uptake of radiolabel onto EA_{IgM} in the absence of C1, always represented <1% of C4

[1] *Abbreviations used in this paper:* E, opsonized sheep erythrocytes; EA_{IgG} and EA_{IgM}, sheep erythrocytes sensitized with IgG and IgM anti-Forssman antibody; EAC4b, sheep erythrocytes bearing C4b; $DGVBS^{++}$, isosmotic dextrose VBS with 1.0% gelatin and 1.0 mM Mg^{++} and 0.15 mM Ca^{++}; GVBS, VBS with 0.1% gelatin; PBS, phosphate-buffered saline; SDS-PAGE, sodium dodecyl sulfate–polyacrylamide gel electrophoresis; VBS, veronal-buffered saline.

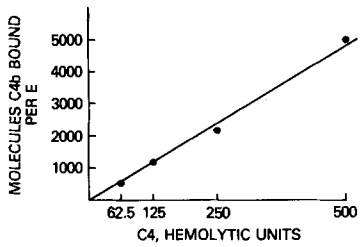

FIGURE 1. ^{3}H-C4 uptake on sheep E as a function of C4 hemolytic input.

input. Specific C4b uptake onto E was linear as a function of C4 input at the doses used (Fig. 1).

Monocyte Isolation and Culture. Human monocytes were isolated from buffy coats with the use of countercurrent elutriation, by a modification of the method of Lionetti et al. (18), as previously described (1). Monocytes were cultured by a modification of the method of Van der Meer et al. (19) in Teflon vessels (Savillex Corp., Minnetonka, MN) at 1×10^6/ml in RPMI 1640 containing 2 mM L-glutamine, 50 μg/ml gentamicin, and 15% heat-inactivated fetal calf serum in 5% CO_2 at 37°C. After 5 d in culture, these cells exhibited characteristics typical of macrophages, including loss of myeloperoxidase activity as demonstrated by histochemical staining, retention of esterase activity, increased phagocytosis of EA_{IgG}, increased protein content, and typical morphology after spreading on glass surfaces. After 6–8 d in culture, macrophages were harvested from the Teflon dishes by vigorous pipetting, washed twice in PBS, and resuspended in RPMI 1640 containing 10 mM Hepes and 50 μg/ml gentamicin, at 2.5×10^5 cells/ml. Preliminary experiments with freshly derived monocytes and monocytes removed from culture after different time intervals indicated that laminin's effect on phagocytosis was most pronounced after 5 d in culture.

Phagocytosis Assay. Eight-well Lab-Tek chambers (Dynatech Laboratories, Inc., Alexandria, VA) were coated with various concentrations of laminin or fibronectin in 0.1 M $NaHCO_3$, pH 9.5, for 2 h at room temperature, and then washed with PBS immediately before use. Macrophages (6.25×10^4 cells) were added in 250 μl and allowed to adhere for 1 h in a 5% CO_2 incubator at 37°C. 100 μl of opsonized E (EA_{IgG} or EAC4b at 1×10^8/ml) were then added, and the chambers were centrifuged at room temperature for 3 min at 50 *g*. The macrophages and phagocytic targets were then incubated without further agitation for 30 min at 37°C. To determine phagocytosis, 200 μl of medium was removed, and hypotonic lysis was performed by addition of 500 μl of PBS diluted 1:4 with distilled water. After completion of lysis, the cells were fixed for 5 min with 0.5% glutaraldehyde in PBS. After staining with Giemsa, phagocytosis was determined by light microscopy and expressed both as the number of E ingested per 100 macrophages (phagocytic index) and as the percentage of macrophages phagocytosing at least one E (percent phagocytosis). At least 200 macrophages were counted in each chamber.

In some experiments, 200 μl of a 1:10 dilution of $F(ab')_2$ in media, prepared from rabbit antilaminin [or preimmune $F(ab')_2$ as a control] were incubated with laminin-coated or uncoated wells for 30 min before the addition of macrophages and opsonized erythrocytes as described above. In experiments involving antibody to CR1, either 25 μl of a 10 μg/ml solution of 3D9 or 25 μl of a 1:100 dilution of HG11 ascites was added to the Lab-Tek chambers at the same time as the macrophages.

Assessment of Macrophage Adherence. Quantitation of the number of macrophages adherent to Lab-Tek chambers under the conditions of the phagocytic assay was performed using the Coulter Counter (Model ZBI; Coulter Electronics, Hialeah, FL). After adherence, phagocytosis, and lysis of extracellular E as described above, wells were treated with five drops of stromatolysing agent (Zap-O-Globin; Coulter Electronics). The contents of each well were aspirated by vigorous pipetting, and the released nuclei were quantitated in the Coulter Counter. Preliminary experiments demonstrated that the results from this

method were identical to those from a technique that quantitates the total DNA extracted from the adherent cells (20).

Results

Effect of Laminin on Phagocytosis of EAC4b. Shown in Fig. 2 is the dose-dependent response of solid phase laminin on macrophage phatocytosis of EAC4b prepared with varying amounts of purified human C4. In these experiments, macrophages were plated onto glass wells that had been coated with laminin for 2 h and from which excess fluid-phase laminin had been removed. As reported by others (3), a small but definite CR1-dependent phagocytosis of EAC4b occurred in culture-derived macrophages in the absence of laminin. Increasing amounts of C4 bound to E caused a small dose-dependent increase in macrophage phagocytosis of E. This contrasts with the activity of resting, freshly derived peripheral blood monocytes in which CR1 does not mediate phagocytosis (1). Exposure to laminin markedly increased the total number of E ingested (Fig. 2*A*) at all levels of C4b opsonization. Laminin also induced up to a fourfold increase in the number of macrophages ingesting E (Fig. 2*B*). The plateau of the dose response curves for the laminin effect on EAC4b phagocytosis was independent of the amount of C4b on the erythrocyte. The plateau occurred at about a 30 μg/ml input of laminin, whether phagocytosis was judged by the phagocytic index or by the percentage of macrophages ingesting E. Significant macrophage phagocytosis of E and EA_{IgM} did not occur in either the absence or presence of laminin (phagocytic index <5).

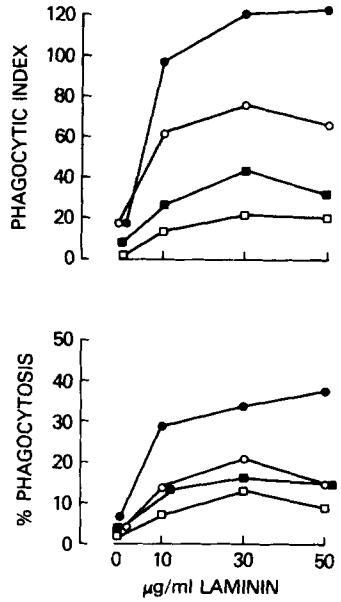

FIGURE 2. Laminin-induced phagocytosis of EAC4b by culture-derived human macrophages. Number of molecules of C4 per E: (●) 5,000; (○) 2170; (■) 1210; and (□) 576. (*A*) Effect of laminin on number of E phagocytosed per 100 macrophages. (*B*) Effect of laminin on percentage of macrophage ingesting at least one E. Data are representative of three experiments. Abscissa represents concentration of laminin used to coat Lab-Tek chambers.

TABLE I
Inhibition of Laminin-induced Phagocytosis by Anti-CR1

Incubation conditions	Phagocytic index*	
	EAC4b (500)‡	EAC4b (1,200)
Laminin (30 μg/ml)	24	48
Laminin + anti-CR_1	4	5
Laminin + anti-IgG	21	38
Buffer	10	14

* Each phagocytic index is the average of determinations from duplicate wells.
‡ Numbers in parentheses refers to the number of C4b molecules on each E.

Phagocytosis of EAC4b was inhibited up to 90% by preincubation of macrophages with mouse monoclonal antibody to human CR1, demonstrating that phagocytosis of EAC4b by macrophages was dependent on the interaction of CR1 with C4b on the surface of the sheep E (Table I). Preincubation of macrophages with HG11, a control monoclonal antibody directed against human IgG, did not significantly inhibit the phagocytosis of EAC4b.

Experiments with C3bi-opsonized E (11) showed that laminin also augmented phagocytosis of these targets by macrophages, even in the presence of anti-CR1 (data not shown). Thus, laminin exposure of macrophages induced both CR1- and CR3-mediated erythrocyte phagocytosis.

Effect of Laminin on Phagocytosis of EA_{IgG}. Experiments with EA_{IgG} demonstrated a similar enhancement of Fc-mediated phagocytosis by laminin (Fig. 3). Plating macrophages in laminin-coated wells for 1 h enhanced Fc-mediated phagocytosis over that observed in uncoated wells when the amount of IgG used to opsonize the E was less than that required to induce maximal erythrocyte phagocytosis. A more than twofold increase in the phagocytic index (Fig. 3*A*) and in the number of macrophages ingesting at least one E (Fig. 3*B*) was observed. Under these same conditions, macrophage phagocytosis of EA_{IgG} was enhanced slightly more by fibronectin than by laminin (Fig. 3). This is consistent with previous reports (1, 2) that fibronectin enhanced the Fc-mediated phagocytosis by freshly derived monocytes when suboptimal amounts of IgG were used to opsonize the E. When the concentration of anti-Forssman IgG used to opsonize the E was reduced even further, a reduction in macrophage phagocytosis occurred, but the relative increase in phagocytosis induced by laminin or fibronectin and the dose response for their effects were unchanged (data not shown).

Assessment of Fibronectin Contamination of the Laminin. Because of the similarity in the effects of laminin and fibronectin on macrophage phagocytic function, we considered the possibility that the laminin preparation was contaminated with fibronectin. The possibility that this potential contamination accounted for the enhancement of macrophage phagocytosis was excluded in several ways. First, laminin examined by SDS-PAGE under reducing conditions appeared pure. Second, the laminin contained <0.1% fibronectin by weight, as detected in an enzyme-linked immunosorbent assay (ELISA) capable of measuring 30 ng/ml of

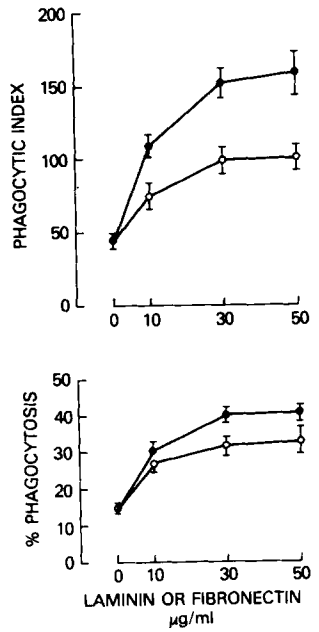

FIGURE 3. Laminin (○)- and fibronectin (●)-induced phagocytosis of EA_{IgG} by cultured human macrophages. (*A*) Increase in total number of E ingested per 100 macrophage. (*B*) Increase in percentage of cells phagocytosing at least one E. Data represent mean of three experiments performed in duplicate ± SEM with a single input of IgG anti-E. As in Fig. 2, abscissa denotes concentration of proteins used to coat chambers.

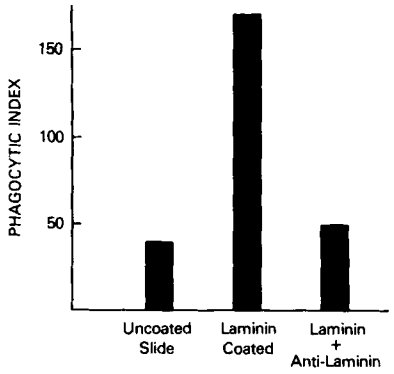

FIGURE 4. Laminin-induced phagocytosis was blocked by preincubation of laminin-coated wells with $F(ab')_2$ of rabbit antilaminin. EAC4b phagocytosis was measured when macrophages were incubated on uncoated wells, laminin-coated wells, or laminin-coated wells preincubated with antilaminin $F(ab')_2$.

mouse fibronectin. Third, the augmenting effect of laminin on phagocytosis could be completely inhibited when $F(ab')_2$ antilaminin was preincubated with laminin-coated wells before the addition of macrophages (Fig. 4); $F(ab')_2$ from preimmune serum failed to inhibit laminin's effect. In an assay capable of measuring as little as 5 ng/ml of rabbit antifibronectin, this antilaminin antiserum

contained no detectable antifibronectin. These results demonstrate that any potential fibronectin contamination of the laminin preparation must be far below the level required for a biological effect of fibronectin on phagocytosis (cf., Fig. 2 and reference 3). Thus, although their effects on macrophage phagocytic function are similar, fibronectin and laminin constitute distinct activation signals to the macrophage.

Assessment of Laminin Interaction With Macrophages and With Opsonized Particles. The design of the previous experiments was such that laminin was adhered to the Lab-Tek chamber, and unbound laminin was washed away before macrophages were added. Thus, it seemed most likely that laminin acted directly and solely on the macrophage, rather than through an interaction with the opsonized particle. To directly test this possibility, macrophages were preincubated with fluid phase laminin at various concentrations for 1 h at 37°C, followed by washing and plating on uncoated wells. This protocol led to a twofold enhancement of EAC4b phagocytosis (Fig. 5). Preincubation of EAC4b with laminin failed to enhance phagocytosis at any dose. This experiment demonstrates that fluid phase laminin can have the same augmenting effect on phagocytosis as surface-adherent laminin. It also provides further evidence that interaction between laminin and the opsonized erythrocyte is not required for laminin's effect on macrophage phagocytosis.

Effect of Laminin on Macrophage Adherence. To assure that laminin did not simply inhibit the binding of nonphagocytic macrophages to the glass slides (21), experiments were performed to quantitate the number of cells remaining in the wells under conditions identical to those of the phagocytosis assay. As shown in Table II, laminin coating caused a small but significant dose-dependent reduction in the number of adherent macrophages. At the highest concentration of laminin used to coat the wells in phagocytosis experiments, there was a 28% reduction in adherent cells. This small reduction in cell binding could not account for the laminin-induced increases in phagocytosis by selection of an especially phagocytic subpopulation, since up to a fourfold increase in the number of phagocytic macrophages occurred on exposure to laminin (Fig. 2*B*). Moreover, the dose response curves of the two laminin effects were different, since increases in

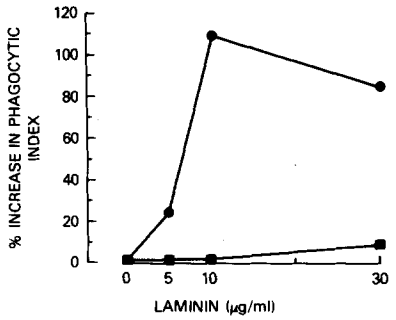

FIGURE 5. Fluid phase laminin induced increased EAC4b phagocytosis by macrophages. Macrophages were incubated with laminin at different concentrations, then washed with buffer three times, and allowed to adhere to glass before phagocytosis of EAC4b was determined (●). Preincubation of EAC4b (■) followed by washing did not enhance phagocytosis.

TABLE II
Inhibition of Macrophage Adherence to Laminin-coated Surfaces

Laminin (μg/ml)*	Percent cells recovered (±SEM)‡
10	89.3 ± 5.18
30	83.5 ± 0.24
50	72.0 ± 5.16

Results of three experiments, each done in duplicate wells. Experiments were done under conditions identical to those used in phagocytosis assays.
* Concentration of laminin used to coat wells.
‡ Percentage of cells recovered from uncoated wells.

phagocytosis reached a plateau at 30 μg/ml of laminin whereas inhibition of binding continued to increase at 50 μg/ml of laminin. Thus, laminin's effect on phagocytosis could not be accounted for by selection of an especially phagocytic subpopulation of cells.

Discussion

These experiments were performed to investigate the effect of the extracellular matrix protein, laminin, on macrophage phagocytosis. In these experiments, we used EAC4b as the primary target to probe the activity of CR1 in mediating phagocytosis. The interaction of CR1 with C4b has been described (22), and the use of EAC4b offers several advantages in studying CR1-mediated phagocytosis. Less time and fewer purified components are required to prepare this intermediate compared with the usual C3b-coated particle. Moreover, EAC3b may interact with CR3, the receptor for C3bi, and C3b on erythrocytes may be degraded by cellular proteases to C3bi (23–25). However, EAC4b rosettes with monocytes are entirely inhibited by 3D9, a monoclonal antibody to CR1, and are not inhibited at all by M1/70, a monoclonal antibody to CR3 (26 and unpublished observations). Moreover, degradation of C4b does not involve a stable intermediate that interacts with CR3. EAC4b bind to phagocytic cells because of a C4b-CR1 interaction, and there is no evidence that C4b or its degradation products interact with CR3 at any time during the phagocytic process. The use of EAC4b thus avoids one of the potential problems involved in the interpretation of data obtained with EAC3b.

Brief exposure of macrophages to laminin induced a major increase in phagocytosis of EAC4b, EAC3bi, and EA_{IgG}. This increase is most likely due to a direct effect of laminin on the macrophage and occurs both with laminin bound to a surface and with fluid phase laminin. Laminin's enhancement of phagocytosis is in many ways analogous to the ability of fibronectin to induce complement-mediated phagocytosis and to enhance antibody-dependent phagocytosis by human monocytes, polymorphonuclear leukocytes, and culture-derived macrophages (1–3). This is quite different from the effect of the lymphokine reported by Gresham and Griffin that enhances CR1-mediated phagocytosis only (27). A direct comparison of the magnitude of the increases in EA_{IgG} phagocytosis suggested that stimulation by laminin leads to slightly less augmentation than that by fibronectin. Nonetheless, the laminin and fibronectin dose responses

were quite similar on a weight basis. Bevilacqua et al. (2) also demonstrated augmented EA_{IgG} phagocytosis by monocytes on fibronectin monolayers. Although Wright et al. (3) did not confirm this, it is clear that the effect of extracellular matrix proteins on Fc-mediated phagocytosis are demonstrable only when concentrations of IgG are used that do not in themselves stimulate maximal phagocytosis. Thus, fibronectin and laminin have a general, rather than receptor-specific, effect on phagocytic function; this suggests that some global change in phagocyte plasma membrane function develops after exposure to these extracellular matrix proteins.

Although laminin and fibronectin share certain properties in addition to phagocytosis enhancement, including the ability to bind proteoglycans and collagen in vitro, there are clear functional differences between them. For example, fibronectin binds to several different types of collagen (the attachment of denatured collagen is the most avid), while laminin appears to bind type IV collagen quite selectively (7). In vitro, fibronectin appears (9) to interact with numerous cell types to mediate attachment and spreading on artificial substrates. By contrast, laminin appears (10, 28) to mediate attachment primarily of cell lines of epidermal origin, although macrophages and fibrosarcoma cells also have been shown to bind laminin. Moreover, while fibronectin is an apparently ubiquitous component of the extracellular matrix, laminin is found only in basement membranes. Distinctly different effects on cell spreading and attachment by these two connective tissue glycoproteins have been demonstrated on cells of the monocyte-macrophage lineage. Giavazzi and Hart (21) reported evidence that monocyte attachment to plastic dishes was inhibited by laminin; this effect was diminished during in vitro differentiation into macrophages. By contrast, fibronectin slightly enhanced substrate attachment and spreading of monocytes and macrophages. We also have observed that both monocytes and macrophages spread poorly on laminin-coated glass compared with uncoated and fibronectin-coated glass. Direct measurements of the number of adherent macrophages demonstrated a 28% decrease in cells adherent to wells coated with laminin compared with control wells. Since laminin increased the number of actively phagocytic macrophages up to fourfold, its augmentation of macrophage phagocytosis could not be due to the selection of a more adherent, more phagocytic subpopulation of cells. Thus, both laminin and fibronectin activate macrophages to a more highly phagocytic state while exhibiting totally different effects on the other plasma membrane-mediated processes of attachment and spreading.

Epithelial tumors with laminin receptors are much more highly metastatic than tumors without receptors (29), apparently because interaction with laminin induces the tumor cell to secrete enzymes that degrade the basement membrane and thus allow passage of the cell from the epithelium to the blood stream. The phagocytic cell is required to undergo the opposite migration, from the blood stream into extravascular areas of inflammation and infection. Thus, interaction of connective tissue components, such as laminin and fibronectin, would logically be expected to transmit to the phagocytic cell the signal that it is out of the bloodstream and in an area of inflammation, where maximal phagocytic function is required. Accumulated data suggest that fibronectin, laminin, and serum

amyloid P component (3) all provide such signals. These very different proteins have a final common effect on the macrophage—to increase its phagocytic potential. To determine the pathway by which occupancy of the various membrane receptors for extracellular matrix proteins affects phagocytic function has become an important question in the biology of these cells.

Summary

Brief exposure of culture-derived human macrophages to laminin, a glycoprotein component of all mammalian basement membranes that has a molecular weight of 1,000,000, led to enhancement of subsequent macrophage phagocytosis of EAC4b, EAC3bi, and EA_{IgG} (sheep eyrthrocytes sensitized with IgG anti-Forssman antibody). This effect on macrophage phagocytosis occurred with both substrate-adherent and fluid phase laminin. Preincubation of macrophages, but not of EAC4b, with laminin led to augmentation of phagocytosis, suggesting that interaction with the phagocytic cell, but not with the opsonized particle, was required for laminin's effect. Laminin-stimulated phagocytosis of EAC4b was blocked entirely by a monoclonal antibody to CR1. Direct comparison of the phagocytic ability of macrophages adherent to laminin- and fibronectin-coated glass slides showed that fibronectin had a somewhat greater enhancing effect on phagocytosis. Nonetheless, the phagocytosis-enhancing effect of laminin was not due to contamination of the purified laminin preparation by fibronectin, since the laminin preparation was free of fibronectin, as determined by sodium dodecyl sulfate–polyacrylamide gel electrophoresis and enzyme-linked immunosorbent assay; in addition, laminin-enhanced phagocytosis was decreased in the presence of laminin-specific antibodies. Laminin inhibited macrophage adherence and spreading, but selection of a laminin-binding macrophage subpopulation could not account for the laminin-induced increases in phagocytosis. We hypothesize that interaction with extracellular matrix proteins may represent an important activation stimulus both to the macrophages normally present in the extravascular compartment and to the phagocytic cells that have emigrated from the bloodstream into areas of inflammation.

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