Relation of the CD11/CD18 Family of Leukocyte Antigens to the Transient Neutropenia Caused by Chemoattractants

By Claes Lundberg and Samuel D. Wright

Adherence of leukocytes to the endothelium is a prerequisite for infiltration and accumulation of cells at an inflammatory site. Recent studies suggest that the CD11/CD18 family of adhesion-promoting receptors plays a crucial role in the initial adherence of polymorphonuclear leukocytes (PMNs) to endothelium. We have studied the effect of the anti-CD18 monoclonal antibody (MoAb) IB4, on movement of PMN in rabbits. Accumulation of PMNs in the skin induced by a local injection of the chemoattractant, zymosan-activated serum (ZAS), was strongly inhibited, in a dose-dependent fashion, by intravenous injection of IB4. A greater than 95% reduction in PMN accumulation was seen with 1 mg IB4/kg body weight, the highest dose used. PMN-dependent plasma leakage in the ZAS-injected skin sites was also inhibited by pretreatment with MoAb IB4, with a similar dose dependence. Histamine-induced plasma leakage, which is PMN independent, was not affected by this treatment. F(ab)$_2$ fragments of IB4 were as effective as the whole immunoglobulin G molecule in reducing PMN accumulation. The half-life of circulating IB4 in rabbits was found to be 11.5 hours. These results are consistent with in vitro studies that show that binding of PMNs to endothelium requires both expression of CD11/CD18 molecules and activation of the PMNs by agonists, and confirm that sites on CD11/CD18 that recognize endothelial cells are blocked by IB4. Other investigators have shown that injection of chemoattractants into the blood stream causes a rapid neutropenia associated with accumulation of PMNs in the lung. We find that intravenous treatment of animals with IB4 did not block the transient accumulation of PMNs in the lung induced by formyl-methionyl-leucyl-phenylalanine, suggesting that this accumulation occurs by a mechanism that does not require CD11/CD18 molecules.

ADHERENCE of leukocytes to the endothelium is a prerequisite for extravasation of leukocytes, and enhanced adhesion at sites of inflammation serves in the recruitment of cells to specific sites. Several types of molecules may participate in the enhanced adhesion observed at sites of inflammation. The endothelial molecules ICAM-1 (ref 1) and ELAM-1 (ref 2) serve as adhesins for polymorphonuclear leukocytes (PMNs) and are strongly induced after 3 hours' exposure of endothelium to cytokines. It is known, however, that PMNs bind endothelium and diapedese within minutes of the introduction of a chemotactic stimulus in tissues,[3] well before the induction of ICAM-1 or ELAM-1. This type of rapid extravasation appears, instead, to depend on the response of PMNs to chemoattractants and on the presence of the CD11/CD18 family of glycoproteins, LFA-1 (CD11a/CD18), CR3 (CD11b/CD18), and p150,95 (CD11c/CD18) on the PMN surface. Stimulation of PMNs with several chemotactic factors causes strong adhesion to unstimulated endothelium in vitro,[4] and essentially all the chemoattractant-stimulated adhesion can be inhibited by treating the PMNs with monoclonal antibodies (MoAbs) against the CD11/CD18 complex.[5-8] Further, PMNs from patients with a genetic deficiency in the CD11/CD18 complex (leukocyte adhesion deficiency) fail to bind unstimulated endothelium in vitro.[5,8]

We studied the movement of PMNs into the tissue of rabbits in response to local injection of chemoattractants. We found that pretreatment of the rabbits with IB4 or F(ab)$_2$ fragments of IB4, an MoAb directed against human CD18, blocks movement of PMNs into dermal sites injected with zymosan-activated serum (ZAS). The dose dependence of the blockade and the half-life of the IB4 in circulation were determined. These studies confirm and extend previous reports using different monoclonal anti-CD18 antibodies,[9-11] and define a dosing regimen that strongly blocks CD18 function.

If chemoattractants are injected systemically rather than locally, a striking transient neutropenia results.[12,13] The number of circulating PMNs declines sharply within 5 minutes and returns to normal levels by 30 minutes. This neutropenia is associated with accumulation of PMNs in the lung. The time course of neutropenia corresponds precisely with the time course of enhanced CD11/CD18-dependent binding to endothelium[8] or to C3bi-coated erythrocytes[14] observed in vitro, suggesting that the neutropenia may result from CD11/CD18-dependent adhesion of PMNs to the endothelium. We found, however, that blockade of CD11/CD18 with IB4 had no effect on neutropenia caused by formyl-methionyl-leucyl-phenylalanine (fMLF). Mechanisms other than CD11/CD18-dependent adhesion must therefore function to produce this type of neutropenia.

MATERIALS AND METHODS

Monoclonal Antibody

The murine anti-CD18 hybridoma cell line IB4[15] was cultured in Dulbecco's modified Eagle's cell culture medium supplemented with 1% fetal calf serum for 11 days. Cross-flow filtration was used to concentrate protein and remove cells and debris. Further purification of the immunoglobulin (Ig) G2a was done by gel filtration (Sephadex G25) and cation exchange (S-Sepharose, Pharmacia, Uppsala, Sweden). IB4 was finally dissolved in phosphate-buffered saline, (PBS), pH 7.4. F(ab)$_2$ fragments were produced by pepsin digestion and purified using anion exchange (Mono Q, Pharmacia, Uppsala,

From the Department of Inflammation Research, Pharmacia AB, Uppsala, Sweden; and the Laboratory of Cellular Physiology and Immunology, The Rockefeller University, New York, NY.

Submitted January 9, 1990; accepted May 22, 1990.

Supported by US Public Health Service grants no. AI22003 and AI24775. S.D.W. is an Established Investigator of the American Heart Association.

Address reprint requests to Samuel D. Wright, PhD, The Laboratory of Cellular Physiology and Immunology, The Rockefeller University, 1230 York Ave, New York, NY 10021.

The publication costs of this article were defrayed in part by page charge payment. This article must therefore be hereby marked "advertisement" *in accordance with 18 U.S.C. section 1734 solely to indicate this fact.*

0006-4971/90/7606-0013$3.00/0

Sweden) and gel filtration (Superose 6). Endotoxin levels in the IgG and $F(ab)_2$ were analyzed using Coatest (KabiVitrum, Stockholm, Sweden) and found to be 1.9 and 0.3 Endotoxin Units (EU)/mg protein, respectively.

Animals and Anesthesia

Male New Zealand White rabbits weighing 1.5 to 2.0 kg were used. The animals were anesthetized with Saffan (6 mg/kg body weight IV; Glaxovet Ltd, Harefield, Uxbridge, England) when surgical anesthesia was required. The backs of the animals were shaved 1 day before the experiment when applicable.

Inflammatory Skin Lesions

After collecting an arterial blood sample from the central ear artery for hematocrit and leukocyte counting, 5 μCi of ^{125}I-labeled human serum albumin was given intravenously as a plasma tracer. Physiologic saline, histamine (10^{-3} mol/L), or ZAS, in a volume of 100 μL, was then injected intradermally (ID) in triplicate sites in the dorsal region of the anesthetized animals. Activation of rabbit serum with zymosan (20 mg/mL zymosan, 60 minutes, 37°C) results in approximately 3 μg of C5a[desArg]/mL serum.[16] The animals recovered from the anesthesia within 5 minutes and were kept in their cages. The ambient room temperature was kept at 25°C. After a period of 4 hours, the animals were reanesthetized, a blood sample was collected for determination of hematocrit, leukocyte count, and circulating mouse IgG levels, and the rabbits were killed by an overdose of anesthetic. All solutions used for injection into animals were assayed for endotoxin and were found to be lower than 0.02 ng/mL (detection limit of assay).

Assay of PMN Infiltration and Plasma Leakage

The injection sites, together with two samples of normal intact skin, were carefully removed, dissected free from fat and muscle tissue, and cut into four smaller pieces. After radioactivity measurements of tissue, blood, and plasma samples were made, the skin lesions were freeze-thawed once, ground in 10 mL 0.05 mol/L phosphate buffer, pH 6.0, containing 0.5% hexadecyltrimethylammonium bromide, and myeloperoxidase (MPO) activity was determined in the supernatants. MPO has previously been shown to be a useful and reliable marker enzyme for PMN accumulation in vivo,[17] and was therefore used to assess PMN accumulation in the inflammatory skin lesions in this study. The MPO activity was assayed by measuring the H_2O_2-dependent oxidation of 3,3',5,5'-tetramethylbenzidine.[18] MPO standards (The Green Cross Corp, Osaka, Japan) were included in each assay, and these results were used to construct a dose-response curve from which the MPO activity of the samples was determined. One unit of MPO activity was defined as the amount of enzyme reducing 1 μmol H_2O_2/min.[19] Albumin extravasation was calculated from the radioactivity of the tissue and plasma sample and is expressed as microliters of plasma exuded per gram wet weight. Values from each animal represent a mean of values from three injection sites.

Experimental Protocols

Protocol I. Ten minutes before the intradermal injections, the animals were treated intravenously with IB4 in doses of 0.03 (n = 5), 0.1 (n = 5), 0.3 (n = 5), 1.0 mg/kg body weight (n = 5), or 1 mL saline/kg body weight (controls; n = 5).

Protocol II. Ten minutes prior to the intradermal injections, the animals were treated intravenously with IB4 $F(ab)_2$ fragments in doses of 0.1 mg/kg body weight (n = 6), 1.0 mg/kg body weight (n = 6), or 1 mL saline/kg body weight (controls; n = 6).

Protocol III. Five minutes after collecting an arterial blood sample from the central ear artery, 1 nmol of fMLF/kg body weight was given intravenously. Additional leukocyte counting was performed at 2, 10, 30, and 60 minutes after the fMLF injection. The rabbits were pretreated with either PBS (1 mL/kg body weight IV) or IB4 (1.0 mg/kg body weight IV) 10 minutes before the fMLF injection. In addition, the myeloperoxidase activity in lungs was determined in three control rabbits that received PBS intravenously and in three rabbits that received fMLF. Two minutes after injection, the animals were killed, their lungs removed, and the myeloperoxidase activity analyzed as described above for skin tissues.

Murine IgG Concentration in Blood

In some experiments (see Fig 6), levels of murine IgG in rabbit plasma were assayed using an enzyme-linked immunosorbent assay (ELISA) technique. Microtiter plates were coated with a goat–anti-mouse IgG+M(H+L) (Jackson Immunoresearch Lab, Inc, Partille, Sweden) overnight, then incubated with known concentrations of murine IgG or with rabbit plasma samples for 2 hours at 37°C. Alkaline phosphatase-conjugated goat–anti-mouse-IgG+M(H+L) was then added (2 hours, 37°C), and p-Nitrophenyl phosphate (Sigma Chemical Co, St Louis, MO) was used as a substrate for a color reaction. OD_{405} was measured in an ELISA reader.

Statistics

Statistical differences between treatments were evaluated using one-way analysis of variance with multiple range testing according to the method of least significant differences. A difference at the 5% level was considered significant and is denoted in the figures by an asterisk. Data in text and figures are given as mean ± SD.

RESULTS

fMLF-Induced Neutropenia

fMLF, 1.0 nmol/kg body weight injected intravenously into rabbits, caused a 94% reduction in circulating peripheral blood PMNs within 2 minutes of injection (Fig 1). Loss of PMNs from the circulation was associated with retention of cells in the lung since the myeloperoxidase in lung tissue rose from 0.27 ± .05 μm/g (n = 3) to 1.18 ± .29 μm/g (n = 3) within 2 minutes of the injection of fMLF. The peripheral PMN count returned to a normal level after a 30-minute

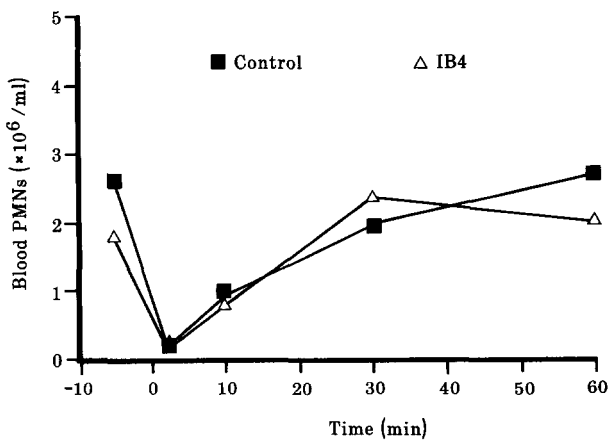

Fig 1. IB4 does not prevent neutropenia in response to fMLF. Animals were pretreated with PBS (n = 6) or with IB4 (1.0 mg/kg body weight, n = 6) 10 minutes before injection with fMLF (1 nmol/kg body weight). At intervals, the density of PMNs in peripheral blood was determined.

period (Fig 1). Animals pretreated with IB4 (1.0 mg/kg body weight IV) 10 minutes before the fMLF injection showed a decrease in peripheral blood PMNs that was identical in extent (93%) and time course to that observed in control rabbits (Fig 1). The inability of anti-CD18 antibody to block transient neutropenia was confirmed in two additional experiments. Injection of ZAS (a source of C5a) caused a similar transient neutropenia, and pretreatment of animals with IB4 had no effect on the time course or extent of the neutropenia (data not shown). We also used a different anti-CD18 MoAb (60.3) and again observed no alteration in the time course or extent of fMLF-induced neutropenia (data not shown). These findings suggest that the CD18 molecules on PMNs are not involved in neutropenia caused by chemoattractants. To verify this surprising result, it was necessary to rigorously prove that the dosing regimen we used was sufficient to completely inhibit CD18 function.

Effect of IB4 on Movement of PMN Into Skin

ZAS injected intradermally into the dorsal region of control rabbits caused accumulation of PMNs at the injection site, measured by myeloperoxidase activity of the homogenized skin lesion (Fig 2A). Saline and histamine did not induce such PMN accumulation. Plasma leakage at the site of injection could be observed for both ZAS and histamine (Fig 2B). The leakage that follows ZAS injection is known to be PMN dependent, whereas histamine-induced plasma leakage is a PMN-independent process.[20]

Treatment of the animals with the MoAb IB4 10 minutes before the intradermal injections caused a dose-dependent decrease in ZAS-induced PMN accumulation (Fig 2A). Virtually complete inhibition of accumulation was observed at the highest dose (1 mg IB4/kg body weight) and significant reduction (>50%) was observed with as little as 0.03 mg/kg body weight. A similar dose-dependent reduction of ZAS-induced plasma leakage was also observed (Fig 2B), but the PMN-independent histamine-induced plasma leakage was unaffected by IB4 treatment (Fig 2B). The dose of IB4 needed for half-maximal inhibition of PMN accumulation in our experiments is approximately 30-fold less than the dose of another anti-CD18 MoAb, 60.3, needed for half-maximal blockade of movement of PMNs into endotoxin-containing sponges.[10]

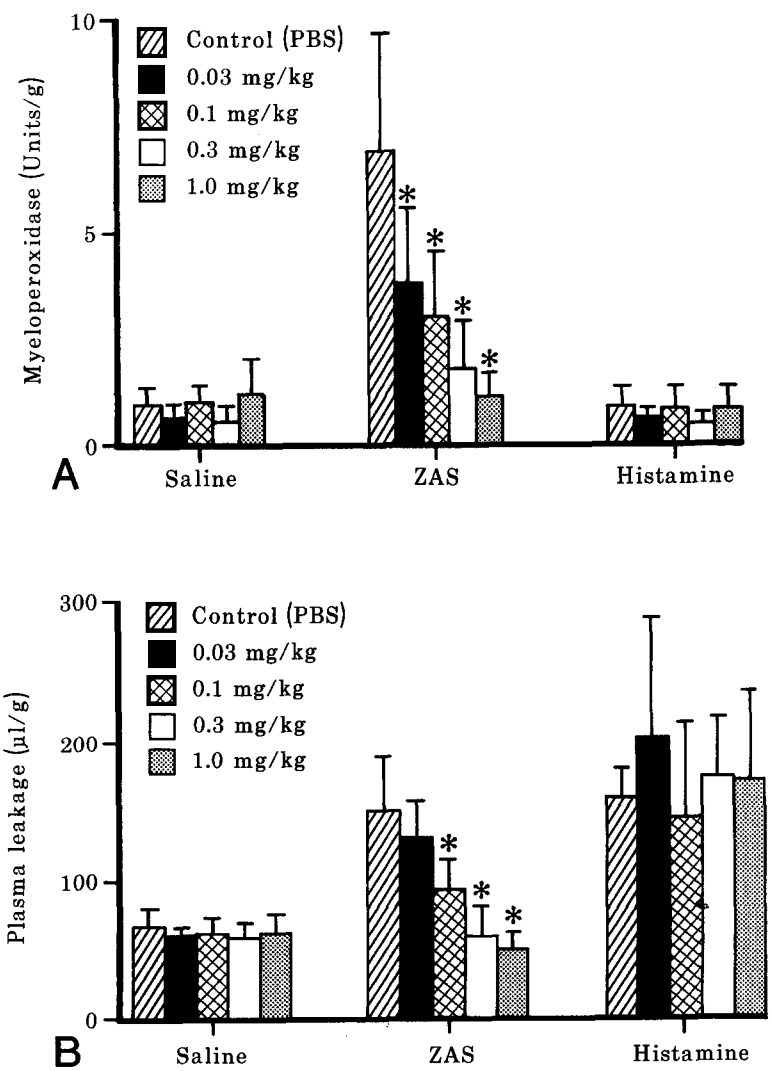

Fig 2. IB4 inhibits PMN accumulation and plasma leakage in response to ZAS. Saline, ZAS, or histamine were injected into animals pretreated with either PBS (control) or IB4 in doses ranging from 0.03 to 1.0 mg/kg body weight IV. Accumulation of PMNs (A) or leakage of plasma (B) was measured after 4 hours (n = 5).

The decreased accumulation of PMNs at the injection site of IB4-treated animals was not due to a paucity of PMNs in the circulation. Peripheral blood PMN counts showed an approximately twofold increase by the end of the 4-hour experimental period, presumably due to the experimental procedure, and IB4 had no significant effect on PMN levels (Fig 3A). Peripheral blood mononuclear cell density, however, increased slightly at the highest IB4 dose (1.0 mg/kg body weight, Fig 3B). The hematocrit of the experimental animals ranged between 33.0 and 35.8, and exhibited no changes after any of the experimental manipulations.

The Fc domain of IB4 was not necessary for the blockade of PMN emigration. $F(ab)_2$ fragments of IB4 caused an 87% inhibition in ZAS-induced accumulation of PMNs in the skin at 1.0 mg/kg body weight (Fig 4). To our knowledge, this is the first demonstration that $F(ab)_2$ fragments of an anti-CD18 mAb are effective in vivo.

Half-life of IB4 in the Circulation

The plasma levels of murine IgG were followed for 48 hours in rabbits that had received 3 mg IB4/kg body weight IV (Fig 5). The half-life of IB4 was found to be 11.5 hours, with circulating levels of IB4 well above saturation for the duration of the experiments described here.

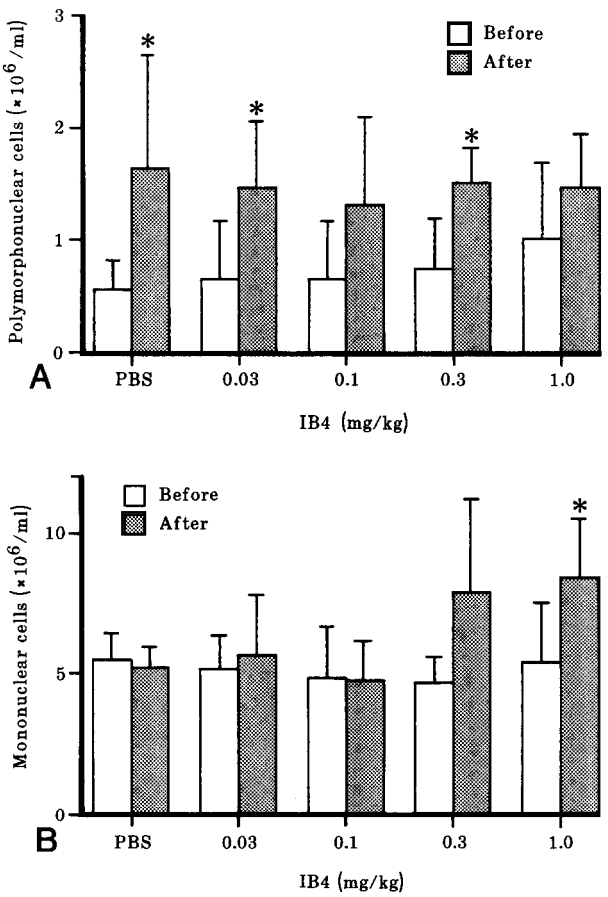

Fig 3. IB4 does not cause neutropenia. The density of peripheral PMNs (A) and mononuclear cells (B) was determined before and 4 hours after injection of PBS (control) or various doses of IB4 (n = 5).

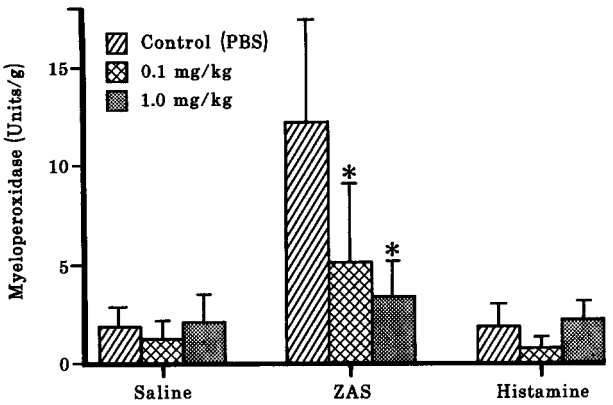

Fig 4. $F(ab)_2$ fragments of IB4 inhibit PMN accumulation in response to ZAS. Animals were pretreated with either PBS (control, n = 6) or $F(ab)_2$ fragments of IB4 in two doses, 0.1 (n = 7) and 1.0 (n = 5) mg/kg body weight IV Saline, ZAS, or histamine were injected intradermally, and accumulation of PMN was determined after 4 hours.

DISCUSSION

Systemic administration of chemoattractants causes transient neutropenia with a time course characteristic of the chemoattractant used. For example, C5a causes very rapid neutropenia (1 minute) that returns to baseline by 15 minutes,[12,13] and tumor necrosis factor causes a slower neutropenia (30 minutes) that returns to baseline by 60 minutes.[21] Neutropenia caused by chemoattractants is associated with an accumulation of PMNs in the capillaries of the lungs, and the rebound in the number of circulating PMNs is

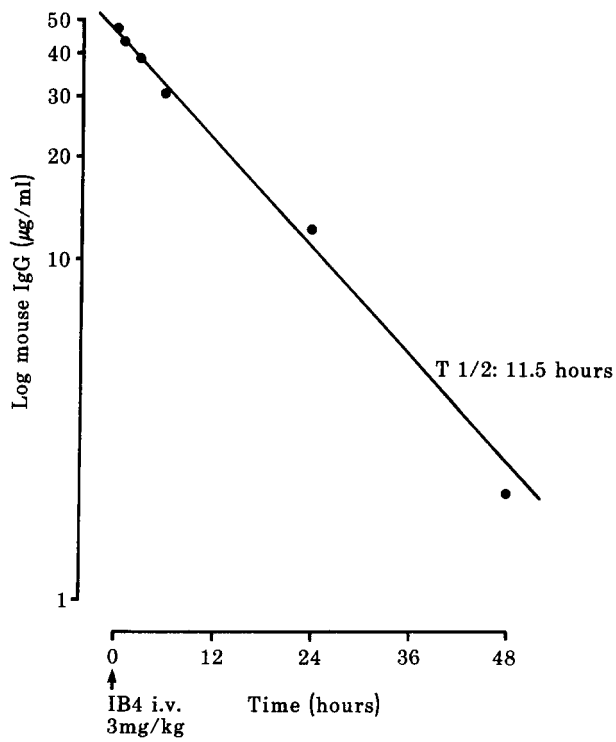

Fig 5. Half-life of circulating IB4. Plasma levels of murine IgG were determined after an intravenous injection of IB4 (3 mg/kg body weight) in rabbits (n = 6).

associated with a loss from the lungs (ref 22 and this study). Thus, the loss of PMNs from the peripheral circulation in response to chemoattractants appears to represent a transient "interaction" of PMNs with the endothelium in the lung. Candidate molecules that may mediate an adhesive interaction include the CD11/CD18 complex on PMNs. These receptors respond to chemoattractants in vitro with transiently enhanced adhesive capacity for endothelium, and the time course of the response to different chemoattractants corresponds closely to the time course of transient neutropenia observed in vivo.[8,23] Furthermore, the drug cytochalasin D has been shown to block both the chemoattractant-induced retention of PMNs in lungs[24] and the capacity of CD11b/CD18 to mediate adhesive events.[25] The data described here, however, suggest that the CD11/CD18 complex is not necessary for transient neutropenia. The concentrations of IB4 we used were sufficient to block by at least 95% the adhesion of PMNs to endothelium in vitro,[8,23] the movement of PMNs across the blood-brain barrier during meningitis in rabbits,[26] and the movement of PMNs into rabbit skin (Fig 2), but we observed no change in the extent or kinetics of the transient neutropenia caused by fMLF (Fig 1). Thus, retention of PMNs in the lung under these conditions is unlikely to be due to a CD11/CD18-dependent adhesion of PMNs with endothelium.

PMNs may aggregate in response to chemoattractants, and the presence of aggregates may prevent passage through the lung. However, PMN aggregation is completely dependent on CD11/CD18 molecules on PMNs and is completely blocked by IB4.[27] Thus, aggregation of PMNs is an unlikely explanation for the phenomenon of transient neutropenia caused by chemoattractants.

Worthen et al[24] recently suggested an alternative mechanism for the neutropenia caused by chemoattractants. fMLF causes a transient increase in the amount of filamentous actin within PMNs,[28] which is associated with a decrease in the deformability of the cells.[24] Since chemoattractants cause PMNs to accumulate in the narrowest capillaries of the lung, their retention may result from an inability of cells to deform sufficiently for passage. The "interaction" of rigid PMNs with the capillary endothelium would thus be one of physical contact rather than adhesion mediated by receptors. Since actin polymerization may contribute to both enhanced cellular stiffness[20] and enhanced CD11/CD18 function,[25] the similar time course observed for changes in CD11/CD18 activity and retention of PMNs in the lung could derive from the dependence of both phenomena on a common antecedent event.

CD11/CD18 molecules appear not to be required for the transient interactions of PMNs in the "marginated pool" with endothelium, since injection of epinephrine into a CD11/CD18-deficient patient caused a normal increase in the number of circulating PMNs.[29] Similarly, our results show that CD11/CD18 molecules are not necessary for the interactions with endothelium that underlie the transient neutropenia caused by chemoattractants. However, CD11/CD18 molecules may play a role in other forms of transient interactions of PMNs with endothelium. Systemic injection of lipopolysaccharide causes a slow (30 minute) neutropenia due principally to accumulation of PMNs in the lung, and recent studies show that blockade of CD18 with MoAb IB4 prevents this response.[30] Thus, transient interaction of PMNs with endothelium may occur by both CD11/CD18-dependent and -independent mechanisms.

ACKNOWLEDGMENT

We are grateful to Inger Fogelberg, Annika Grahn-Westin, and Elisabeth Hammar for excellent technical assistance, and P.A. Detmers, W.A. Muller, S.K. Lo, and A. Hermanowski-Vosatka for critical reading of the manuscript.

REFERENCES

1. Dustin ML, Rothlein R, Bhan AK, Dinarello CA, Springer TA: Induction by IL 1 and interferon-gamma: Tissue distribution, biochemistry, and function of a natural adherence molecule (ICAM-1). J Immunol 137:245, 1986

2. Bevilacqua MP, Pober JS, Mendrick DL, Cotran RS, Gimbrone MA Jr: Identification of an inducible endothelial-leukocyte adhesion molecule. Proc Natl Acad Sci USA 84:9238, 1987

3. Cybulsky MI, Colditz IG, Movat HZ: The role of interleukin-1 in neutrophil leukocyte emigration induced by endotoxin. Am J Pathol 124:367, 1986

4. Harlan JM: Leukocyte-endothelial interactions. Blood 65:513, 1985

5. Harlan JM, Killen PD, Senecal FM, Schwartz BR, Yee EK, Taylor FR, Beatty PG, Price TH, Ochs HD: The role of neutrophil membrane glycoprotein GP-150 in neutrophil adhesion to endothelium in vitro. Blood 66:167, 1985

6. Zimmerman GA, McIntyre TM: Neutrophil adherence to human endothelium in vitro occurs by CDw18 (Mol, MAC-1/LFA-1/GP 150,95) glycoprotein-dependent and -independent mechanisms. J Clin Invest 81:531, 1988

7. Smith CW, Rothlein R, Hughes BJ, Mariscalco MM, Rudloff HE, Schmalstieg FC, Anderson DC: Recognition of an endothelial determinant for CD18-dependent human neutrophil adherence and transendothelial migration. J Clin Invest 82:1746, 1988

8. Lo SK, Detmers PA, Levin SM, Wright SD: Transient adhesion of neutrophils to endothelium. J Exp Med 169:1779, 1989

9. Arfors K-E, Lundberg C, Lindblom L, Lundberg K, Beatty PG, Harlan JM: A monoclonal antibody to the membrane glycoprotein complex CD18 inhibits polymorphonuclear leukocyte accumulation and plasma leakage in vivo. Blood 69:338, 1987

10. Price TH, Beatty PG, Corpuz SR: In vivo inhibition of neutrophil function in the rabbit using monoclonal antibody to CD18. J Immunol 139:4174, 1987

11. Nourshargh S, Rampart M, Hellewell PG, Jose PJ, Harlan JM, Edwards AJ, Williams TJ: Accumulation of 111In-neutrophils in rabbit skin in allergic and non-allergic inflammatory reactions in vivo. J Immunol 142:3193, 1989

12. Lundberg C, Marceau F, Hugli TE: C5a-induced hemodynamic and hematologic changes in the rabbit. Role of cyclooxygenase products and polymorphonuclear leukocytes. Am J Pathol 128:471, 1987

13. O'Flaherty JT, Showell HJ, Ward PA: Neutropenia induced by systemic infusion of chemotactic factors. J Immunol 118:1586, 1977

14. Wright SD, Meyer BC: Phorbol esters cause sequential

activation and deactivation of complement receptors on polymorphonuclear leukocytes. J Immunol 136:1759, 1986

15. Wright SD, Rao PE, VanVoorhis WC, Craigmyle LS, Iida K, Talle MA, Westberg EF, Goldstein G, Silverstein SC: Identification of the C3bi receptor of human monocytes and macrophages by using monoclonal antibodies. Proc Natl Acad Sci USA 80:5699, 1983

16. Wagner JL, Hugli TE: Radioimmunoassay for anaphylatoxins: A sensitive method for determining complement activation products in biological fluids. Anal Biochem 136:75, 1984

17. Lundberg C, Arfors K-E: Polymorphonuclear leukocyte accumulation in inflammatory dermal sites as measured by 51Cr-labelled cells and myeloperoxidase. Inflammation 7:247, 1983

18. Suzuki I, Ota H, Sasagawa S, Sakatani T, Fujikura T: Assay method for myeloperoxidase in human polymorphonuclear leukocytes. Anal Biochem 132:345, 1983

19. Worthington Enzyme Manual. Freehold, NJ, Worthington Biochemical Corporation, 1972

20. Lundberg C, Bjrk J, Lundberg K, Arfors K-E: Polymorphonuclear leukocyte (PMNL) dependent and independent microcirculatory plasma leakage. Prog Appl Microcirc 1:86, 1983

21. Ulich TR, delCastillo J, Keys M, Granger GA, Ni R-X: Kinetics and mechanisms of recombinant human interleukin 1 and tumor necrosis factor-alpha-induced changes in circulating numbers of neutrophils and lymphocytes. J Immunol 139:3406, 1987

22. Meyrick BO, Brigham KL: The effect of single infusion of zymosan-activated plasma on the pulmonary microcirculation of sheep. Am J Pathol 114:32, 1984

23. Lo SK, VanSeventer GA, Levin SM, Wright SD: Two leukocyte receptors (CD11a/CD18 and CD11b/CD18) mediate transient adhesion to endothelium by binding to different ligands. J Immunol 143:3325, 1989

24. Worthen GS, Schwab B III, Elson EL, Downey GP: Mechanics of stimulated neutrophils: Cell stiffening induces retention in capillaries. Science 245:183, 1989

25. Detmers PA, Olsen E, Cohn ZA: Cytochalasin D inhibits the binding activity of complement receptors on human neutrophils. FASEB J 2:A1450, 1988

26. Tuomanen EI, Saukkonen K, Sande S, Cioffe C, Wright SD: Reduction of inflammation, tissue damage, and mortality in bacterial meningitis in rabbits treated with monoclonal antibodies against adhesion-promoting receptors of leukocytes. J Exp Med 170:959, 1988

27. Buyon JP, Hopkins P, Abramson SB, Winchester R: Differential participation of epitopes on alpha and beta chains of the LFA family in neutrophil aggregation as defined by workshop monoclonal antibodies, In McMichael AJ (Ed): Leucocyte Typing III. Oxford, UK, Oxford University Press, 1988, p 844

28. Wallace PJ, Wersto RP, Packman CH, Lichtman MA: Chemotactic peptide-induced changes in neutrophil actin conformation. J Cell Biol 99:1060, 1984

29. Buchanan MR, Crowley CA, Rosin RE, Gimbrone MA, Babior BM: Studies on the interaction between GP-180-deficient neutrophils and vascular endothelium. Blood 60:160, 1982

30. Cooper JA, Neumann PH, Wright SD, Malik AB: Pulmonary vascular sequestration of neutrophils in endotoxemia: Role of CD18 leukocyte surface glycoprotein. Am J Respir Dis 139:A301, 1989