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Abnormal binding properties of blood monocytes in rheumatoid arthritis

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Abstract

Blood monocytes from patients with RA exhibited a greater binding to monolayers of umbilical cord vein endothelium than monocytes from control subjects (mean 42% increase; $p<0.01$). When control monocytes were added to TNF or IL-1 treated endothelium their adhesion was enhanced (mean 24% increase; $p<0.05$), whereas the number of monocytes from RA patients binding to TNF or IL-1 treated monolayers was less than that adhering to untreated endothelial cells (mean 22% inhibition; $p<0.02$). The surface expression of CD11b/CD18 on RA monocytes was increased and pretreatment of normal and RA cells with an anti-CD18 monoclonal antibody inhibited their attachment to untreated and cytokine-treated endothelial cells. Normal blood monocytes activated with LPS demonstrated an enhanced binding to untreated cultures (mean 23% increase; $p<0.05$) and an inhibited attachment to cytokine-treated endothelial cells. This study suggests that blood monocytes in RA may be activated and that this property modifies the attachment of these cells to normal and "inflammatory" endothelium.

Introduction

Macrophages occupy a prominent position in the immunopathogenesis of rheumatoid arthritis (RA) and their proliferation within the rheumatoid synovium may depend upon the recruitment of blood monocytes [1]. For monocytes to enter synovial tissue they must first bind to adhesion determinants on the surface of vascular endothelium. In RA, circulating monocytes may be predisposed to enter inflammatory lesions and to investigate this possibility, we compared the adherent properties of rheumatoid monocytes with control monocytes using monolayers of umbilical cord vein endothelium. Since inflammatory cytokines act on endothelial cells to increase their adhesiveness for leucocytes [2], a further characterisation was undertaken using endothelial monolayers treated with tumour necrosis factor (TNF) and interleukin-1 (IL-1). Both control and RA monocytes were examined for their expression of the β_2 leucocyte integrins.

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Materials and methods

Monocytes were isolated from the blood of 15 RA patients and 15 control subjects (patients attending a rheumatology clinic but with no evidence of

inflammatory disease) by a standard technique. A quantitative monolayer adhesion assay was used to evaluate their adherent properties [3]. The monocytes were labelled with ^{51}Cr and added to monolayers of umbilical cord vein endothelium that were either untreated or pretreated with 10 U/ml TNF or IL-1 for 4 h. In order to assess the binding characteristics of activated monocytes, cells were treated with lipopolysaccharide for 18 h [4]. FACS analysis was employed to monitor the expression of CD11a, CD11b, CD11c/CD18 on the surface of monocytes.

Results

Blood monocytes from RA patients were more adherent than control monocytes (mean 42% increase; $p<0.01$) to untreated endothelial monolayers. The enhanced adhesiveness of the rheumatoid monocytes was not related to the patients' disease activity. Figure 1 shows that treatment of endothelial cells with TNF or IL-1 enhanced the binding of normal monocytes (mean 27 and 22% increase, respectively; $p<0.05$). However, there were fewer rheumatoid monocytes binding to TNF [mean 25% inhibition; $p<0.02$) and IL-1 (mean 20% inhibition; $p<0.02$)] treated endothelium when compared with their basal adhesion. Of the CD11/CD18 family of adhesion molecules, FACS analysis revealed that there was increased expression of CD11b ($p<0.005$) and its β chain CD18 ($p<0.001$) on rheumatoid monocytes when com-

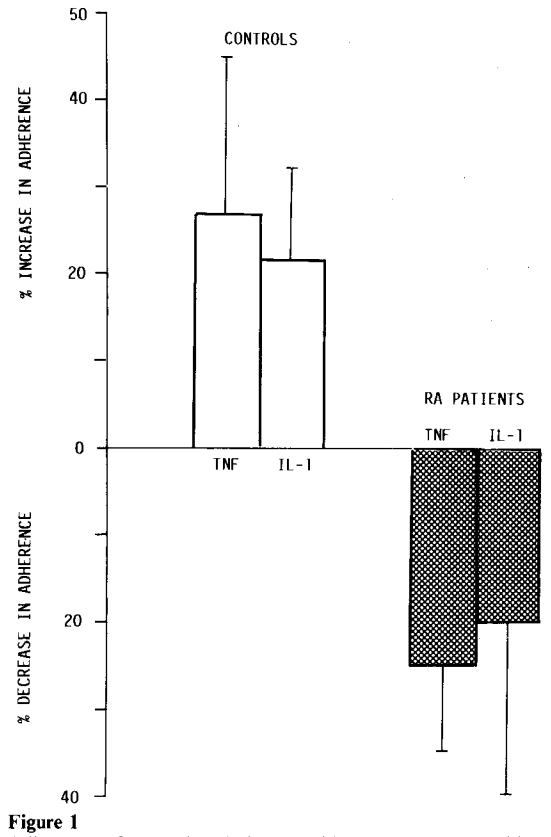

Figure 1
Adherence of control and rheumatoid monocytes to cytokine-treated endothelial monolayers. Results are the mean of 15 experiments. The adherence of control monocytes to endothelium pretreated with IL-1 or TNF was increased, whereas the attachment of rheumatoid monocytes to cytokine-treated monolayers was inhibited.

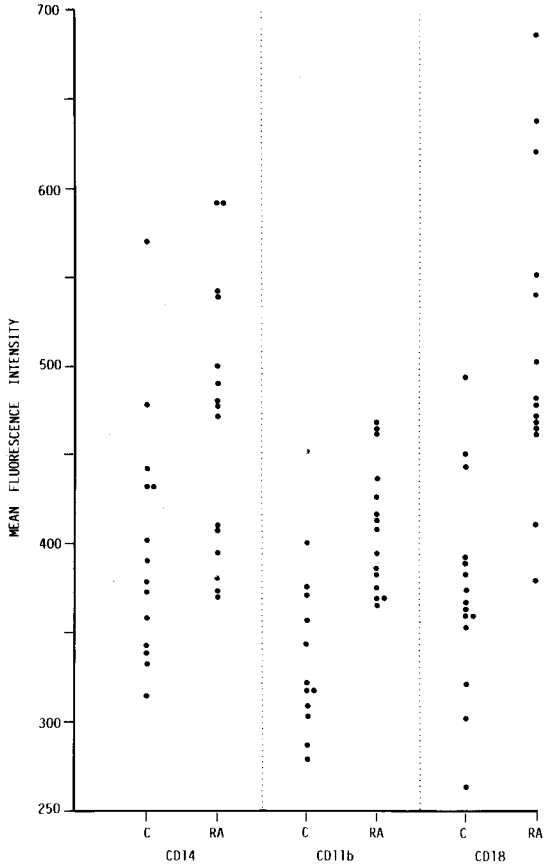

Figure 2
Expression of CD14, CD11b and CD18 is enhanced on the surface of rheumatoid monocytes. Each symbol represents the mean fluorescence intensity of monocytes from one RA or control subject.

pared with controls (Fig. 2). The expression of CD14 was enhanced also on RA monocytes.
When blood monocytes from control subjects were incubated with LPS, their binding to untreated endothelial monolayers was augmented (mean 23% increase; $p < 0.05$). In contrast, the addition of LPS-stimulated monocytes to endothelial treated with IL-1 produced a 27% inhibition of adhesion.

Discussion

This study shows that rheumatoid monocytes are more adherent than control monocytes to untreated monolayers of endothelial cells but less adherent to monolayers pretreated with either TNF or IL-1. Circulating monocytes in RA are believed to be activated [5, 6] and in this context it is of interest that LPS-stimulated monocytes exhibit an enhanced adhesion to untreated endothelium and an impaired binding to IL-1-treated endothelium. Monocyte attachment to endothelium is partially CD18-dependent [7] and the augmented binding of rheumatoid monocytes to untreated endothelium may be related to the upregulation of CD11b/CD18 on their surface. Adhesion molecules other than CD11/CD18 are likely to regulate monocyte–endothelial cell interaction and the impaired adherence of rheumatoid monocytes to TNF- and IL-1-treated endothelium could be due to the down-regulation of such determinants following monocyte activation.
We propose that the distribution of macrophages in distinct areas of rheumatoid synovium [8] could arise from the selective binding of blood monocytes to unstimulated and cytokine-activated endothelium of the local microvasculature.

Acknowledgement

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