Lipopopolysaccharide induces functional ICAM-1 expression in rat alveolar epithelial cells in vitro

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Lipopopolysaccharide (LPS)-induced lung inflammation is known to increase pulmonary intercellular adhesion molecule-1 (ICAM-1) expression. In the present study, L2 cells, a cell line of alveolar epithelial cells, were stimulated with LPS, and ICAM-1 expression was studied. ICAM-1 protein on L2 cells peaked at 6 (38% increase; P < 0.01) and 10 (48% increase; P < 0.001) h after stimulation with Escherichia coli and Pseudomonas aeruginosa LPS, respectively. ICAM-1 mRNA expression was markedly increased, with a peak at 2–4 (E. coli) and 4–6 (P. aeruginosa) h. Adherence assays of neutrophils to LPS-stimulated L2 cells showed a threefold increase in adherence (P < 0.001). Pretreatment of the neutrophils with anti-lymphocyte function-associated antigen-1 and anti-Mac-1 antibodies reduced adherence by 54% (P < 0.001). Analysis of immunofluorescence staining for ICAM-1 showed an exclusive apical expression of ICAM-1. These results indicate that LPS upregulates functional active ICAM-1 on the apical part of the membrane in rat pneumocytes.

intercellular adhesion molecule-1; alveolar epithelium; endotoxin; neutrophil adherence

INTERCELLULAR ADHESION MOLECULE-1 (ICAM-1; CD54) is a cell surface glycoprotein of 76–114 kDa and a member of the immunoglobulin supergene family. Its structure contains a cytoplasmic tail, a transmembrane region, and five extracellular domains binding to counterreceptors on leukocytes (28, 32). ICAM-1 was described for the first time in 1986 (27), and its proposed function as a ligand for lymphocyte function-associated antigen-1 (LFA-1; CD11b/CD18) was confirmed in further studies (9, 20, 21). The second ligand for ICAM-1 is Mac-1 (CD11a/CD18), another member of the β2 subfamily of integrins. This group of antigens is localized on leukocytes, also known as leukocyte cell adhesion molecules. Interactions between ICAM-1 expressed on epithelial and endothelial cells and the β2-integrins play an important role in a wide variety of immune mechanisms. Most current among them are cell-cell contact-mediated immune mechanisms (3, 33) and leukocyte-endothelial and leukocyte-epithelial adhesion processes. Adhesion to the vascular endothelium mediated through ICAM-1-integrin interactions reveals a key role in emigration of white blood cells to sites of inflammation (30, 31, 38). The relevance of leukocyte-epithelium adhesion provided by ICAM-1 has not been clearly defined yet.

In lung tissue, ICAM-1 is found on both type I and type II alveolar epithelial cells. The majority of alveolar epithelial cells are made of thin, squamous type I cells, whereas surfactant-producing type II cells have a cuboidal shape and cover just a small part of the alveolar surface. These distinct cell types have not only been shown to be different in their basic expression but also in the upregulation of ICAM-1 depending on culture conditions (4, 5, 26). Previous in vitro studies have investigated the effects of different stimuli on expression and upregulation of ICAM-1 on type II cells in vitro. Stimulation with tumor necrosis factor (TNF-α) and interferon (IFN)-γ (1) and the effect of mechanical factors (14) as well as direct exposure of type II cells to pathogens (40) inducing ICAM-1 upregulation have already been studied. In a large number of inflammatory and antigen-induced models of lung diseases such as asthma (22), reperfusion-induced lung injury (16), lung damage caused by immunocomplexes (23), hyperoxia (17, 25), and endotoxin and cytokine exposure (2, 18, 35), a possible relevance of upregulated epithelial ICAM-1 could be shown. However, its functional significance still remains to be determined. In addition, in acute lung injury and acute respiratory distress syndrome, ICAM-1 probably plays a crucial role, although the great complexity of these immunologic processes is incompletely understood. In sepiic acute respiratory distress syndrome, interaction between pathogens and macrophages or leukocytes results in release of cytokines (19, 33, 39), causing lung damage. This process is believed to be mediated through proteases and oxidative metabolites of neutrophils that adhere to the pulmonary endothelium (36). The role of adhesion processes to alveolar epithelium in the pathogenesis of acute lung injury, namely the role of ICAM-1 in this context, still has to be defined. However, it has been previously shown (29) that the adherence of neutrophils might promote epithelial cell killing in vitro, although an oxygen metabolite-independent manner was concluded. In vivo, enhanced expression of lung
ICAM-1 is presumed to be the result of direct inflammatory stimuli (e.g., endotoxins) in concert with secondary effects (i.e., cytokines and chemokines) mediated by inflammatory cells (33). In this study, the direct effects of different endotoxins on ICAM-1 expression in a type II alveolar epithelial cell line are evaluated.

MATERIALS AND METHODS

L2 cells. L2 cells (American Type Culture Collection, Manassas, VA), representing a donally isolated cell line from the adult rat lung, are of alveolar epithelial cell type II origin (8). These cells were cultured in Dulbecco's modified Eagle's medium (DMEM; Life Technologies, Basel, Switzerland) supplemented with 10% fetal bovine serum (FBS), 1% penicillin-streptomycin, and 1% HEPES buffer as the culture medium. Passages 35–47 were used for the following experiments.

Stimulation with lipopolysaccharide. Confluent monolayers of L2 cells were exposed to Escherichia coli serotype 055:B5 lipopolysaccharide (LPS; 100 μg/ml; Sigma, Buchs, Switzerland) or Pseudomonas aeruginosa serotype 10 LPS (100 μg/ml; Sigma) diluted in DMEM-1% FBS. At intervals of 2 h, the medium of selected wells was replaced with 100 μl/well (96-well plates) or 2 ml/well (6-well plates) of the LPS solution. Stimulation was completed after 12 h. Control wells and wells that were reserved for background values were simply filled with DMEM-1% FBS. The following cell-based enzyme-linked immunosorbent assays (ELISA) were made the same day.

RNA extraction and Northern blot analysis. Total cellular RNA was isolated from confluent monolayers of stimulated L2 cells. Following manufacturer's instructions, the cells were lysed by directly adding a monophasic solution of phenol and guanidine isothiocyanate (TRIzol Reagent, Life Technologies) to each well. RNA was precipitated with chloroform and, finally dissolved in RNase-free water. Total amounts of cellular RNA were determined by absorbance at 260 nm. Four micrograms of RNA were then loaded on a 1.5% agarose gel, electrophoresed, and blotted onto a nylon transfer membrane (GeneScreen, New England Nuclear). After hybridization, the blots were washed two times for 20 min in 0.1% SDS at 65°C, the membranes were hybridized overnight at 65°C with rat-specific 32P-labeled cDNA for ICAM-1, which was previously produced through a random primer DNA labeling system (Life Technologies) with [γ-32P]ATP and rat ICAM-1 DNA (a gift from Dr. P. A. Ward, Department of Pathology, University of Michigan, Ann Arbor, MI). After hybridization, the blots were washed two times for 20 min in wash solution I (1 mM EDTA, 40 mM Na2HPO4, pH 7.2, and 5% sodium dodecyl sulfate (SDS)) for 4 h at 65°C, the membranes were hybridized overnight at 65°C with rat-specific 32P-labeled cDNA for ICAM-1, which was previously produced through a random primer DNA labeling system (Life Technologies) with [γ-32P]ATP and rat ICAM-1 DNA (a gift from Dr. P. A. Ward, Department of Pathology, University of Michigan, Ann Arbor, MI). After hybridization, the blots were washed two times for 20 min in wash solution I (1 mM EDTA, 40 mM Na2HPO4, pH 7.2, and 5% SDS) at 65°C and two times for 20 min in wash solution II (1 mM EDTA, 40 mM Na2HPO4, pH 7.2, and 1% SDS) at 65°C. The membranes were exposed to X-OMAT-AR film (KODAK) at −70°C. Changes in ICAM-1 mRNA expression were verified by densitometric analysis.

Direct cellular ELISA for ICAM-1. L2 cells were plated at concentrations of 7,500 cells/well in DMEM-10% FBS on tissue culture-treated 96-well microtiter plates (NUNC Products, Life Technologies) and incubated (37°C, 5% CO2) for 5 days to form confluent monolayers. After stimulation with LPS as described in Stimulation with lipopolysaccharide, the cells were fixed with PBS-3% paraformaldehyde for 5 min at room temperature. After each incubation step, the cells were washed three times with 1× PBS (200 μl/well). Incubation for 1 h at 4°C with the primary antibody (except the wells that were selected to determine the background) was performed with monoclonal mouse anti-rat ICAM-1 antibody (1A29; Serotec, Oxford, UK) diluted in PBS-5% FBS to a concentration of 0.5 μg/ml. After a washing step, the wells were incubated with the secondary goat anti-mouse IgG conjugated with horseradish peroxidase (1:2,500 in PBS-5% FBS; 1 h at 4°C). The washing procedure was repeated, 200 μl of 0-phenylenediamine dihydrochloride solution (Sigma) were added to each well, and the microtiter plates were incubated for 5 min at room temperature. Finally, the reaction was stopped with 3 M H2SO4. The plates were read at 492 nm on a microtiter plate reader (BioConcept, Allschwil, Switzerland). The optical density values were interpreted as upregulation of ICAM-1 in comparison to that in the control wells after subtraction of the background value.

Immunofluorescence staining for ICAM-1. L2 cells were cultured in Lab-Tek chamber slides (NUNC Products, Life Technologies) at 30,000 cells/well for 4 days to form confluent monolayers. For immunofluorescence staining, the cells were fixed at room temperature for 15 min with PBS-3% paraformaldehyde-0.1% saponin (300 μl/well). The cells were washed three times with PBS-0.1% saponin and incubated at 4°C for 45 min with 5 μg/ml of 1A29 antibody in PBS-5% FBS-0.1% saponin. The cells were washed again, and 100 μl of Cy3-goat anti-mouse IgG conjugate (1.5 μg/ml in PBS-5% FBS-0.1% saponin; Jackson ImmunoResearch Laboratories, West Grove, PA) and fluorescein-phalloidin (Cytoskeleton, Denver, CO) were simultaneously added to each well. To show unspecific binding of the secondary antibody, incubation with the primary antibody was omitted in selected wells. After an incubation for 45 min at 37°C, the cells were washed again and finally embedded in Vectashield Gel (Reactolab, Servion, Switzerland). Immunofluorescence staining was examined with confocal laser scanning microscopy (Zeiss LSM3, JenLab, Germany). The staining was processed with an image analysis program to enhance the contrast between ICAM-1 monolayers. This resulted in a change of colors, with ICAM-1 staining appearing green and actin staining appearing red.

Isolation of neutrophils. Venous blood was obtained from healthy adult volunteers and anticoagulated with citrate-dextrose solution (Sigma). Neutrophils were isolated by gradient centrifugation over Ficoll-Paque (Amersharm Pharmacia Biotech, Dübendorf, Switzerland) followed by 1% dextran sedimentation for 1 h to separate neutrophils from erythrocytes. After centrifugation of the supernatant, the contaminating erythrocytes were lysed with distilled water followed by the addition of 2.7% NaCl to stop hypotonic lysis. The remaining neutrophils were resuspended in Hank's balanced salt solution without calcium and magnesium (HBSS; Life Technologies). The entire procedure was performed at room temperature.

Assay of neutrophil adherence to L2 monolayers. L2 cells were plated in DMEM-10% FBS on tissue culture-treated 96-well microtiter plates (NUNC Products, Life Technologies) and grown to confluency at 37°C with 5% CO2. After stimulation with E. coli serotype 055:B5 LPS (100 μg/ml) in DMEM-1% FBS for 2, 4, 6, and 24 h, the cells were washed with DMEM. One hundred thousand neutrophils were added to each well, and the microtiter plates were incubated for 30 min at 37°C with 5% CO2. The nonadherent cells were removed by carefully washing the cells two times with DMEM. Finally, the number of adherent neutrophils was...
determined by counting them in a defined area. In this experiment, the neutrophils were preincubated at 37°C for 15 min with both the monoclonal mouse anti-human LFA-1/CD11a and Mac-1/CD11b antibodies (10 µg/ml; Becton Dickinson, Basel, Switzerland). Neutrophils that were used for positive control values were preincubated with the nonspecific binding monoclonal mouse anti-human CD40 antibody (10 µg/ml; Becton Dickinson). Before being added to the L2 monolayers, the neutrophils were washed once with HBSS. To evaluate the single effect of each antibody, L2 cells were stimulated for 4 h with E. coli LPS and the same procedure was performed but with each of the two antibodies alone and together.

For ICAM-1-blocking studies on L2 cells, after 4 h of stimulation, the cells were preincubated with 10 µg/ml of anti-ICAM-1 antibody (1A29) in DMEM for 30 min at 37°C with 5% CO2. At the same time, the neutrophils were preincubated with anti-FcyRIII (CD16) and anti-FcyRII (CD32) antibodies (Becton Dickinson) at a concentration of 10 µg/ml for each antibody in DMEM for 15 min at 37°C. The L2 cells and neutrophils were washed once with HBSS, and the neutrophils were incubated with L2 cells as described above.

Statistical analysis. The results are expressed as means ± SE after subtraction of negative control values. Significances were calculated by comparing individual group means with Student’s t-test.

RESULTS

Effects of LPS on mRNA for ICAM-1. To determine whether upregulation of ICAM-1 protein reflects mRNA for ICAM-1 expression, Northern blot analysis was performed. L2 cells were stimulated with E. coli and P. aeruginosa LPS (100 µg/ml) for various periods of time, and total RNA was extracted. Figure 1A shows a representative Northern blot demonstrating ICAM-1 expression after stimulation for 2, 4, 6, 8, 10, and 12 h with E. coli LPS. Unstimulated cells showed almost no constitutive mRNA for ICAM-1. Stimulation with E. coli LPS revealed an induction of ICAM-1 mRNA in L2 cells, with a peak at 2–4 h (7.4-fold increase) as shown by densitometry. Stimulation of L2 cells with P. aeruginosa LPS for 2, 4, 6, 8, 10, and 12 h upregulated ICAM-1 (Fig. 1B). But in contrast to stimulation with E. coli LPS, ICAM-1 mRNA peaked at 4–6 h (7.2-fold increase), with a continuous decrease thereafter.

Effects of LPS on L2 cell ICAM-1 protein expression. ICAM-1 expression in L2 cells was measured by cell-based ELISA after exposure to E. coli or P. aeruginosa LPS. ICAM-1 upregulation in response to E. coli LPS increased ICAM-1 by 31% after 4 h (P < 0.01) and by a

Fig. 1. Time course of intercellular adhesion molecule-1 (ICAM-1) mRNA level after stimulation with Escherichia coli (serotype 055:B5; A) and Pseudomonas aeruginosa (serotype 10; B) lipopolysaccharide (LPS; 100 µg/ml) in confluent monolayers of L2 cells. Total cellular mRNA was extracted, and ICAM-1 mRNA was detected by Northern blot analysis (A and B, top). Four micrograms of total cellular mRNA were loaded in each lane. ICAM-1 mRNA peaked at 2–4 (E. coli LPS) and 4–6 (P. aeruginosa LPS) h. No constitutive mRNA expression was detectable. Below each representative blot, methylene blue staining of 18S and 28S rRNA bands shows equal loading. To verify upregulation of mRNA for ICAM-1, densitometry was performed, showing maximal expression after E. coli stimulation at 2 h (a 7.4-fold increase; A, bottom) and after P. aeruginosa LPS at 4 h (a 7.2-fold increase; B, bottom).
maximum of 38% after 6 h (P < 0.01), which persisted (Fig. 2A). After 2 h, however, there was no significant change in ICAM-1 expression detectable (increase of 2%; P > 0.05). Interestingly, with P. aeruginosa LPS, the increase in ICAM-1 expression after 2 h of stimulation was not significant (8%; P > 0.05); after 4, 6, 8, 10, and 12 h, increases of 20 (P < 0.01), 35, 40, 48, and 46%, respectively (all P < 0.001), were seen compared with those in unstimulated control cells (Fig. 2B). In contrast to stimulation with E. coli LPS, the peak of ICAM-1 upregulation in response to P. aeruginosa LPS occurred at 10 h.

Localization of ICAM-1 protein on L2 cells. To obtain more information about the biological function of membrane-bound ICAM-1, we analyzed immunofluorescence staining of L2 cells using a confocal laser scanning microscope. Using this system, we were enabled to examine both the apical and basolateral parts of the cell. Costaining for ICAM-1 (Fig. 3B) as well as for actin (Fig. 3A) was used for assessment of the thickness of the cells. To ensure that ICAM-1-detecting antibodies did not miss possible intracellular pools of ICAM-1 or ICAM-1 expressed at the bottom of the cell, the cells were permeabilized with saponin. In Fig. 3C, the frontal view of a representative cell simultaneously shows a green fluorescence signal representing ICAM-1 and a red color representing actin-bound fluorescence-labeled phalloidin. The side section (Fig. 3D) clearly demonstrates that ICAM-1 is expressed on the apical part of the membrane of the L2 cells (a). In contrast, no ICAM-1 expression was seen on the basolateral part of the membrane (Fig. 3D, b and c) nor were there any intracellular pools of ICAM-1 detectable. No unspecific binding of the secondary antibody was found (data not shown).

Adherence of neutrophils to LPS-stimulated L2 monolayers. L2 cells were exposed to E. coli LPS for 2, 4, 6, and 24 h. After the monolayers were washed, neutrophils were added and incubated for 15 min. Nonadherent neutrophils were washed away. An increase in adherence of 69% 2 h after stimulation was detected on stimulated cells (P < 0.001). Adherence was greatest at 4 and 6 h, with a threefold increase (P < 0.001). To determine to what extent interaction between ICAM-1 and its counterreceptors LFA-1/CD11a and Mac-1/CD11b were responsible for adherence, the neutrophils were simultaneously preincubated with anti-LFA-1 and anti-Mac-1 antibodies. These blocking antibodies together significantly reduced adherence at 0, 2, 4, 6, and 24 h by 40, 61, 54, 49, and 38%, respectively (all P < 0.001; Fig. 4A). Blocking with anti-LFA-1 or anti-Mac-1 antibody alone after 4 h of LPS stimulation decreased adherence by 24 (P < 0.01) and 33% (P < 0.01), respectively, with the assumption that both antibodies together have an additive effect (Fig. 4B). After 4 h of stimulation, ICAM-1 was also blocked directly on L2 cells before the neutrophils were added, which showed a diminished adherence of 49% (P < 0.001; Fig. 4C). These results suggest that the interaction between ICAM-1 and LFA-1 and/or Mac-1 is partly involved in the adherence of neutrophils to pneumocytes.

**DISCUSSION**

This study demonstrates that LPS upregulates ICAM-1 expression on L2 cells. Compared with mRNA levels for ICAM-1, ICAM-1 protein expression peaked with a 2- to 4- (E. coli LPS) to 4- to 6- (P. aeruginosa LPS) h delay, suggesting de novo synthesis of ICAM-1 as described in similar studies on ICAM-1 upregulation on alveolar epithelial cells (1, 9, 22). These data were reproducible with two different endotoxins, although different dynamics of ICAM-1 upregulation were seen. Performing these studies with primary cultures of rat alveolar epithelial cells showed the same upregulation (data not shown). The analysis of the ultrastructural localization of ICAM-1 on L2 cells showed ICAM-1 to be expressed only on the apical part of the cell membrane.
Increased adherence of neutrophils to LPS-stimulated L2 cells being partially blocked by specific anti-ICAM-1 antibodies emphasizes a possible biological function of apically expressed ICAM-1.

Upregulation of ICAM-1 protein expression was only 40% (E. coli) and 50% (P. aeruginosa) compared with the baseline expression of ICAM-1. An explanation for the comparable modest ICAM-1 upregulation could be found in the pathophysiological processes that occur in lung inflammation. Endotoxin-induced lung inflammation is the result of a very complex cascade of synergistic processes leading to poorly understood interactions between different cell types, mainly mediated by cytokines such as TNF-\(\alpha\), interleukin-1, and interleukin-8 (19, 33, 39). In an in vitro study (34), ICAM-1 expression on L2 cells previously stimulated with a mixture of LPS, TNF-\(\alpha\) and IFN-\(\gamma\) was extremely upregulated. These three components in combination could be considered as an approach to inflammatory processes in vivo as described above. Consequently, it is obvious that the direct stimulation of alveolar epithelial cells by LPS in vitro represents a part of the whole process increasing ICAM-1 on these cells in vivo. This might be the explanation for the modest increase in ICAM-1 in vitro compared with that in vivo (4).

The exact molecular mechanism of LPS-induced injury to epithelial or endothelial cells has not been clearly defined yet. Evidence has grown that a soluble form of CD14, a macrophage and granulocyte membrane protein, forms complexes, with LPS binding to specific signal-transducing receptors on epithelial or endothelial cells (13, 26, 37). In contrast, myeloid cells express CD14 as a receptor for complexes of LPS and LPS binding protein. Nevertheless, other studies (10, 12) indicated that CD14 is expressed not only by myeloid cells but also by epithelial cells. Moreover, CD14 mRNA in mouse lung tissue has been shown to be upregulated by LPS and TNF-\(\alpha\) in vivo, although regulation of CD14 gene expression differs in epithelial and myeloid cells (11). To determine whether L2 type II alveolar epithelial cells express CD14 mRNA, which may be upregulated by LPS or cytokines like TNF-\(\alpha\) or IFN-\(\gamma\), RT-PCR was performed. The results clearly showed that L2 cells express constitutive high levels of CD14 mRNA that are not influenced by stimulation with LPS, TNF-\(\alpha\), or IFN-\(\gamma\) for 6 h (data not shown). Accordingly, increased levels of mouse lung CD14 mRNA after systemic exposure to LPS in vivo (10) seem to have cellular sources other than type II alveolar epithelial cells.

LPS consists of three components: lipid A, core polysaccharide, and the O antigen. Because virtually all LPS-induced immune responses are lipid A dependent, differences in molecular structure (although highly conserved among gram-negative bacteria) or different three-dimensional conformations of lipid A or LPS in solution might be responsible for the different effects they exert on L2 cells concerning ICAM-1 upregulation (37). Consequently, different binding interactions between lipid A or LPS and soluble CD14 could lead to a different three-dimensional structure presented to the hypothesized complex-binding receptor on the cells, resulting in a different signal transduction. Further studies that focus on the molecular basis of LPS-induced activation of epithelial or endothelial cells will be necessary to elucidate this problem.
Because previous studies (6, 24) have raised the question about the biological role of ICAM-1 expression on alveolar epithelial cells, we determined the exact ultrastructural localization of ICAM-1 on L2 cells to get more insight into its biological function. Interestingly, ICAM-1 was found to be exclusively expressed on the apical part of the cell membrane. These results are supported by previous immunohistological examinations of mouse lungs where ICAM-1 was found only on the luminal surface of alveolar epithelial cells (4, 18).

Guzman et al. (15) showed ICAM-1 to be expressed just on one side of the surface of freshly isolated human type II cells. These results suggest that even in nonconfluent type II cells, independent of cell-cell contact, there is a cell surface area where ICAM-1 is exclusively expressed, although possible species-specific differences have to be considered. On endothelial cells, the apical expression of ICAM-1 is well known to be essential for emigration of white blood cells out of the bloodstream in the direction of the site of inflammation (38). If these principal processes of inflammation are applied to the anatomy of the lung, one would rather expect a more basolateral expression of ICAM-1 on alveolar epithelial cells to enable white blood cells to emigrate from the pulmonary interstitium into the alveolar space. In addition, we showed that enhanced adhesion of neutrophils to LPS-stimulated L2 cells is partly mediated by ICAM-1. Other alveolar epithelial cell adhesion molecules such as vascular cell adhesion molecule-1 (CD106) and LFA-3 (CD58) are probably involved in this process (6, 7). This may be one reason for only partial blocking adhesion of neutrophils to L2 cells by anti-ICAM-1 antibodies.

Summarized, our studies reveal more evidence that ICAM-1 expressed on the apical part of the alveolar epithelial cell membrane may have a different biological function compared with ICAM-1 on the vascular endothelium (1, 23). First, ICAM-1 probably provides adherence of white blood cells to the luminal surface of alveolar epithelial cells in vivo, which is a hypothesis (5, 15, 23) that is supported by our in vitro results. In vivo, this process could serve as a protection mechanism against microbial pathogens present in the alveolar space in the case of lung inflammation. Adhering white blood cells also cause lung injury through release of oxygen radicals as described above. This is supported by Mulligan et al. (23), who demonstrated the protective effect of intratracheally instilled antibodies to ICAM-1 in a model of immune complex-induced lung injury. Second, because ICAM-1 also plays a role in cell-cell contact-mediated immune mechanisms, as for example in B- or T-lymphocyte interaction (3), it may advance the immunoreactivity of white blood cells in the alveolar lumen by interaction with their LFA-1 counterreceptor.

In conclusion, our results indicate that L2 cells or alveolar epithelial cells respond to direct stimulation.
with LPS by upregulation of ICAM-1 expression on the apical part of the cell membrane. This is probably an important immunologic defense mechanism in lung inflammation on one hand but could also contribute to endotoxin-induced lung injury on the other hand. Further studies have to be performed to evaluate the suitability of this concept to develop therapeutic strategies against acute lung injury in vivo.

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