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Synovial cells are potent antigen-presenting cells for superantigen, staphylococcal enterotoxin B (SEB)

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SUMMARY

There is ample evidence suggesting that superantigens may act as a triggering factor in the pathogenesis of rheumatoid arthritis (RA). We investigated whether superantigen could activate T cells in the presence of synovial cells. T cells were cultured with SEB in the presence of interferon-gamma (IFN-γ)-treated synovial cells. T cell proliferation and activation were assessed by ^{3}H-thymidine incorporation and IL-2 production. The expression of HLA class II antigens and adhesion molecules on synovial cells was detected by flow cytometer. In the presence of IFN-γ-treated synovial cells, T cells proliferated vigorously and produced IL-2 in response to SEB. A low SEB-induced T cell response was noticed in the presence of untreated synovial cells. Allogeneic as well as autologous IFN-γ-treated synovial cells markedly enhanced SEB-induced T cell proliferation. IFN-γ-treated synovial cells had increased expression of HLA class II antigens and intercellular adhesion molecule-1 (ICAM-1) adhesion molecules. MoAbs towards these antigens markedly inhibited the SEB-induced T cell response. These results indicate that activated synovial cells are potent antigen-presenting cells for SEB to T cells, and that superantigens may play a critical role in the pathogenesis of RA through activated synovial cells.

Keywords superantigen staphylococcal enterotoxin B antigen-presenting cell synovial cell rheumatoid arthritis

INTRODUCTION

Rheumatoid arthritis (RA) is a systemic autoimmune disease characterized by marked inflammation of the synovial membranes of multiple joints [1]. It has been suggested that the autoimmune disease occurs in a genetically susceptible patient following a triggering event such as a bacterial or viral infection. Seropositive RA patients also have certain susceptible genes such as HLA class II, the DR4 and DR1 alleles [2].

Exogenous superantigens are produced by different organisms such as mycoplasma and bacteria, including *Staphylococcus aureus* and *Streptococcus pyogenes* These superantigens can bind directly to HLA class II antigens on antigen-presenting cells (APC) without antigen processing [3,4]. Such a process activates T cells bearing specific Vβ elements of the T cell receptor (TCR). There is a growing body of evidence suggesting that superantigens may play an important role in the autoimmune response. Experimentally induced infection of mice with mycoplasma arthritides results in chronic joint inflammation [5]. In addition, human streptococcal infections may be associated with arthropathy and/or a pronounced autoimmune response [6]. Paliard *et al.* [7] reported that the number of Vβ14$^+$ T cells is significantly higher in the synovial fluid of affected joints compared with peripheral blood. These findings suggest that superantigens may be implicated in the pathogenesis of RA.

The present study was undertaken to investigate the role of synovial cells in the activation of T cells in the presence of a superantigen.

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MATERIALS AND METHODS

Reagents

SEB was purchased from Sigma Chemical Co. (St Louis, MO). IFN-γ was a generous gift from Shionogi Co. (Tokyo, Japan).

Cells

Synovial cells used in this study were obtained from four patients with RA and one patient with osteoarthritis (OA) undergoing total knee replacement due to joint deterioration. RA patients were diagnosed as suffering from either definite or classical RA fulfilling the revised American Rheumatism Association criteria for RA [8]. The synovial tissue was cut with scissors into small pieces and incubated for 15 min at 37°C

in RPMI 1640 medium containing 2 mM L-glutamine, 50 U/ml penicillin, and 50 μg/ml streptomycin, and treated with 100 μg/ml collagenase (Sigma) and 3·3 mg/ml dispase (Godo Shunsei Co., Tokyo, Japan). After washing, cells were cultured with RPMI 1640 medium supplemented with 10% heat-inactivated fetal calf serum (FCS; GIBCO, Gaithersburg, MD). Synovial cells from the third to sixth passage which resembled the previously reported 'fibroblast-like' synovial cells, were used in the following experiments. Cells were later washed three times with PBS and treated with 50 μg/ml mitomycin C (MMC) (Kyowa, Tokyo, Japan) for 30 min at 37°C.

Peripheral blood mononuclear cells (PBMC) were isolated from heparinized blood by Ficoll–Conrey density gradient centrifugation (Daiichi Pharmaceutical Co., Tokyo, Japan). Adherent MNC were obtained using the plastic plate adherent method, and defined as monocytes. T cells were separated from non-adherent MNC by E-rosette formation with neuraminidase-treated sheep erythrocytes (Nippon Bio-Test Laboratories Inc., Tokyo, Japan) and then passed through a nylon wool column (Wako Pure Chemical Industries Ltd., Osaka, Japan). The purity of T cells separated from PBMC by these methods was >97·8%.

Monoclonal antibodies

Purified MoAbs to human HLA-DR, HLA-DQ, intercellular adhesion molecule-1 (ICAM-1) and vascular cell adhesion molecule-1 (VCAM-1) were purchased from Immunotech S.A. (Marseille, France).

Assay of proliferative responses

Purified T cells (1×10^5) and MMC-treated synovial cells (1×10^4) were cultured with different concentrations of SEB in triplicates of 0·2 ml volumes in flat-bottomed 96-well microplates (Costar 3595, Cambridge, MA) for 4 days. Cultures were pulsed with 0·2 μCi of ^{3}H-thymidine/well for the last 16 h of the culture period. Each sample was harvested and radioactivity determined with a liquid scintillation counter. Data are presented as an average of triplicate values.

Antibodies on inhibitory effect of anti-HLA-DR, anti-HLA-DQ, anti-ICAM-1, and anti-VCAM-1 SEB-induced proliferative responses of T cells

Purified T cells (1×10^5) were stimulated with 0·1 μg/ml SEB in the presence of IFN-γ-treated synovial cells. MoAbs to HLA-DR, HLA-DQ, ICAM-1 and VCAM-1 were added 30 min before initiation of culture at a concentration of 10 μg/ml.

Assay for IL-2 production

T cells (1×10^6) were cultured with 1×10^5 MMC-treated synovial cells in 2-ml volumes in 24-well culture plates (Costar 3524) and stimulated with SEB for 24 h. IL-2 concentration was measured in collected culture supernatants using an ELISA IL-2 assay kit from Otsuka Co. (Tokyo, Japan). Briefly, plates precoated with MoAb to IL-2 were incubated with samples, followed by incubation with polyclonal rabbit anti-IL-2 antibody, and later reacted with goat anti-rabbit immunoglobulin to horseradish peroxidase. Excess reactants were removed between each step by a triplicate washing with 0·01 M phosphate buffer pH 7·4 containing 1% bovine serum albumin (BSA). The addition of enzyme substrate produced a chromogenic product with absorbance of 490 nm. The limitation of the sensitivity was 50 pg/ml.

Flow cytometric analysis

A two-step surface staining was performed by incubating 1×10^6 cells with 5 μl of the appropriate MoAb for 30 min at 4°C. This was followed by washing and a further incubation with FITC-conjugated goat anti-mouse IgG (Coulter Immunology, Hialeah, FL) for 30 min at 4°C. Stained cells were counted with an EPICS PROFILE II flow cytometer (Coulter).

Statistical analysis

Statistical analysis of the results was performed using Student's *t*-test for unpaired samples.

RESULTS

Proliferative response of PBMC by SEB

In preliminary experiments we determined the number of PBMC stimulated with SEB (1·0 μg/ml). The proliferative response stimulated by SEB was maximal when 1×10^5/ml of PBMC were used. The response reached a peak on day 4 of culture, and the optimal dose of SEB was 1·0–10·0 μg/ml. Incorporation of ^{3}H-thymidine into PBMC in the absence of SEB was 154 ± 17 ct/min, and 6086 ± 539 ct/min in the presence of 1·0 μg/ml SEB.

Effect of the presence of synovial cells on SEB-induced T cell proliferation

Purified T cells and autologous synovial cells did not respond to SEB in the absence of monocytes. When 1×10^5 T cells were cultured with 1·0 μg/ml SEB, in the presence of 1×10^4 synovial cells pretreated with 1000 U/ml IFN-γ, ^{3}H-thymidine incorporation of T cells progressively and markedly increased, reaching a peak on day 4 of culture (Fig. 1a). In addition, SEB-induced T cell proliferation was also detected in the presence of untreated synovial cells, but it was distinctly

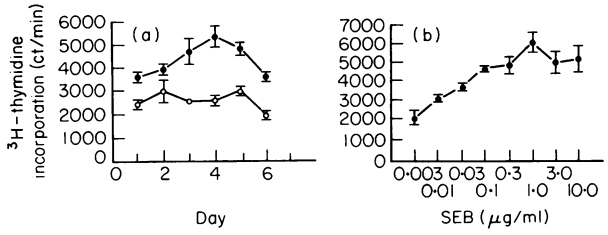

Fig. 1. SEB-induced T cell proliferation in the presence of untreated or IFN-γ-treated synovial cells. (a) Time course: purified T cells (1×10^5 cells/well) were cultured with 1·0 μg/ml SEB in the presence of untreated (○) or IFN-γ-treated (●) synovial cells (1×10^4 cells/well) for 1–6 days. The proliferative response of T cells to SEB is expressed by ^{3}H-thymidine incorporation. Results are mean ± s.d. of three separate experiments. (b)Effect of SEB concentration: T cells (1×10^5) were cultured with various concentrations of SEB in the presence of 1×10^4 IFN-γ-treated synovial cells. Cultures were harvested on day 4 after 6-h pulse of ^{3}H-thymidine. Proliferative responses are expressed as Δct/min of ^{3}H-thymidine incorporation. Results are mean ± s.d. of three separate experiments.

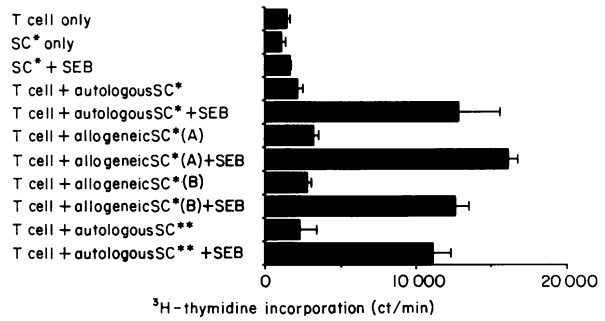

Fig. 2. SEB-induced T cell proliferation in the presence of autologous or allogeneic synovial cells. Synovial cells were obtained from three patients (autologous synovial cells, allogeneic synovial cells from A and B) with rheumatoid arthritis (RA) and one patient with osteoarthritis (OA). Purified T cells (1×10^5 cells/well) were cultured with 1 μg/ml SEB in the presence of synovial cells pretreated with or without IFN-γ for 4 days. The proliferative response of T cells is expressed as Δct/min of ^{3}H-thymidine incorporation. Results are mean $\pm$ s.d. from three separate experiments. There was no significant difference among autologous synovial cells and allogeneic synovial cells (A or B) from patients with RA and synovial cells from patient with OA. SC*, IFN-γ-treated synovial cells from patients with RA; SC**, IFN-γ-treated synovial cells from patients with OA.

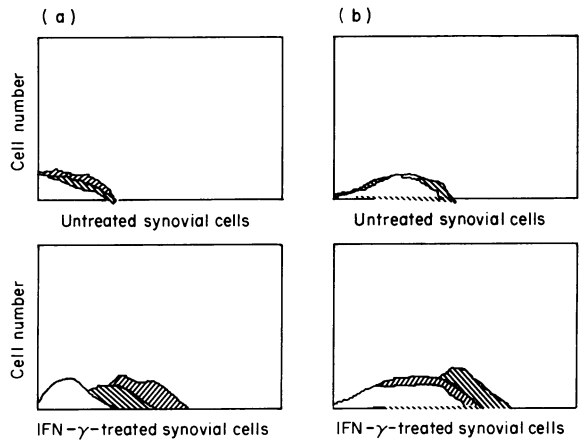

Fig. 3. Flow cytometric analysis of HLA-DR, HLA-DQ, intercellular adhesion molecule-1 (ICAM-1) and vascular cell adhesion molecule-1 (VCAM-1) expression on the untreated or IFN-γ-treated synovial cells. Synovial cells (1×10^6/well) were cultured with or without IFN-γ (1000 U/ml) for 3 days. The abscissa represents a logarithmic scale of the relative fluorescence intensity. (a) The expression of HLA-DR and HLA-DQ antigens on synovial cells.The expression of HLA-DR (▨) and HLA-DQ antigens (▧) was determined by an immunofluorescence staining on a flow cytometer. Mean fluorescence intensity (MFI) of untreated synovial cells stained with control, anti-HLA-DR, and anti-HLA-DQ antibody was 0·5, 1·3, and 0·8, respectively. MFI of IFN-γ-treated synovial cells stained with control, anti-HLA-DR, and anti-HLA-DQ antibody was 2·5, 44·3, and 25·3, respectively. (b) Expression of ICAM-1 and VCAM-1 adhesion molecules on the synovial cells. MFI of untreated synovial cells stained with control, anti-ICAM-1 (▧), and anti-VCAM-1 (▨) antibody was 16·1, 25·7, and 19·5, respectively. MFI of IFN-γ-treated synovial cells stained with control, anti-ICAM-1, and anti-VCAM-1 antibody was 19·6, 55·7, and 17·2, respectively.

lower than that measured in the presence of IFN-γ-treated synovial cells. Culture of T cells with various concentrations of SEB in the presence of IFN-γ-treated synovial cells for 4 days resulted in augmentation of T cell proliferation in a dose-dependent manner, reaching a peak at 1·0 μg/ml of SEB (Fig. 1b). Finally, synovial cells were pretreated with different concentrations of IFN-γ (0–10 000 U/ml), followed by the addition of untreated or IFN-γ-treated cells into the culture of T cells and SEB. SEB-induced T cell proliferation increased by IFN-γ in a dose-dependent manner, and reached a plateau at 1000 U/ml of IFN-γ.

SEB-induced T cell proliferation in the presence of autologous or allogeneic synovial cells and in the presence of synovial cells from OA patients

We investigated whether allogeneic as well as autologous synovial cells were capable of inducing T cell proliferation in response to SEB. As shown in Fig. 2, the level of SEB-induced T cell proliferation in the presence of IFN-γ-treated allogeneic or autologous synovial cells was not different. Without SEB, ^{3}H-thymidine incorporation of T cells in the presence of IFN-γ-treated allogeneic or autologous synovial cells was less than 3000 ct/min. These results indicate that the T cell proliferation was induced in response to SEB and the SEB-induced T cell proliferation in the presence of synovial cells is not restricted by HLA antigens. We also investigated SEB-induced T cell proliferation in the presence of synovial cells from OA patients. As shown in Fig. 2, SEB-induced T cell proliferation was also observed in the presence of IFN-γ-treated synovial cells from OA patients.

Expression of HLA-DR, HLA-DQ, ICAM-1 and VCAM-1 antigens on IFN-γ-treated or untreated synovial cells

Synovial cells were cultured in Petri dishes with or without 1000 U/ml IFN-γ for 3 days, and the expression of HLA-DR, HLA-DQ, ICAM-1 or VCAM-1 was detected by an immunofluorescence method. In contrast to the expression of HLA-DR or HLA-DQ on the untreated synovial cells, the mean fluorescence intensities of HLA-DR and HLA-DQ antigens were markedly increased on IFN-γ-treated synovial cells. Furthermore, the expression of HLA-DR antigens was predominant over that of HLA-DQ on IFN-γ-treated synovial cells. The expression of ICAM-1 was also increased on the IFN-γ-treated synovial cells. However, treatment of synovial cells with IFN-γ did not influence the expression of VCAM-1 (Fig. 3b).

Blocking of SEB-induced T cell proliferation by anti-HLA-DR and anti-HLA-DQ antibodies

Addition of anti-HLA-DR MoAb to a co-culture of SEB, T cells and IFN-γ-treated synovial cells significantly inhibited T cell proliferation (Fig. 4). Anti-HLA-DQ antibody had a similar suppressive effect, but the inhibitory effect of anti-HLA-DR antibody was significantly higher than that of anti-HLA-DQ antibody. Addition of both antibodies to the culture produced a synergistic effect and markedly inhibited the T cell proliferation. These findings indicate that SEB-induced T cell proliferation in the presence of IFN-γ-treated synovial cells is necessary for the binding of SEB to HLA class II antigens on synovial cells.

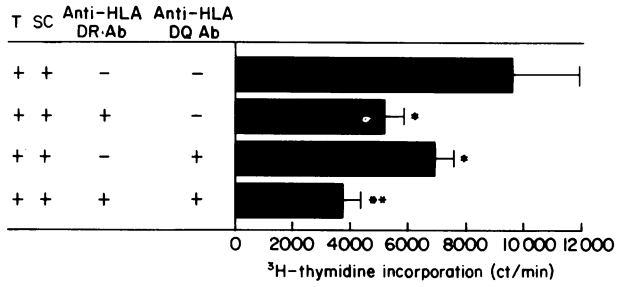

Fig. 4. Blocking of SEB-induced T cell proliferation by anti-HLA-DR and anti-HLA-DQ antibodies. Anti-HLA-DR and/or anti-HLA-DQ antibodies were added to SEB (1·0 μg/ml), T cells (1×10^4 cells/well) and IFN-γ-treated synovial cells (1×10^4 cells/well), and cultured together for 4 days. The proliferative response of T cells to SEB is expressed as Δct/min of ^{3}H-thymidine incorporation. Results are mean ± s.d. of four separate experiments. Asterisks refer to group comparisons using Student's *t*-test. A significant reduction from that without anti-HLA-DR/DQ antibodies is shown as $*P < 0.05$ and $**P < 0.01$.

Effect of anti-ICAM-1 and anti-VCAM-1 MoAbs on SEB-induced T cell proliferation in the presence of IFN-γ-treated synovial cells

The addition of anti-ICAM-1 MoAb to a co-culture of SEB, T cells and IFN-γ-treated synovial cells markedly and significantly blocked T cell proliferation (Fig. 5). On the other hand, anti-VCAM-1 MoAb did not significantly inhibit SEB-induced T cell proliferation. The inhibitory effect of anti-ICAM-1 antibody was greater than that of anti-HLA-DR and anti-HLA-DQ antibody. These results suggest that ICAM-1 expression on IFN-γ-treated synovial cells may play an important role in SEB-induced T cell proliferation.

IL-2 production in SEB-induced T cell proliferation in the presence of IFN-γ-treated synovial cells

We also measured IL-2 in the culture supernatant of T cells and SEB in the presence of untreated or IFN-γ-treated synovial cells. As shown in Fig. 6, the concentration of IL-2 in the supernatant in the presence of untreated synovial cells was similar to that of T cells and SEB in the absence of synovial cells. IL-2 production was significantly inceased in the presence of IFN-γ-treated synovial cells compared with that in the presence of untreated synovial cells. The addition of anti-HLA-DR antibody to the culture in the presence of IFN-γ-treated synovial cells completely inhibited IL-2 production.

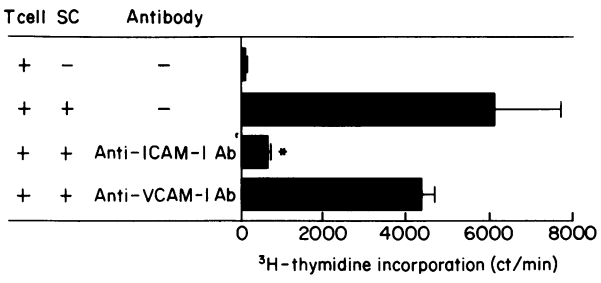

Fig. 5. Effects of anti-intercellular adhesion molecule-1 (ICAM-1) and anti-vascular cell adhesion molecule-1 (VCAM-1) antibodies on SEB-induced T cell proliferation in the presence of IFN-γ-treated synovial cells. Anti-ICAM-1 or anti-VCAM-1 antibody were added to SEB (1·0 μg/ml), T cells (1×10^5 cells/well) and IFN-γ-treated synovial cells (1×10^4 cells/well), and cultured for 4 days. The proliferative response of T cells to SEB is expressed as Δct/min of ^{3}H-thymidine incorporation. Results are mean ± s.d. of three separate experiments. A significant reduction from that without anti-ICAM-1/VCAM-1 antibodies is shown as $*P < 0.05$.

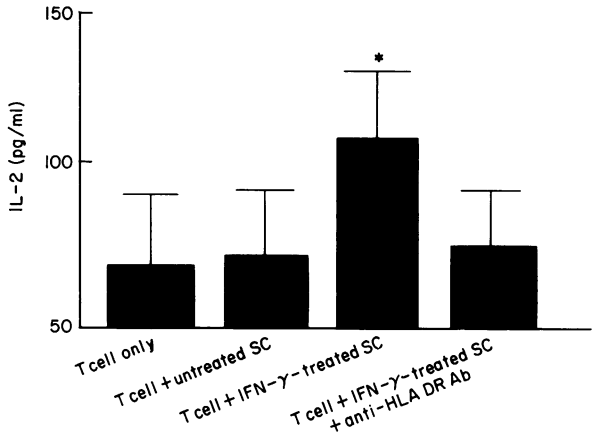

Fig. 6. IL-2 production in SEB-induced T cell proliferation in the presence of IFN-γ-treated synovial cells. T cells were stimulated with SEB (10 μg/ml) in the presence of untreated or IFN-γ-treated synovial cells with or without anti-HLA-DR MoAb for 3 days. The concentration of IL-2 in the culture supernatants was measured by an immunoenzymatic assay, and expressed in pg/ml. Values represent the mean ± s.d. of triplicate determinations. Results represent a summary of three experiments ($*P < 0.05$).

DISCUSSION

The present study demonstrated that T cells in the presence of IFN-γ-treated synovial cells were able to proliferate vigorously and produce IL-2 in response to SEB. In addition, T cell activation was markedly reduced in the presence of untreated synovial cells. Our results also indicated that allogeneic IFN-γ-treated synovial cells served as APC.

Bacterial exotoxins are introduced as exogenous superantigens activating a wide range of T cells bearing particular Vβ families. Mls antigen in mice is also a superantigen recently described to be the mouse retroviral gene product [9–15]. *Staphylococcus aureus* produces most components of exogenous superantigens, such as SEA, SEB, SEC1, SEC2, SEC3, SED, SEE, and toxic shock syndrome toxin 1 (TSST1). *Streptococcus pyogenes* and mycoplasma also produce exogenous superantigens, such as streptococcus pyogenic enterotoxins (SPEs, SPE A and SPE B) and mycoplasma arthritides mitogen (MAM), respectively.

Unlike conventional antigens, superantigens, such as SEs, do not require processing by accessory cells to be presented [3,4]. APC functions for superantigens have thus far been reported for human keratinocytes, dendritic cells, and mononuclear phagocytes [16–18]. However, to our knowledge, antigen presentation of superantigens has never been reported

for synovial cells except by Mourad *et al.* [19]. They showed that staphylococcal enterotoxin A (SEA) bound to synoviocytes and could induce inflammatory cytokine (IL-6 and IL-8) gene expression in these cells. In the present study, we demonstrated that synovial cells could serve as APC for superantigen. In the presence of IFN-γ-treated synovial cells, purified T cells were able to proliferate in response to SEB. In addition, the expression of HLA class II antigens (HLA-DR and HLA-DQ antigens) was markedly increased on the surface of synovial cells after treatment of IFN-γ for 3 days. Our results also demonstrated that anti-HLA-DR and anti-HLA-DQ antibodies blocked SEB-induced T cell activation in the presence of IFN-γ-treated synovial cells. These findings are in accordance with previous reports that superantigens, such as SEB, must bind directly to APC, and do so with high affinity to MHC class II molecules [20,21]. We also demonstrated that the level of SEB-induced T cell proliferation in the presence of allogeneic and autologous IFN-γ-treated synovial cells was the same. These results support the notion that superantigens activate T cell proliferation in an MHC-unrestricted manner [3, 20–22].

The present study also showed that IFN-γ treatment of synovial cells increased the expression of ICAM-1 as well as HLA class II antigens. Anti-ICAM-1 MoAb markedly blocked SEB-induced T cell proliferation with IFN-γ-treated synovial cells, whereas anti-VCAM-1 antibody did not. Adhesion molecules are not only extensively involved in cell adhesion *per se*, but also in signal transduction mediating cellular events, such as cell activation and proliferation [23]. A wide variety of adhesion-dependent lymphocyte interactions, in both antigen-dependent and -independent processes, are mediated through interaction of the integrin lymphocyte function-associated antigen 1 (LFA-1) with the immunoglobulin supergene ICAM-1, and of the immunoglobulin supergene family LFA-2 (CD2) with LFA-3. These interactions include T cell interaction with APC, cytotoxic T cell-mediated killing, and lymphocyte binding to the endothelium [24]. Whether adhesion molecules are required for T cell activation by superantigens has not been fully evaluated. Kotb *et al.* [25] reported that SEB and staphylococcal M protein stimulate T cells independent of accessory cells when a combination of phorbol myristate acetate, IL-1 and IL-6 is added. In contrast, a dependence on LFA-1 for the formation of homotypic aggregation of B cells in response to superantigen TSST-1 was described by Mourad *et al.* [26]. These findings suggest that LFA-1 and/or its ligand ICAM-1 may serve to provide accessory molecules for T cell activation in response to SEB. Furthermore, very late antigen 4 (VLA-4) and its ligand, VCAM-1 have also been demonstrated to co-stimulate CD3 MoAb-mediated and superantigen-mediated $CD4^+$ cell activation. The specific interaction of LFA-1/ICAM-1 and VLA-4/VCAM-1 not only leads to T cell proliferation, but also regulates the production of IL-2 and IL-4, and augments the level of secreted granulocyte-macrophage colony-stimulating factor (GM-CSF) [27].

In the present experiment, SEB-induced T cell proliferation in the presence of IFN-γ-stimulated synovial cells was blocked by anti-ICAM-1 MoAb. These results indicate that an LFA-1/ICAM-1 adhesion pathway plays a critical role in SEB-induced T cell proliferation in the presence of IFN-γ-treated synovial cells. In contrast, anti-VCAM-1 MoAb did not significantly inhibit SEB-induced synovial cells, because VCAM-1 expression on synovial cells was not induced by IFN-γ. Whether the VLA-4/VCAM-1 pathway mediates SEB-induced T cell proliferation by synovial cells is not clear. Using immunohistochemical methods, it was previously demonstrated that synovial cells from patients with RA aberrantly express HLA class II antigens and adhesion molecules including ICAM-1, VCAM-1 and LFA-3 [28, 29]. As shown in Fig. 3, IFN-γ induced the expression of HLA class II antigens and ICAM-1 adhesion molecules. IFN-γ treatment of synovial cells facilitated adhesive interaction with T cells [30, 31], a process mediated by the LFA-1 and ICAM-1 pathway [32, 33]. Our results documented that IFN-γ-treated synovial cells provided costimulatory signals to support superantigen-induced proliferation of resting peripheral blood-derived T cells. Thus, the LFA-1/ICAM-1 pathway may play an important role in mediating T cell proliferation in response to SEB, as the anti-ICAM-1 MoAb inhibited over 80% of SEB-induced T cell proliferation. Results of experiments using IFN-γ-treated synovial cells are consistent with *in vitro* studies using either recombinant ICAM-1 protein or ICAM-1-expressing cells to provide costimulation for T cell proliferation in response to superantigen [34–37].

It has been suggested that superantigens play a role in the pathogenesis of RA, since T cells bearing a particular TCR Vβ family expand in the affected joints [7, 38]. However, the role of superantigen in RA remains controversial, since T cell expansion in affected joints has been reported to express different TCR Vβ repertoires [7, 38–45]. Paliard *et al.* [7] demonstrated the preferential usage of TCR Vβ14, which is rarely used in peripheral blood T cells, and suggested a role for superantigen in the pathogenesis of RA. Furthermore, Howell *et al.* [38] analysed TCR Vβ chain gene expression of IL-2 receptor-positive synovial cells. They described three dominant gene families in the synovium, Vβ3, Vβ14 and Vβ17, that were similar in the fourth complementarity-determining region (CDR). Given that the binding sites for superantigens have been mapped to the CDR4s of TCR Vβ chains, synovial localization of T cells bearing Vβ3 with significant CDR4 homology indicates that Vβ-specific T cell activation by superantigen may play a role in RA.

The present study demonstrated that synovial cells serve as APC for SEB and induce T cell activation. The potency as APC of synovial cells for other superantigens was not investigated. Previous study demonstrated keratinocytes were able to present both SEB and SEA to T cells [16]. So synovial cells may be able to present other superantigens to T cells. Although the role of superantigens remains unclear, it is evident that superantigen-producing microbes can induce a broad spectrum of pathogenic responses. Friedman *et al.* [45] have previously suggested that during natural infections, superantigens may lead to immunostimulation and autoimmunity when a relatively small number of T cells are activated. This observation may be due to a lower dose of the superantigen, or to the presence of a smaller fraction of host T cells reacting with the autoimmune-inducing superantigens.

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