Introduction Rheumatoid arthritis (RA) is a systemic, chronic, inflammatory disease, which affects at least 1% of the human population. Patients with RA show humoral and cellular immune responses to various cartilage matrix proteins, i.e. type II collagen, proteoglycan (aggrecan), link protein, HC-gp39. Systemic immunization of BALB/c mice with human cartilage proteoglycan (PG) induces progressive polyarthritis. One approach to study these immunological events related to the initiation and maintenance of any autoimmune disease is to transfer the disease adoptively under controlled conditions. Here we describe a transfer system of the erosive autoimmune arthritis by injecting spleen cells from arthritic BALB/c mice into mice with severe combined immunodeficiency disease (SCID).

Materials and Methods Female BALB/c mice, aged 8-12 weeks, were injected intraperitoneally with 100 g of cartilage PG in Freund’s adjuvant at three-week intervals. After the 3rd injection the limbs were examined on a daily basis and abnormalities due to the arthritic changes in the peripheral joints were recorded on a scale of 0-4 per each paw with a maximum cumulative arthritis score of 16 for each animal. The appearance of joint swelling and redness was considered as the day of the onset of arthritis. Spleen cells and lymph node cells were collected from arthritic animals and Lympholyte-purified lymphocytes were injected into SCID mice intraperitoneally with or without 100 g of cartilage PG (antigen). The cell transfer was repeated on day 7, and if it was necessary, on day 28. SCID mice were assessed for arthritis and scored as described above. The cellular immune response was measured by antigen-specific Interleukin-2 (IL-2) production and T-cell proliferation, anti-PG antibody levels (both hetero- and auto-antibodies) were determined by ELISA.

Results The most effective transfer of arthritis was achieved by using either splenocytes (1x10^6 cells) or Lympholyte-purified spleen lymphocytes (2x10^6 cells) together with PG (100 g intraperitoneally). SCID mice injected with splenocytes exhibited earlier arthritis (24±6.4 days after the injection) with lower severity (3.9±1.8 cumulative arthritis score), than those injected with purified lymphocytes (30±10.3 days onset and 7.7±1.9 severity score) (Fig.1). All these cell-transfers from arthritic BALB/c mice into SCID mice resulted in 100% incidence, and the histopathology of the transferred disease was indistinguishable from those described in donor BALB/c mice. In contrast to the lack of immune response PG antigen alone did not induce arthritis in SCID mice. While the non-specific lymphocyte stimulation (Concanavalin A- for T cells, LPS- for B cells) revealed no differences between the groups, the in vitro stimulation with PG was positive only in arthritic (lymphocyte plus PG-injected) SCID mice (Fig. 2.). The human PG co-injected arthritic SCID mice possessed a 600-fold higher antibody titer when compared with the SCID mice injected only with cells. This antibody titer in arthritic SCID mice was even higher than the titer measured in the donor arthritic BALB/c mice (Fig. 3.).

Discussion/Conclusion This is the first report of an adoptive transfer of (PG-induced) arthritis into SCID mice. We observed 100% incidence of arthritis in the SCID mice administered with immunologically active cells and the specific antigen (aggrecan) at the same time. SCID mice injected only with antigen-specific lymphocytes, however, could not develop arthritis. The clinical and histological appearance of arthritis in SCID mice was indistinguishable from the PG-induced arthritis in the donor BALB/c mice. This in vivo responsiveness and incidence of arthritis closely correlated with in vitro tests: T- and B-lymphocytes from cell plus antigen-injected SCID mice exhibited a high proliferation, antibody production and IL-2 secretion, whereas lymphocytes from SCID mice injected without PG antigen co-administration did not respond to in vitro stimulation. This observation clearly suggests that the presence of accessible antigen is pivotal in the maintenance of arthritis, whereas the antigen processing and the presentation of arthritogenic epitopes are intact in SCID mice.

Our finding provides a very useful method to further investigate the mechanism of the development of arthritis and offers a special condition for studying cell-cell interaction, lymphocyte responses in vivo and in vitro conditions and the function of various lymphocyte subsets in any form of experimental arthritis.

**Fig. 1.** Severity of arthritis in SCID mice, transferred with lymphocyte-enriched spleen cell suspension. Mice were injected on day 1, 7 and 28 (days after the first injection on the x-axis). Only the mice co-injected with PG developed arthritis.

**Fig. 2.** T cell response (proliferation) of lymphocytes from arthritistransferred SCID mice. The lymphocytes from SCID mice transferred with lymphocytes and lymphocytes plus PG show identical response to the non-specific stimulation (Con-A: open bar, LPS: hatched bar), however, the PG stimulation provokes immune response only from lymphocyte and PG co-immunized SCID mice (closed bars).

**Fig. 3.** Humoral response to human PG (hetero antigen: hatched bar) and mouse PG (auto antigen: open bar) in arthritic BALB/c and lymphocyte-transferred SCID mice. The SCID mice, which were not co-administered with antigen (PG) show a 600-fold lower antibody titer, when compared with either arthritic BALB/c mice or PG co-administered SCID mice.

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Poster Session - Rheumatoid Arthritis - Hall E