A crucial role for macrophages in the pathology of K/B × N serum-induced arthritis

Samuel Solomon, Narendiran Rajasekaran, Elvira Jeisy-Walder, Scott B. Snapper and Harald Illges
Figure 1
Depletion of macrophages by Clodronate liposomes treatment: F4/F80 antibody immunostaining for macrophages. Liver sections showing macrophage depletion before (A) and after (B) Clodronate liposomes treatment. Bone marrow sections showing macrophage depletion before (C) and after (D) Clodronate treatment. Arrows indicate macrophages.
Figure 2:
(A) Macrophage depleted mice are resistant to arthritis. Macrophages were depleted by Clodronate liposomes treatment. Two i. p. injections of 200 µl of clodronate liposomes were done on days -3 and -1 to deplete macrophages prior to K/BxN sera transfer. 100 µl of K/BxN sera was injected i.p. on day 0. The mice were assessed for arthritis development by ankle thickness and clinical index score on days 0, 3, 5, 7 and 9. Mice treated with 100µl K/BxN sera alone and vehicle control, 100µl K/BxN sera and clodronate liposomes. (Tilted squares) - mice treated with clodronate liposomes prior to K/BxN sera transfer. (Squares) - mice treated with PBS liposomes prior to KBN sera transfer. Data are expressed as mean ± SEM; n=8-9.

(B) Ankle joint histology shows absence of pannus tissue in macrophage depleted mice. Ankle joints from macrophage non-depleted (A) show extensive pannus infiltration and cartilage erosion whereas (B) macrophage depleted mice show no inflammation of joints.

Figure 2
Figure 3

(A) Macrophage reconstituted mice are susceptible to arthritis. Macrophages were depleted by Clodronate liposomes treatment. Two i.p. injections of 200 µl of clodronate liposomes were done on day day-8 and -6. On day 0, peritoneal macrophages (2x10^7) were injected i.p followed by 100 µl of K/BxN sera. The mice were assessed for arthritis development by ankle thickness and clinical index score on days 0, 3, 5, 7, and 9. (Triangles) - mice depleted of macrophages and reconstituted with peritoneal macrophages again prior to K/BxN sera. (Squares) - mice depleted of macrophages and not reconstituted again with macrophages prior to KBN sera. Data are expressed as mean ± SEM; n=4.

(B) Ankle joint histology. Ankle joints from macrophage depleted and K/BxN sera transferred shows no signs of arthritis (A) whereas the macrophage reconstituted mice are susceptible to arthritis after K/BxN sera transfer (B) shown here by pannus tissue and cartilage erosion of ankle joints (arrow).
Figure 4
Mast cell degranulation in the absence of macrophages.
(A) Total and degranulating mast cell counts in the synovium of mice. Group A - Arthritic Balb/c mice after K/BxN sera injection. Group B - Balb/c mice treated with clodronate liposomes. Total mast cells Group A vs. Group B (P = 0.0655. P < 0.05 is considered significant).
(B) Histological staining showing the presence of mast cells in the inflamed synovium

Figure 4
Figure 4

B
Figure 5

CD40 deficient mice are not protected from sera induced arthritis. CD40\(^{-/-}\) mice were injected with 200µl of serum from arthritic K/BxN animals on day 0. Arthritis was evaluated by measuring clinical index and ankle thickening every two days (Materials and methods). Data are expressed as a mean +/- SEM. Hematoxylin-eosin staining of ankle sections from CD40\(^{-/-}\) mice treated with K/BxN sera and sacrificed on day 4. (A) Control BALB/c mice treated with K/BxN sera showing synovitis on day 4. (B) CD40\(^{-/-}\) mice showing synovitis. B = bone, C = cartilage, S = synovium.
Figure 6
WASP is not required in arthritis induced by K/BxN serum transfer. WASP-deficient mice were injected with 200µl of serum from arthritic K/BxN animals on day 0. Arthritis was evaluated by measuring clinical index and ankle thickening every two days (Materials and methods). Data are expressed as a mean +/- SEM. Hematoxylin-eosin staining of ankle sections from CD40-/- mice treated with K/BxN sera and sacrificed on day 4. (A) Control C57Bl/6 mice treated with K/BxN sera showing synovitis on day 4. (B) WASP deficient mice B = bone, C = cartilage, S = synovium.