Involvement of TLR 9 in the Pathogenesis of Erosive Autoimmune Arthritis and Osteoclastogenesis

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Disclosure Statement

Nothing to disclose

Toll-like receptors

- Receptors of the innate immune system
- Recognize **PAMPs** (Pathogen-Associated Molecular Patterns) that are expressed by pathogenic organisms (bacteria, viruses):
  - TLR1/ 2/ 6: bacterial l
  - TLR4: LPS
  - TLR5: flagellin
  - TLR3/ 7/ 8/ 9: nucleic

➢ Binding of ligands leads to **ac** **athways and transcription facors** (NFκB, AP-1, IRF3, IRF7 **inflammatory cytokines** (TNF-α, IL-6, IL-12, type I Inte

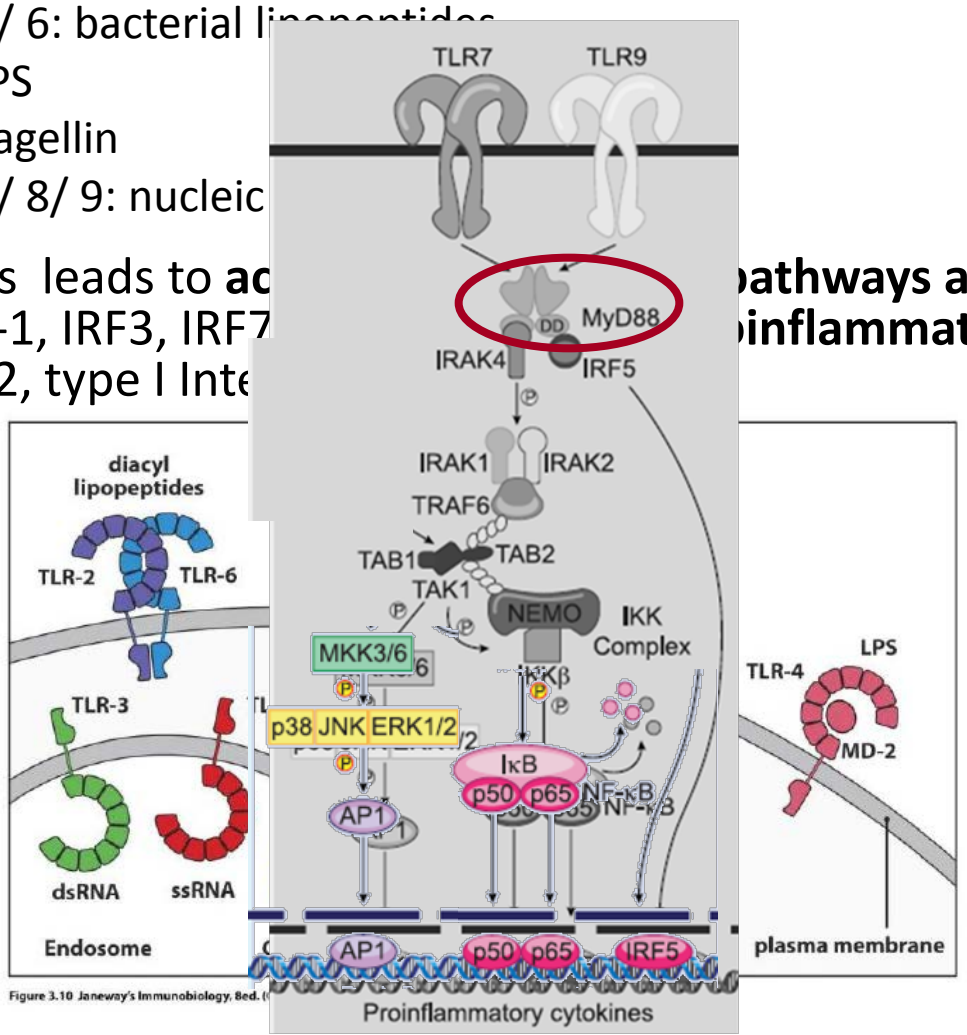
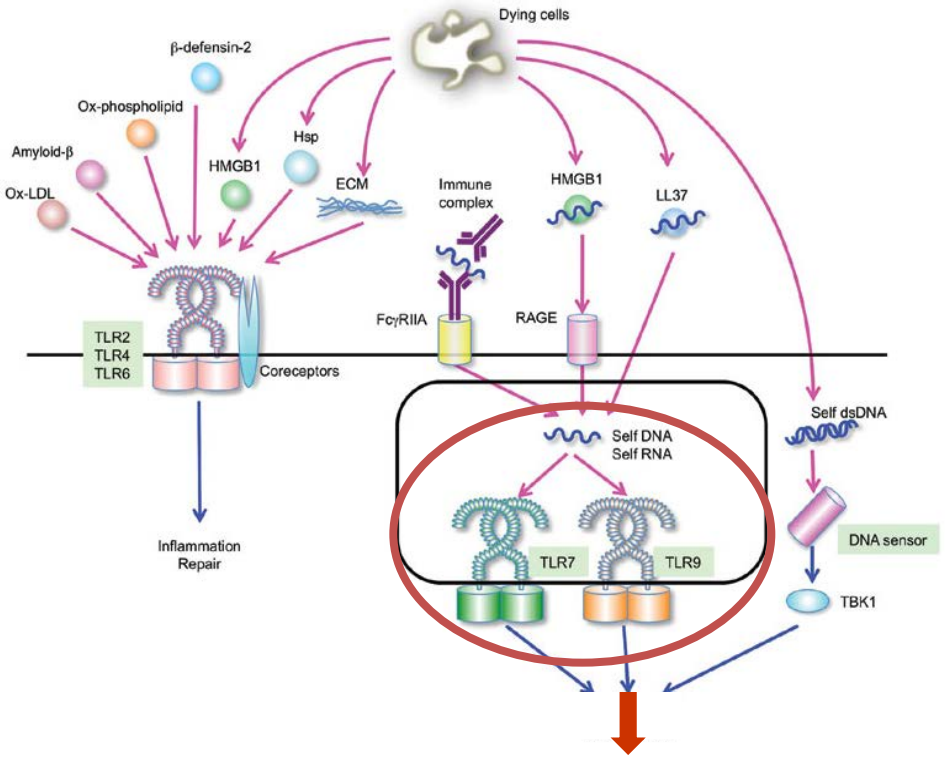


Figure 3.10 Janeway's Immunobiology, 8ed. (

Toll-like Receptors

- Recognize also **DAMPs** (Damage-Associated Molecular Patterns) endogenous ligands released by dying cells in the course of infection, trauma, tissue injury



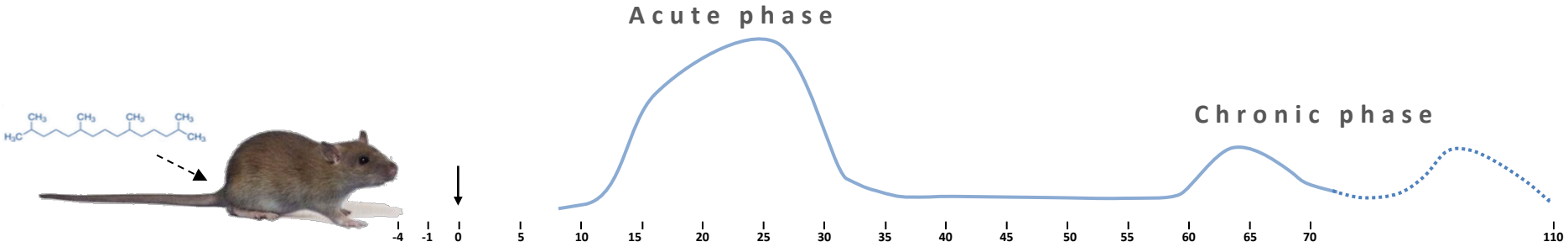
Adapated from Kawai and Akira, Nat. Immunol. 2010

Are Toll-like Receptors Involved in the Pathogenesis of Rheumatoid Arthritis?

Pristane-Induced Arthritis

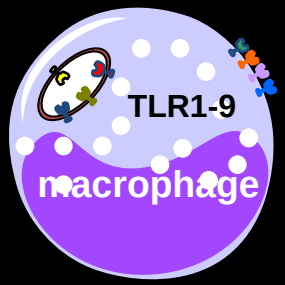
The best model to study autoimmune arthritis (RA)

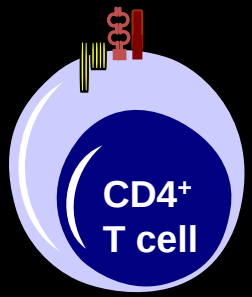
- **Arthritis induced in DA rats** by subcutaneous injection of **the mineral oil pristane**[1]
- **Pristane induces apoptosis/necrosis** in draining lymph nodes → **release of DAMPs**[2]
- **Acute and chronic** phase
- **Autoantibodies** which are also present in RA
  (RF, anti-RA33 (hnRNP-A2)[3], anti-carbamylated proteins[4])
- **T cell autoimmunity** (against hnRNP-A2[3] and related ribonucleoproteins)
- **Transferable by T cells and TLR7/9 activated APC**[5]

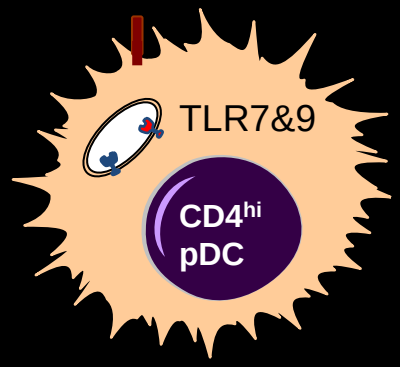


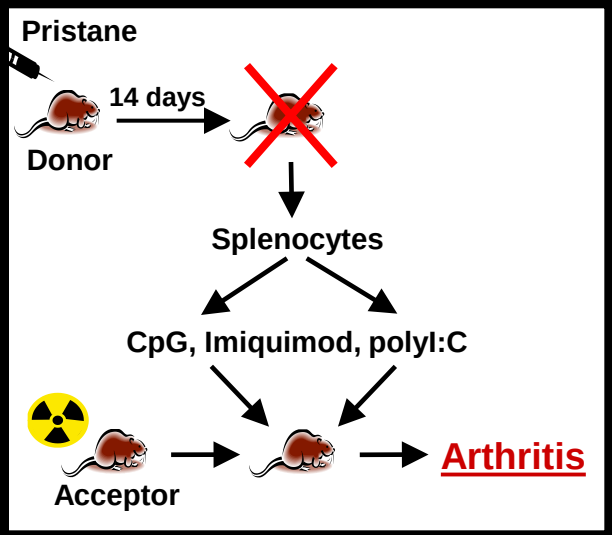
(1) Tuncel et al. Plos One 2016; (2) Herman et al. Autoimmunity 2012; (3) Hoffmann et al. J Immunol 2007; (4) Stoop et al. Ann Rheum Dis 2015; (5) Hoffmann et al. J Autoimm 2011.

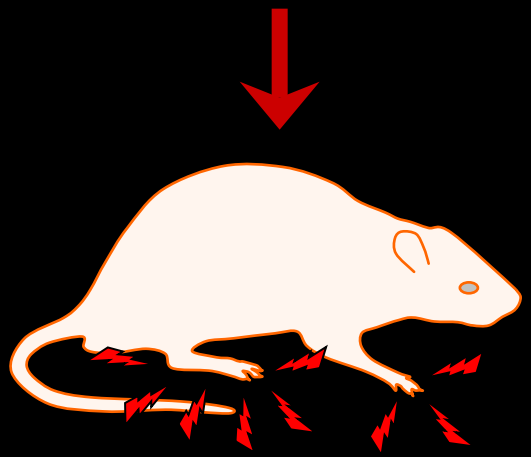
Disease transfer is mediated by T cells and macrophages activated *in vitro* by ligands of TLR 7/9

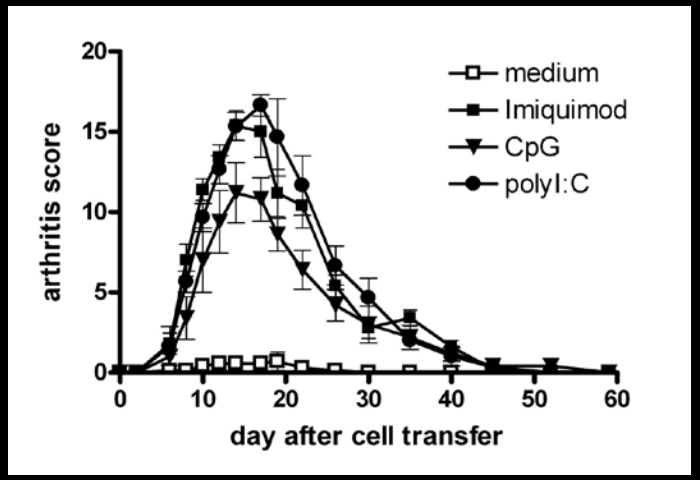








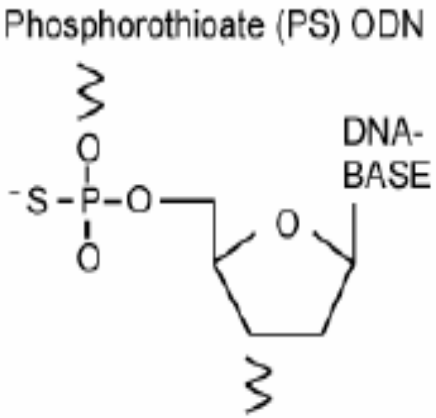


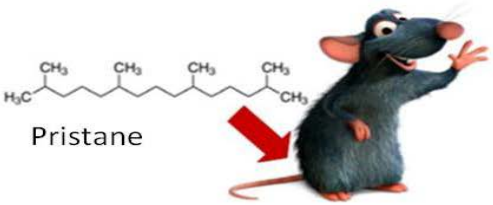


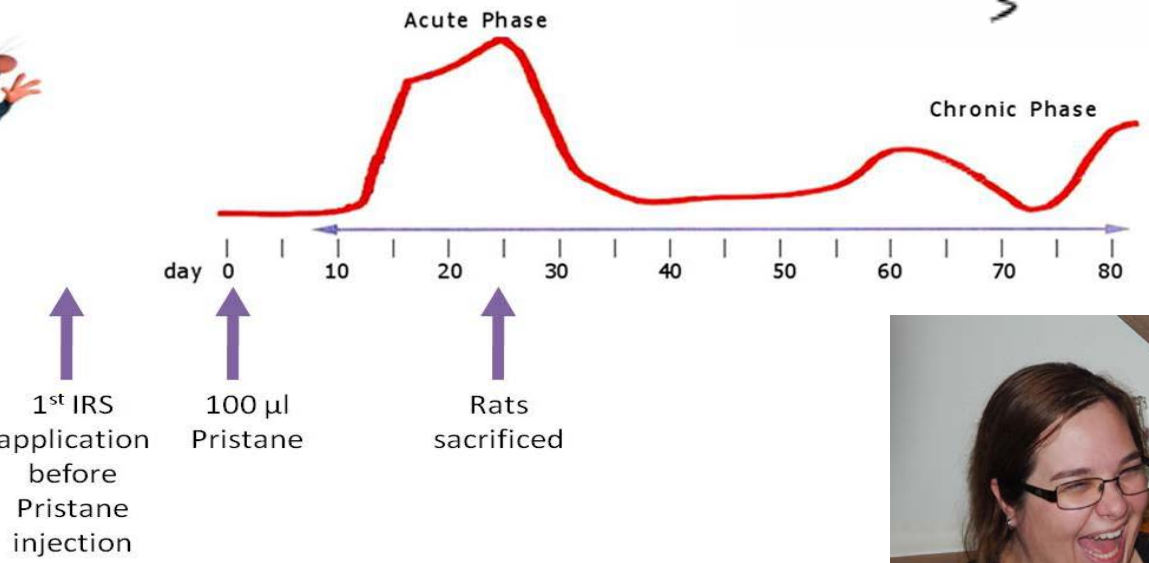
Hoffmann et al, 2011

Can TLR inhibitors influence the development of PIA?

- **Inhibitory oligonucleotides** (ODN):
  - **TLR7** (IRS 661)
  - **TLR9** (IRS 869)
  - **TLR7/TLR9** (IRS 954)
- ODN applied twice/week at the base of the tail
- PBS as placebo control

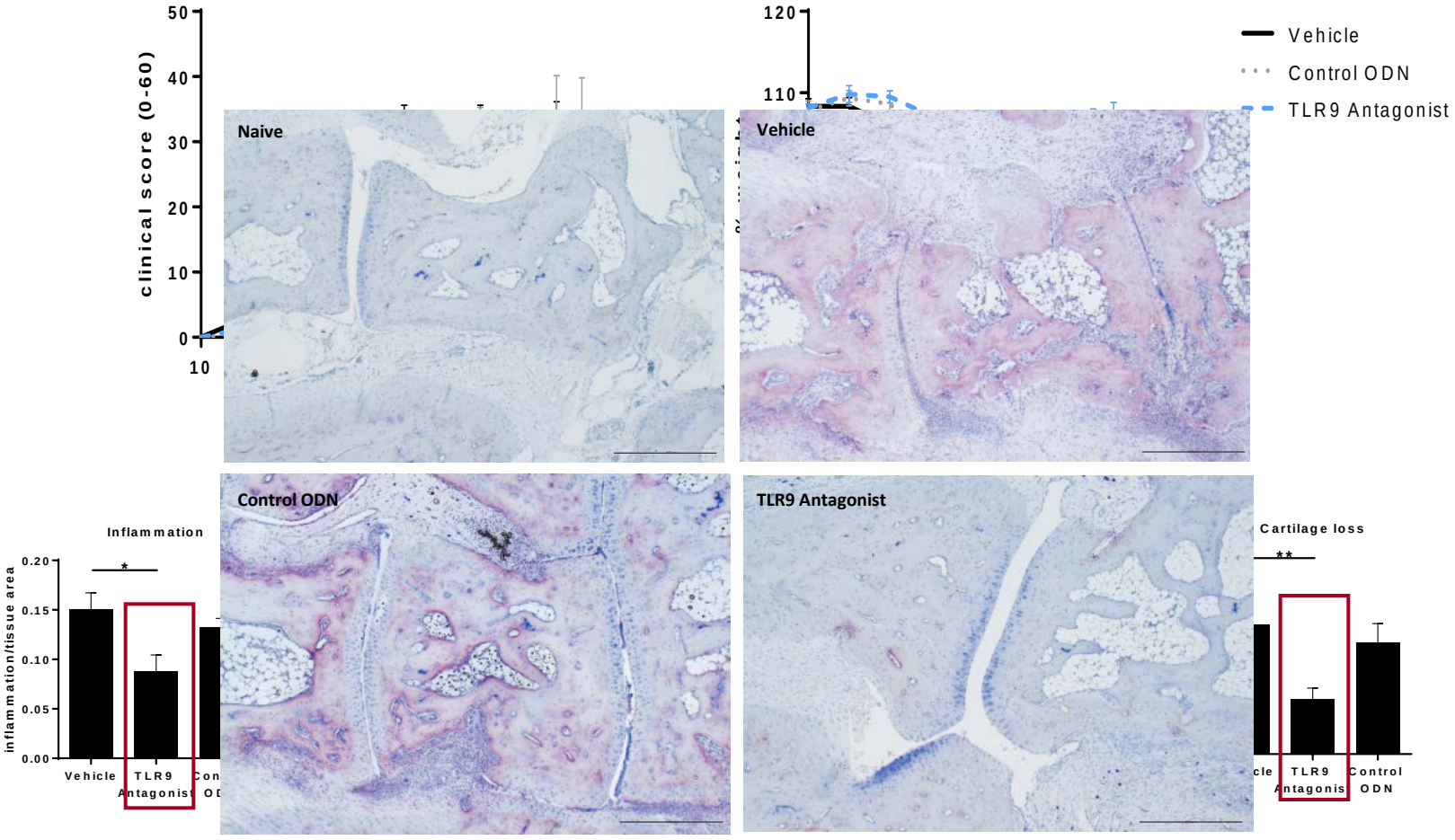




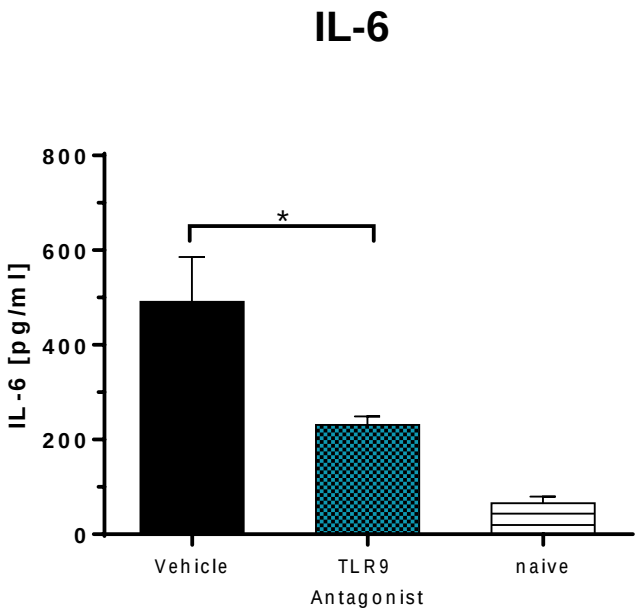


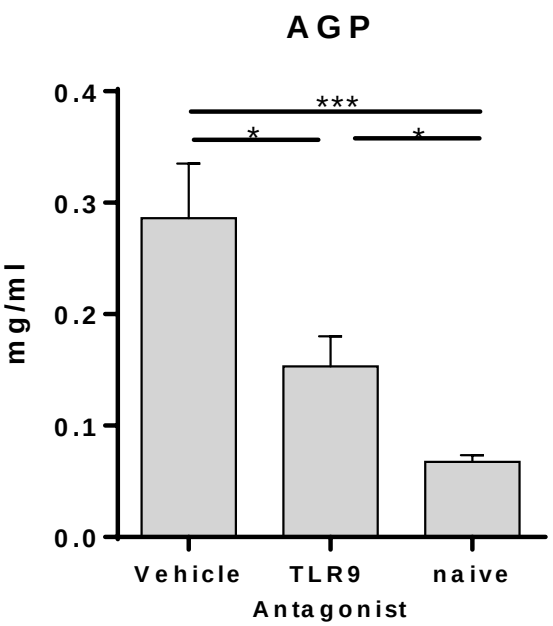
Anita Fischer, PhD thesis

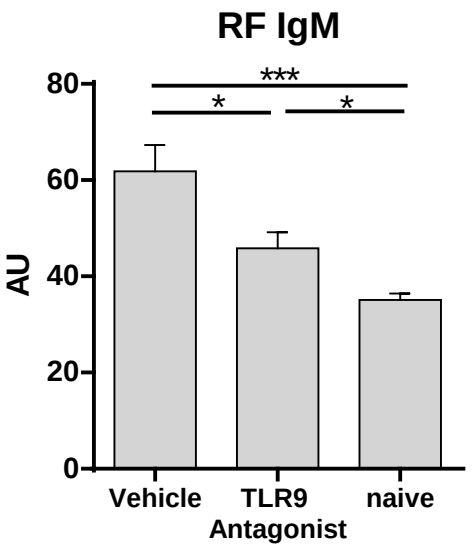
Treatment with a TLR9 inhibitor reduces severity of PIA



IL-6, AGP and RF-IgM are significantly reduced in sera of rats treated with the TLR9 inhibitor

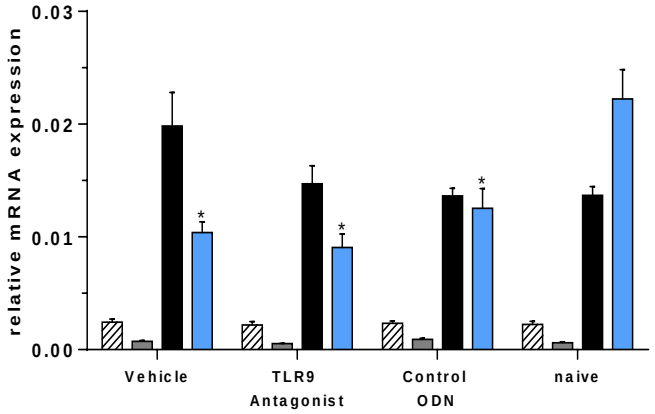






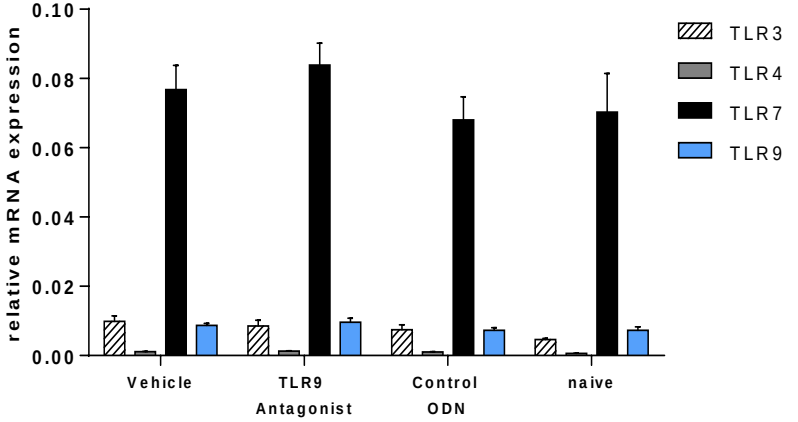
TLR expression in lymph node and spleen

Lymphnode



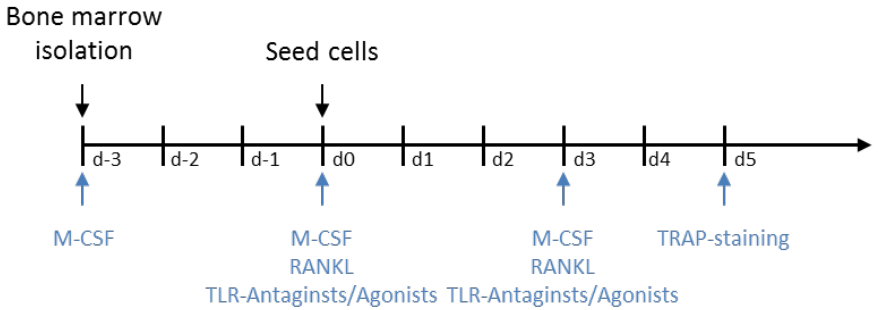
High expression of TLR7 and TLR9
Low expression of TLR3
Very low expression of TLR4

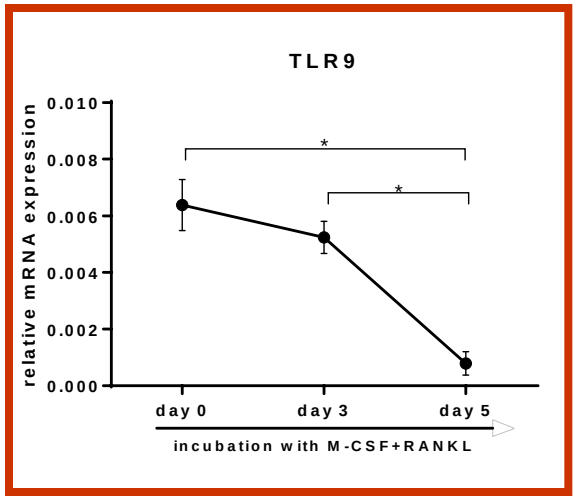
Spleen

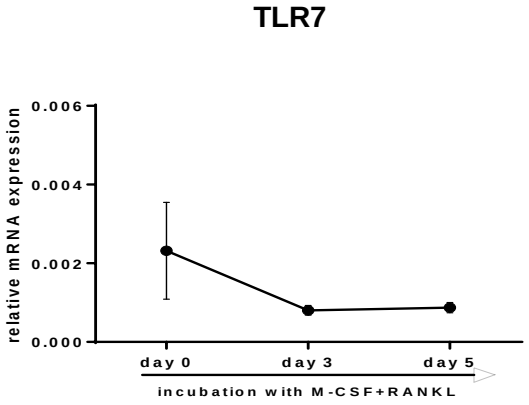


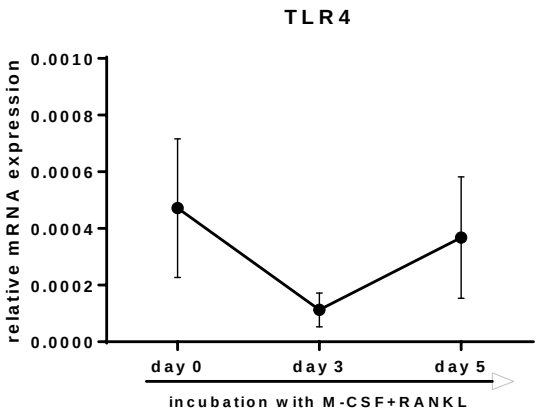
High expression of TLR7
Low expression of TLR3 and TLR9
No expression of TLR4

TLR expression during osteoclastogenesis



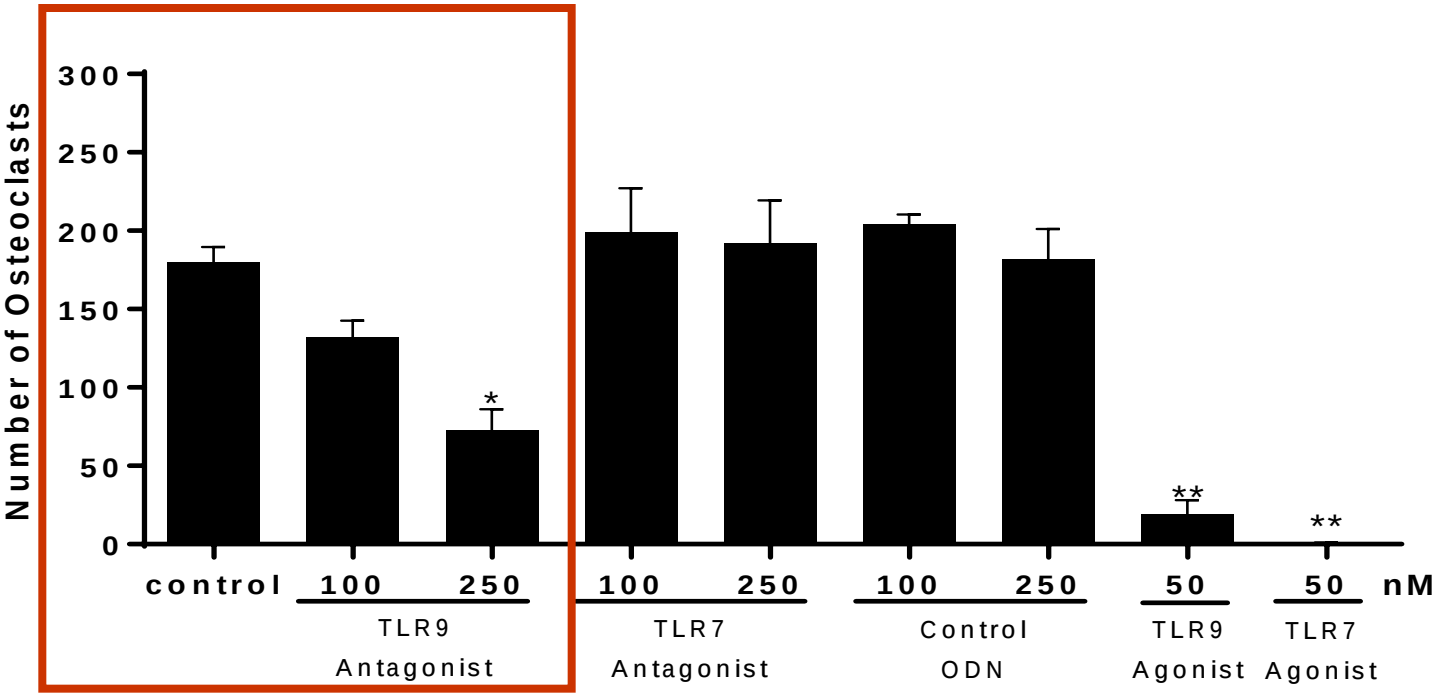






RANKL induces downregulation of TLR9

Influence of TLR9 and TLR7 antagonists on osteoclastogenesis



The TLR9 antagonist reduces osteoclast differentiation

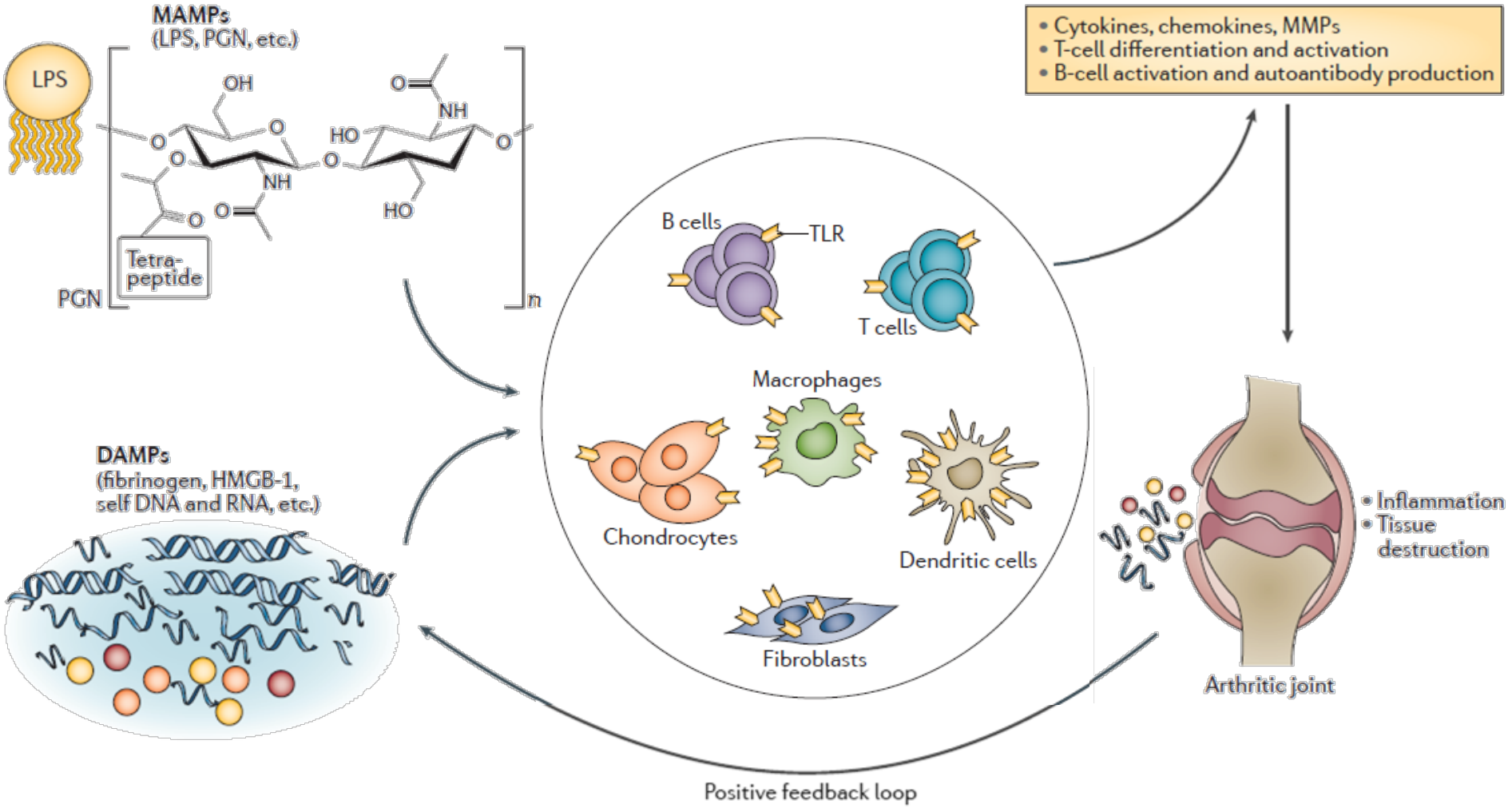
TLR9 and PIA

- TLR9 is presumably activated by DNA derived from apoptotic and/or necrotic cells.
- TLR9 seems to play a crucial role in the T cell driven initial phases of PIA arthritis via induction of proinflammatory cytokines promoting arthritogenic autoimmune responses.
- TLR9 seems to be involved in osteoclastogenesis.
- **Relevance for human RA?**

There is some evidence….

- **Expression of TLR3, -7 and -9 in the synovial tissue of RA patients**. [Radstake et al, AmJ Path 2004; Roelofs et al,A&R 2005; Kyburz et al, A&R 2003]
- **Endogenous TLR ligands in RA synovial fluid.** [Schett et al, A&R 1997; Yu et al, A&R 1997; Taniguchi et al A&R 2003]
- **Supernatants from cultured RA synovial membrane can activate macrophages in a MyD88 (TLR) dependent manner**. [Sacre et al, Ann Rheum Dis 2007]

Contribution of TLRs to chronic Inflammation and Autoimmunity in Arthritis



Joosten et al., Nat. Rev. 2016

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Masterswitch