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Anti-Ly9 (CD229) antibody treatment reduces marginal zone B cell numbers and salivary gland inflammation in a mouse model of Sjögren's Syndrome

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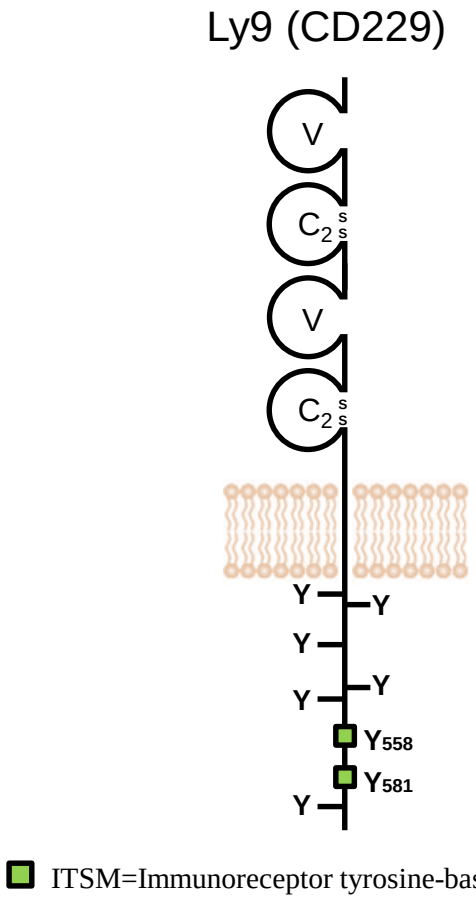
Ly9 (CD229) IS AN IMPORTANT IMMUNE-MODULATOR OF AUTOIMMUNITY AND B CELLS



• Unique **SLAM family** molecule with 4 Ig-like domains.

• Ly9 interacts in a **homophilic** manner.

• Expression restricted to lymphocytes. Highly expressed on innate-like lymphocytes, such as NKT cells and MZ B cells.

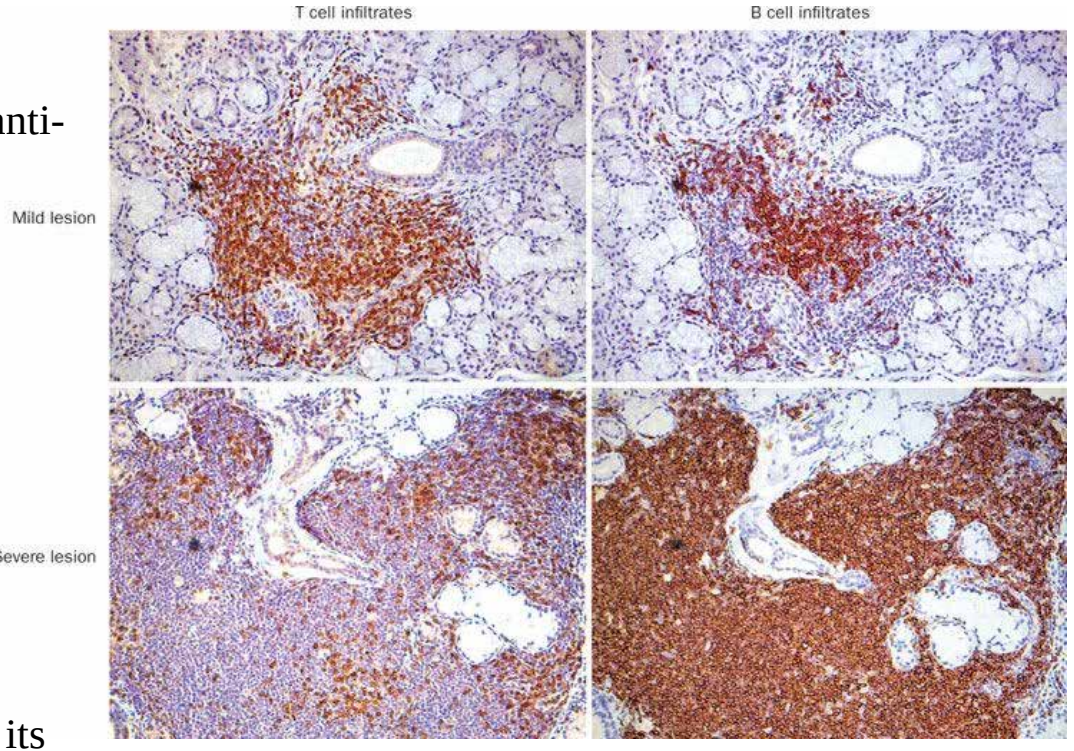
•Contains two immunoreceptor tyrosine-based switch motifs (ITSM) in its cytoplasmic tail. Recruits SAP adaptor molecule or phosphatases (SHP2 or SHIP-1).

•Aged *Ly9*-deficient mice **spontaneously** develop features of systemic **autoimmunity** (de Salort et al., Front Immunol 2013)

•Ly9 is a negative **regulator** for the development of **marginal zone** B cells (Cuenca et al., *J Immunol* 2015)

SJÖGREN'S SYNDROME (SjS) MAY BE A POTENTIAL TARGET FOR Ly9 TARGETING

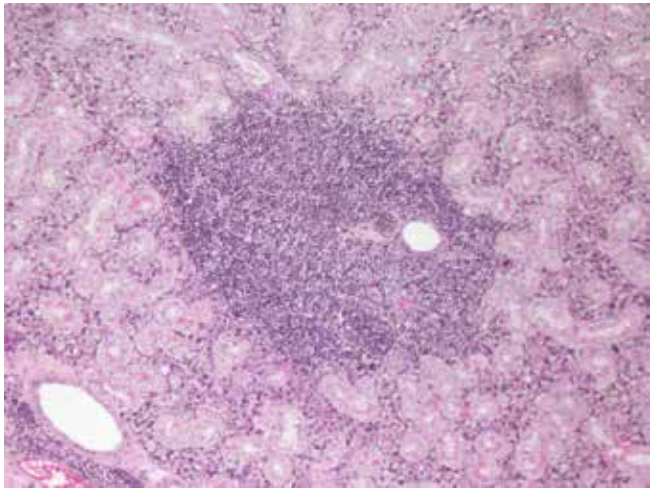
- SjS is a common chronic **autoimmune** disease affecting **salivary/lacrimal glans** (dryness) but also extraglandular tissues
- Presence of **autoantibodies** (anti-dsDNA, anti-Ro, Rheumatoid Factor) are key markers
- B-cell hyperactivity induces **MZ B cell expansion** that can lead to **MZ-B cell lymphoma**
- **B cells play a key role** in both humans and mice SjS
- All current **therapies have failed**
- **Murine models** are essential to understand its pathophysiology



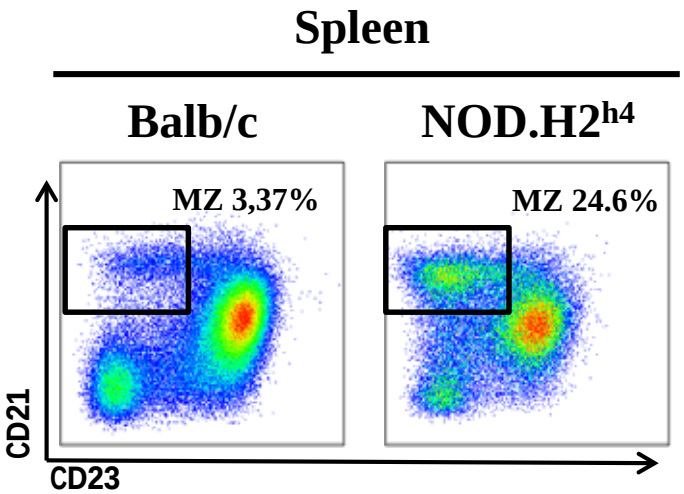
Salivary gland tissue with lymphocytic infiltrate. Voulgarelis, M. & Tzioufas, A. G. Nat. Rev. Rheumatol. 6, 529–537 (2010). Lymphocytic CD3 and CD20 cell infiltrate. x200

THE NOD.H-2^{h4} MICE: AN SPONTANEOUS MODEL OF SJS-LIKE DISEASE

- The NOD.H-2^{h4} mice **spontaneously** develope SjS-like disease but not diabetes
- At **24 weeks-old** all female mice have **SjS-related autoantibodies** and salivary gland infiltrates
- The NOD.H-2^{h4} mice show abnormal **MZ B cell pool enlargment**



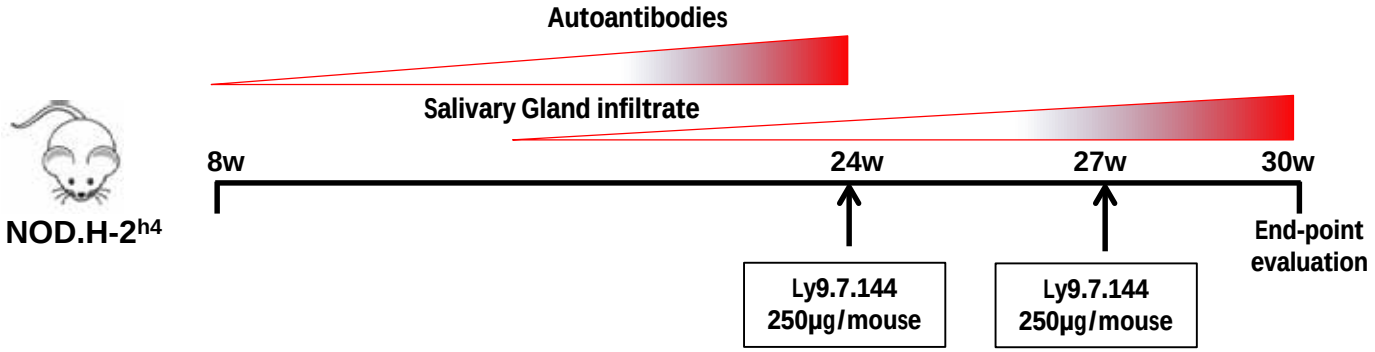
Salivary Gland infiltrate of NODH2h4 mice. 10x.



Previous results demonstrated that **treatment of mice with Ly9.7.144**, an agonistic mAb against moue Ly9, had a **dramatic effect on splenic MZ and B1 B cells**. Since these two subsets of innate-like B cells have been postulated to be **essential players in SjS** in both humans and mice, we decided to test the hypothesis that injection with **Ly9.7.144** may be able to **hinder** the development of **autoimmunity in NOD.H-2^{h4}** mice

Specific Objectives

1. To test the ability of Ly9.7.144 to **deplete the MZ B cells** and other subsets in the murine model of SjS NOD.H-2^{h4}.
2. To check if treatment with anti-Ly9 mAb have an impact on **salivary gland infiltrates** of d NOD.H-2^{h4} mice.
3. To assess if Ly9.7.144 treatment is able to **reduce the levels of autoantibodies** in NOD.H-2^{h4} mice.
4. To prove if anti-Ly9 mAb is able to **protect** NOD.H-2^{h4} mice **from developing** SjS disease.

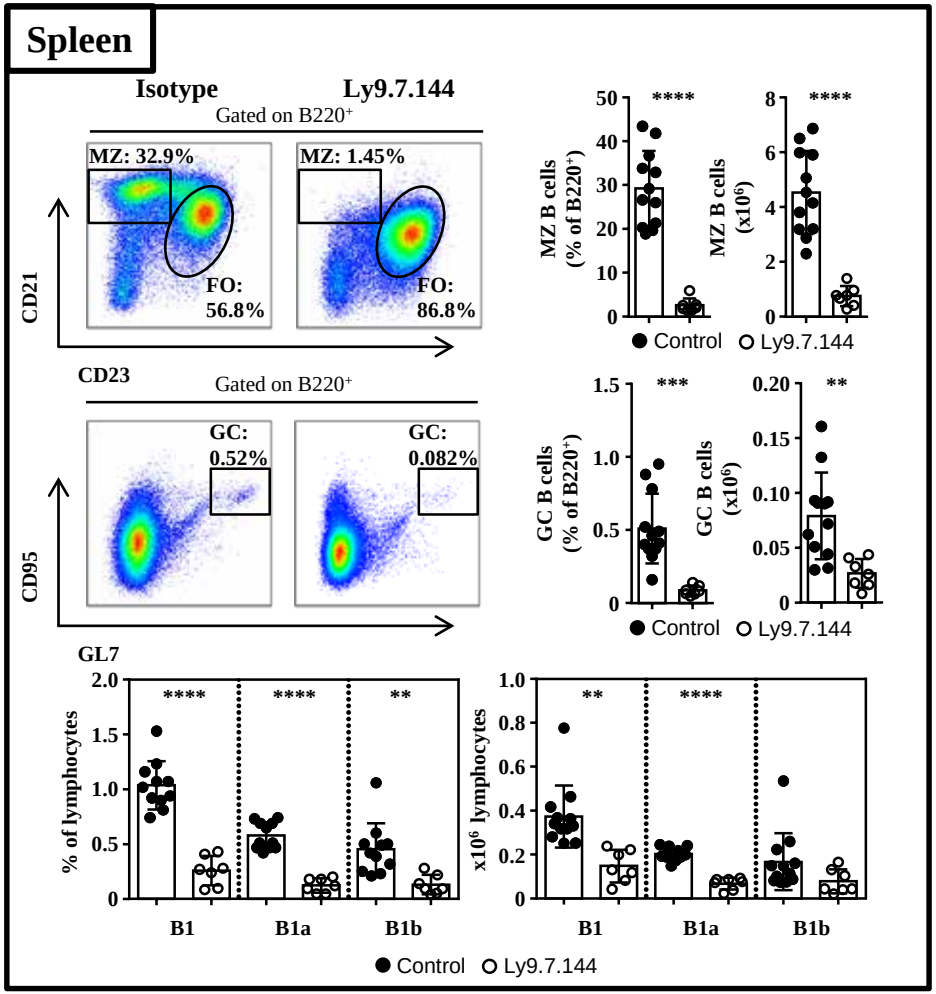


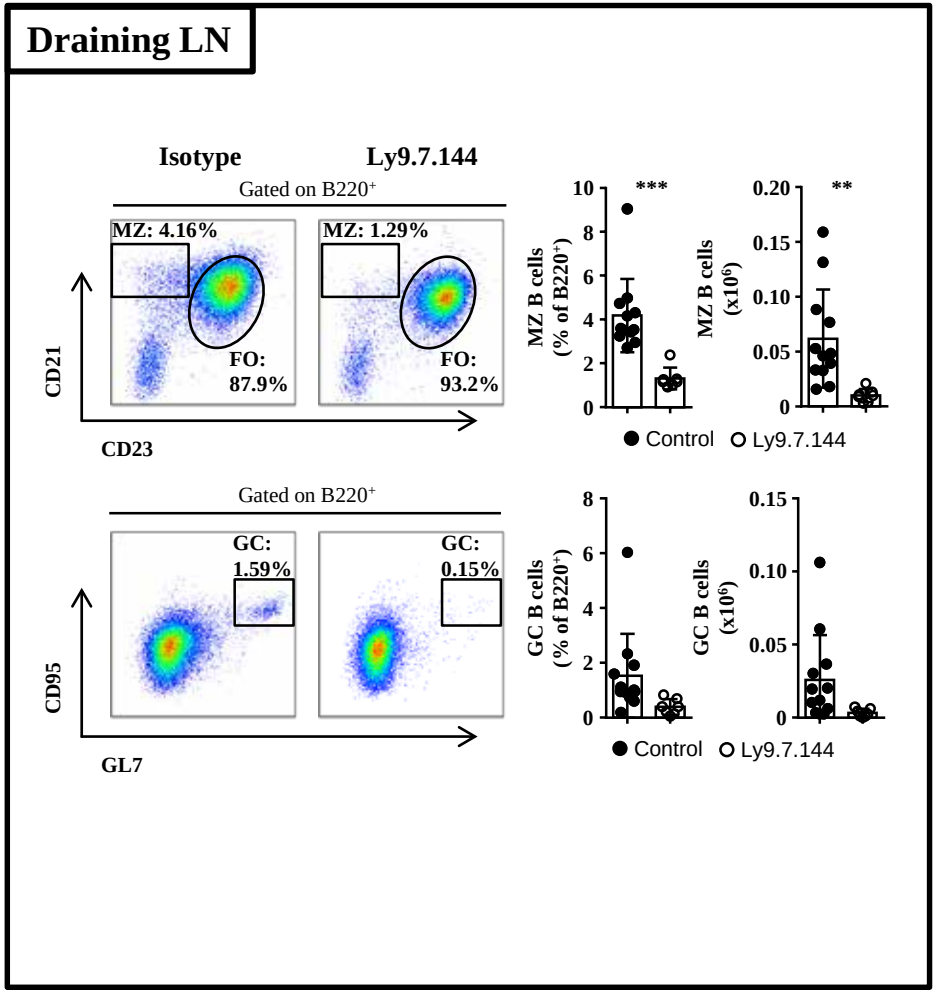
Read-out:

Organs
- Spleen
- Draining LN
- Salivary Gland
- Kidney

Plasma

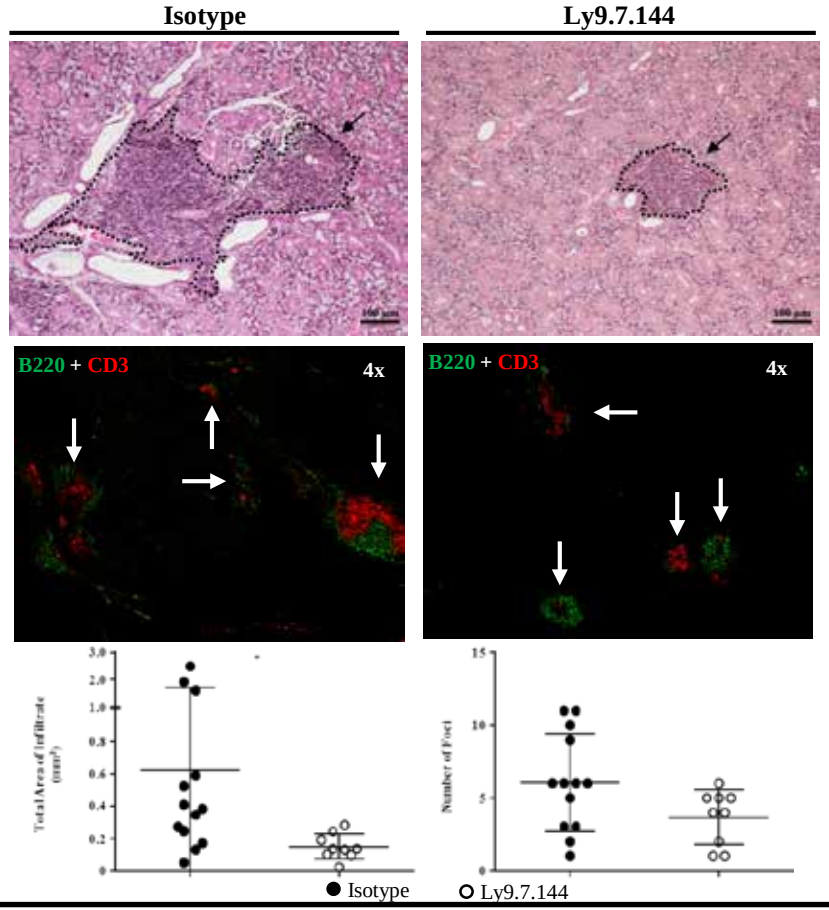
Results: MZ, B1 and GC B cells are depleted in different lymphoid organs

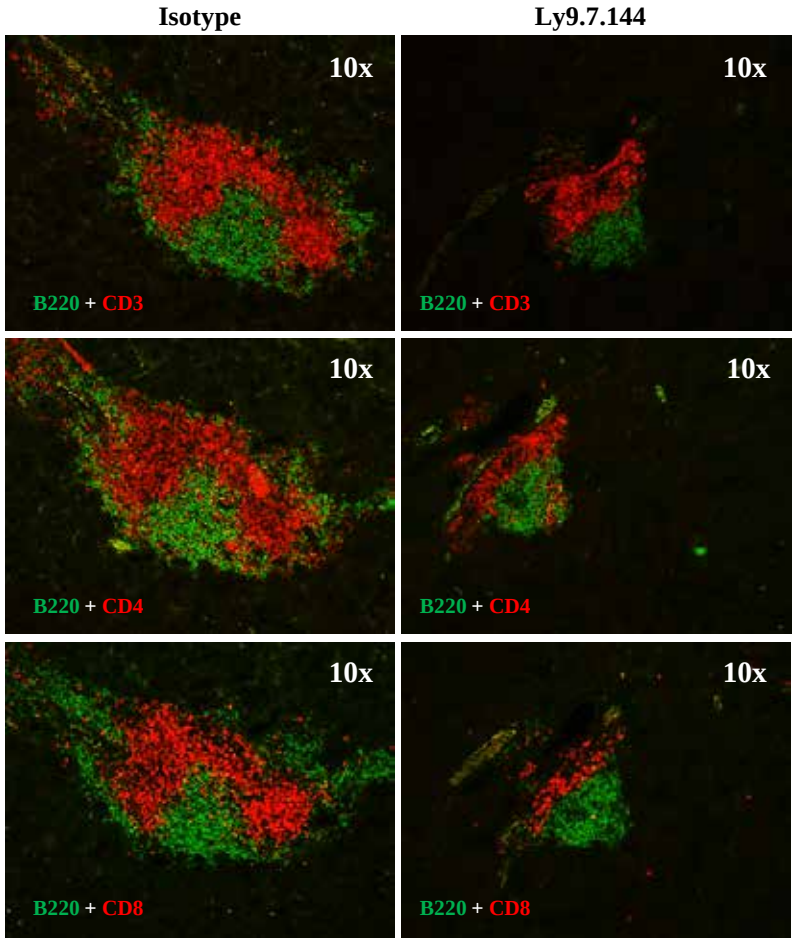




Results: Salivary gland area of infiltrate is reduced in anti-Ly9 treated NOD.H-2^{h4} mice

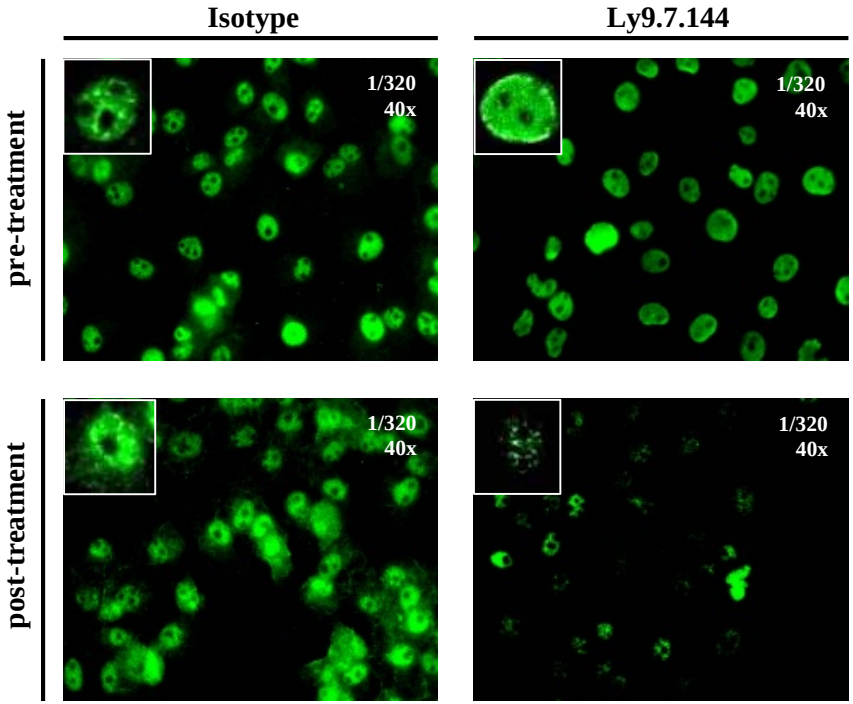
Salivary Gland

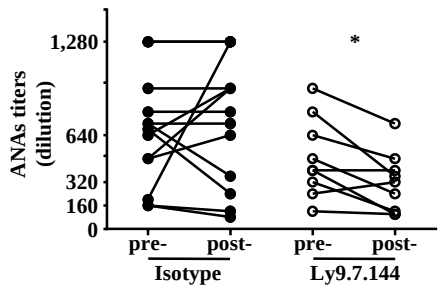


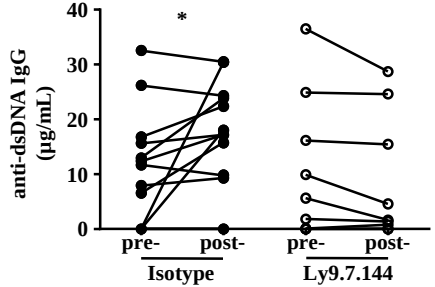


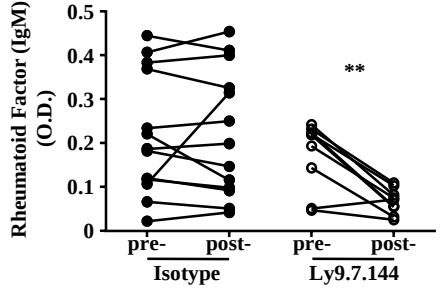
Results: Treatment with Ly9.7.144 is able to affect autoantibodies levels

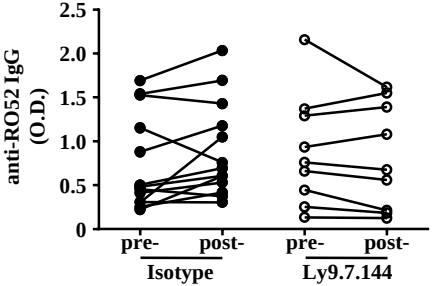
Serum





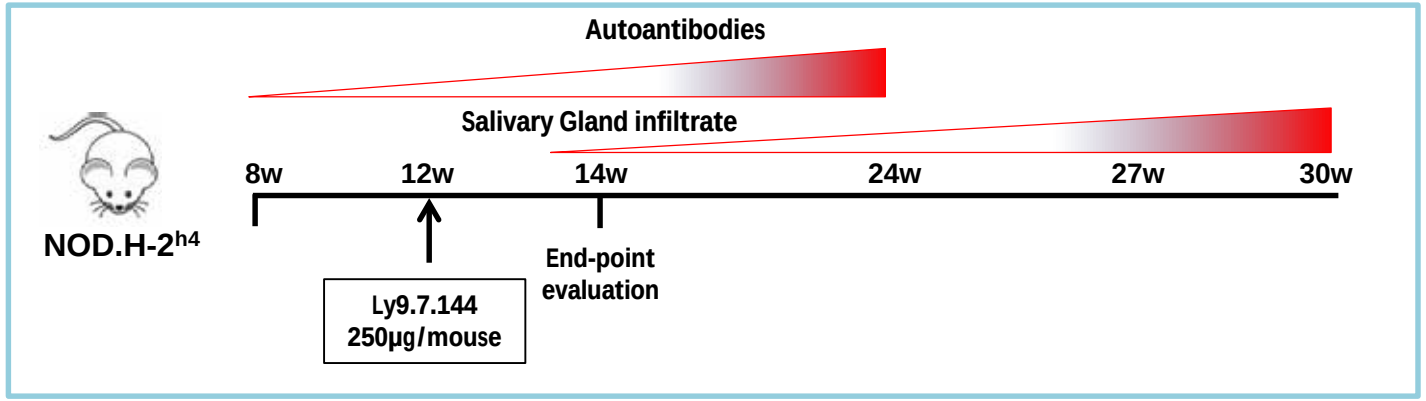


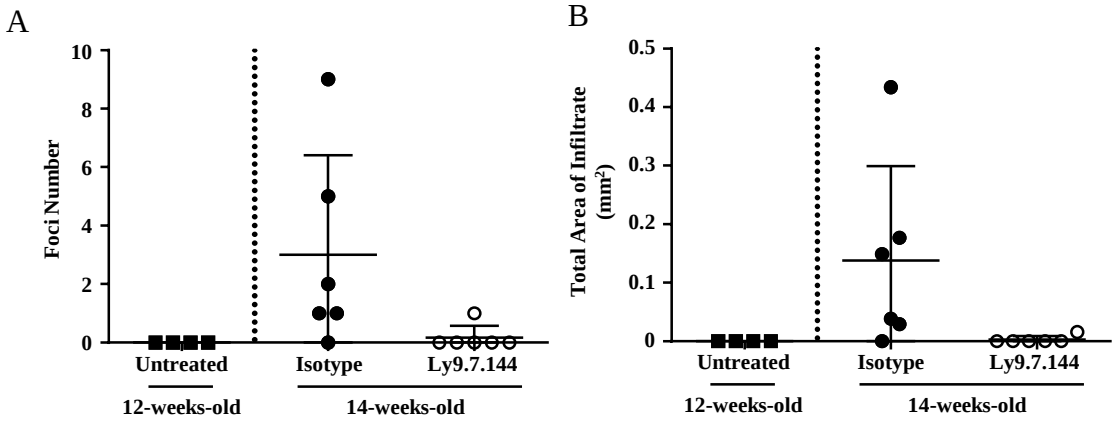




Results: Early treatment with Ly9.7.144 is able to protect from salivary gland infiltration

Salivary Gland





CONCLUSIONS

- Ly9 (CD229) targeting is able to deplete MZ, B1 and GC B cells in the spleen and draining lymph nodes
- Mice treated with the mAb anti-Ly9 show reduced area of lymphocyte infiltrates in the salivary glands
- The administration of Ly9.7.144 is capable to affect autoantibody levels in mouse sera
- Early anti-Ly9 mAb treatment before disease onset impedes salivary gland infiltration
- In contrast with the B cell depletion therapies, the selective deletion of MZ, B1 and GC B cells should be regarded as an attractive new therapeutic approach

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THANKS!

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