B6-hIL18R1/hIL18RAP

Strain Name： B6/JGpt-*Il18r1*$^{em1Cin(hIL18R1)}$*Il18rap*$^{em1Cin(hIL18RAP)}$/Gpt
Strain Type： Knock-in
Strain Number： T053316
Background： C57BL/6JGpt

Description

Interleukin-18 (IL-18) that belongs to the IL-1 family, is a potent pro-inflammatory cytokine involved in host defense against infections and regulates the innate and acquired immune response. IL-18 receptor (IL-18R) is requisite for IL-18 signaling. The alpha chain (IL-18R α or IL18R1) has a weak binding affinity for IL-18. Upon binding of IL-18 to IL-18 R α , the other subunit—the IL-18 receptor accessory protein (IL18RAP, also known as IL-18R β)—is then recruited to form a heterotrimeric complex for high-affinity binding, initiation of cellular signaling, up-regulation of the expression of a variety of inflammatory cytokines, such as Il-4, Il-13, IFN- γ , and TNF, which facilitates cell differentiation/survival, and FasL which facilitates apoptosis. The broad biological role of the IL-18 cytokine on immune cells have revealed its potential role in inflammatory and autoimmune diseases.

Systemic lupus erythematosus (SLE) is a female-biased, chronic systemic autoimmune disorder .Elevated expressions of IL18RAP mRNA and protein were observed in neutrophils from SLE patients, and that IL18RAP expression is positively correlated with disease activity[1]. In the MRL/lpr mouse model of spontaneous lupus erythematosus, The lymphocytes of MRL/lpr mice were found to have an overexpression of Il18rap; In addition, IL-18R α -deficient MRL/lpr mice exhibited decreased levels of proteinuria and serum anti-DNA antibodies, attenuation in renal pathology, and longer survival[2]. In systemic-onset juvenile idiopathic arthritis (SJIA), an overexpression of IL18RAP in neutrophils was observed in patients with active disease[3]. In addition, the majority of studies on IL18RAP focus on genetic association analyses that encompass a diverse spectrum of conditions including cancer, cardiovascular disease, autoimmunity, and infections. In autoimmune diseases, the rs9’’17997 single nucleotide polymorphism (SNP) in IL18RAP was found to have a divergent role, conferring risk for celiac disease but protection for type I diabetes[4]. Not only affecting disease susceptibility, IL18RAP expression in synovial tissues was shown to associate with treatment response in rheumatoid arthritis patients[5].

In addition, the IL-18/IL-18R axis represents double-edged swords in cancer, as their activation may promote tumor development and progression or oppositely, enhance anti-tumor immunity and limit tumor growth. Studies have found that in mouse models of inflammation-driven colon carcinogenesis induced by azoxymethane and dextran sulfate, Il18−/− and Il18R−/− mice are highly susceptible to colitis and colorectal cancer development, indicating that the IL-18/IL-18R axis has a protective effect on the development of colorectal cancer and an anti-tumor activity[6]. Downregulation of T-cell cytotoxic marker IL18R1 promotes cancer proliferation and migration and is associated with dismal prognosis and immunity in lung squamous cell carcinoma. Other studies indicate that IL-18 has a dual role in tumors, as it may exert proinvasive, proangiogenic, and immune-regulatory activities in different tumor models. In a melanoma mouse model, IL-18 enhances the development of lung metastases through the induction of PD-1 on NK cells. A subset of human melanomas expressing IL-18R shows enhanced prometastatic activity in nude mice relative to IL-18R-negative melanomas[7]. Other reports have shown that IL-18 has a pro-cancer effect on advanced gastric cancer, in a subset of melanomas, and in T cell acute leukemia(T-ALL). Along this line, high levels of IL-18 were found in different cancers, and IL18 gene polymorphisms were associated with some cancers[8].

GemPharmatech used the gene editing technology to develop the B6-hIL18R1/hIL18RAP humanized mouse model, and replaced the *Il18r1* gene and *Il18rap* gene fragments of B6 mice with the corresponding human *IL18R1* gene and *IL18RAP* gene fragments. This strain is an ideal model for studying autoimmune diseases and cancer.

Strategy

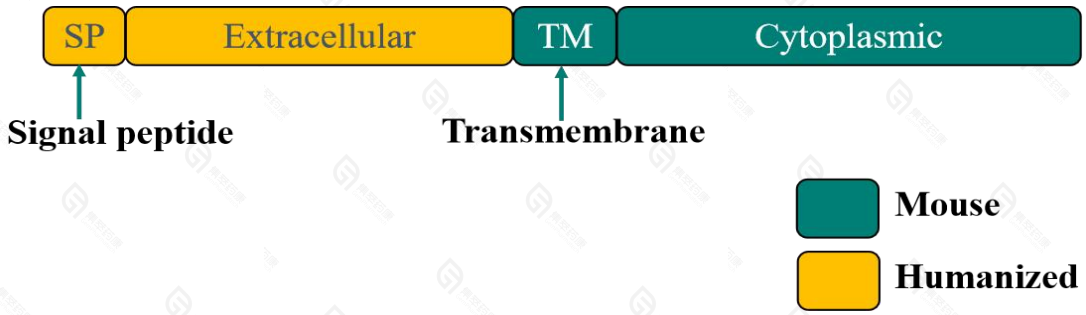


Fig 1. Schematic diagram of IL18R1 humanization strategy on B6-hIL18R1/hIL18RAP mice.

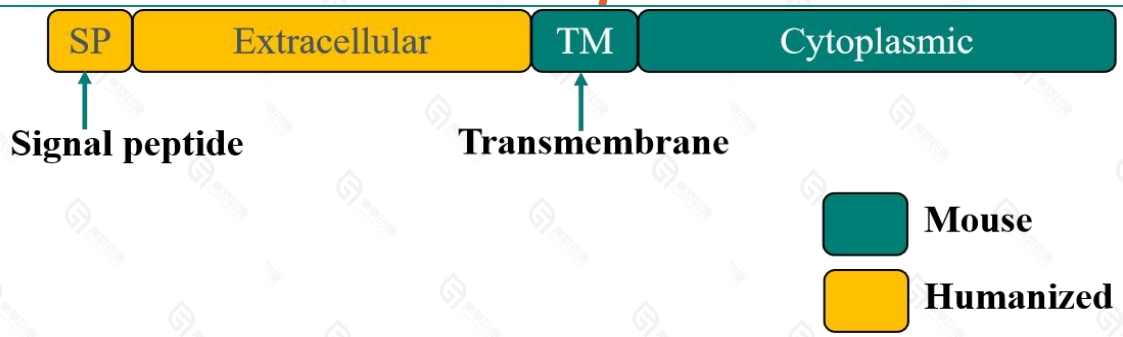


Fig 2. Schematic diagram of IL18RAP humanization strategy on B6-hIL18R1/hIL18RAP mice.

Applications

1. Efficacy evaluation of human IL18R inhibitors or agonist
2. Safety study of anti-hIL18R antibody
3. Research on autoimmune diseases
4. Research on immune-oncology

Supporting Data

1. **Detection of *IL18R1* mRNA expression**

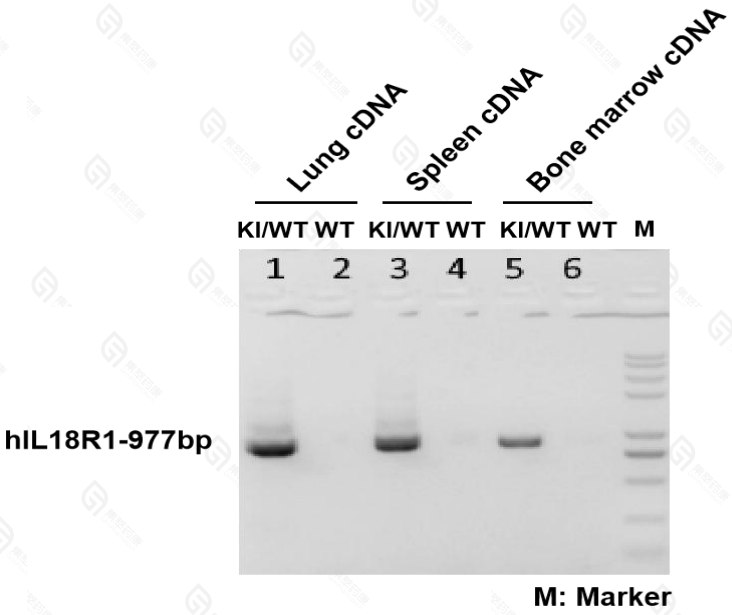


Fig 3. Detection of *hIL18R1* mRNA expression in B6-hIL18R1/hIL18RAP mice.
Human *IL18R1* mRNA was detectable only in lung tissue ,spleen tissue and bone marrow tissue of heterozygous B6-hIL18R1/hIL18RAP mice (KI/WT; KI/WT) but not in wild type C57BL/6(WT) mice.

2. Detection of *IL18RAP* mRNA expression

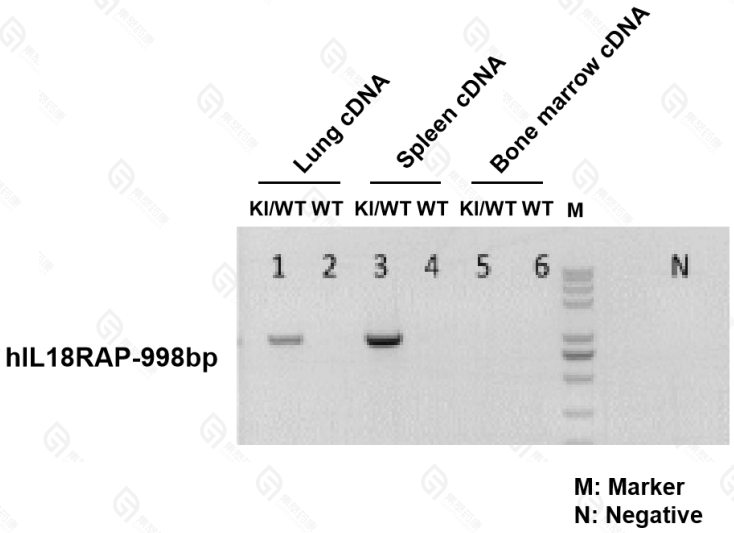


Fig 4. Detection of *hIL18RAP* mRNA expression in B6-hIL18R1/hIL18RAP mice.
Human *IL18RAP* mRNA was detectable only in lung tissue and spleen tissue of heterozygous B6-hIL18R1/hIL18RAP mice (KI/WT; KI/WT) but not in wild type C57BL/6(WT) mice.

3. Analysis of IL18R1 expression

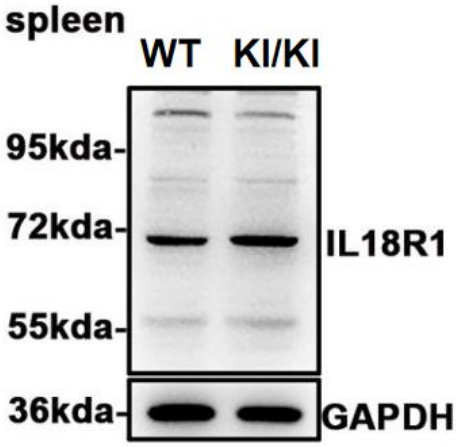


Fig 5. Analysis of IL18R1 expression in B6-hIL18R1/hIL18RAP mice by western blot.
Spleen samples were collected from B6 and homozygous B6-hIL18R1/hIL18RAP mice (KI/KI;KI/KI) and analyzed by western blot with anti-IL18R1 antibody. Human IL18R1 was expressed in the tissue of spleen in homozygous B6-hIL18R1/hIL18RAP mice. The anti-IL18R1 antibody used in this study is cross-reactive with human and mice IL18R1.

4. Analysis of IL18RAP expression

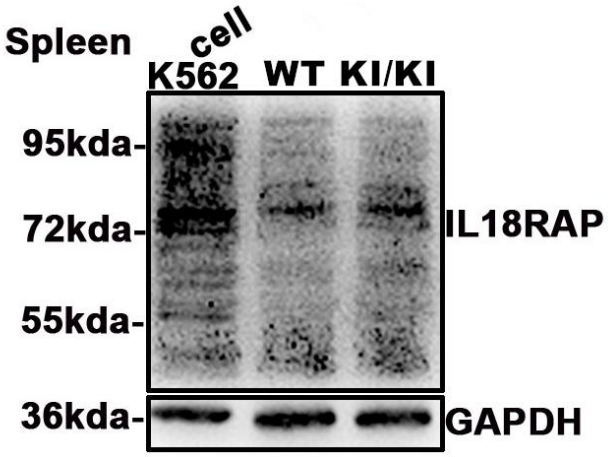
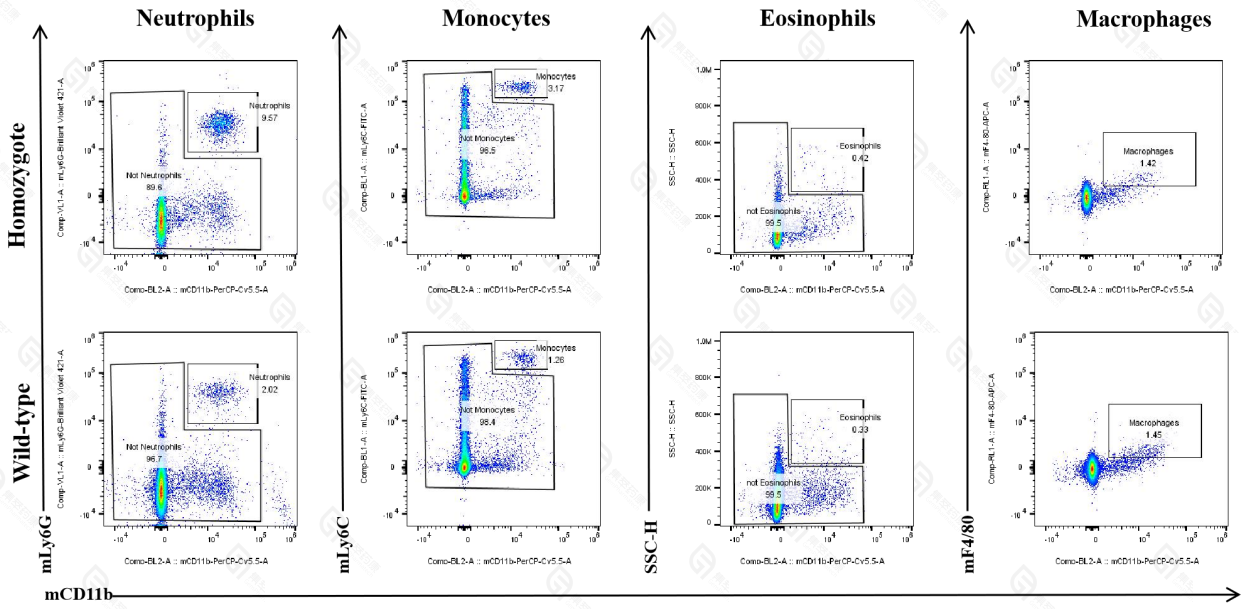


Fig 6. Analysis of IL18RAP expression in B6-hIL18R1/hIL18RAP mice by western blot.
Spleen samples were collected from B6 and homozygous B6-hIL18R1/hIL18RAP mice (KI/KI;KI/KI) and analyzed by western blot with anti-IL18RAP antibody. K562 cells were used as the positive control. Human IL18RAP was expressed in the tissue of spleen in homozygous B6-hIL18R1/hIL18RAP mice. The anti-IL18RAP antibody used in this study is cross-reactive with human and mice IL18RAP.

5. Analysis of immune cell subpopulation in blood



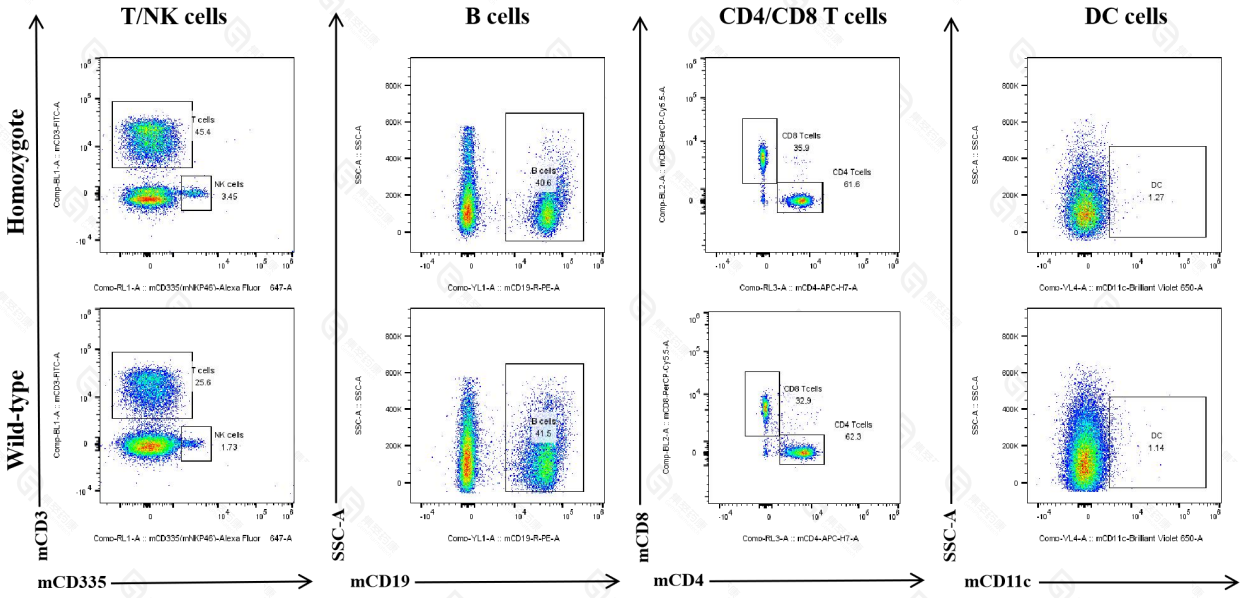
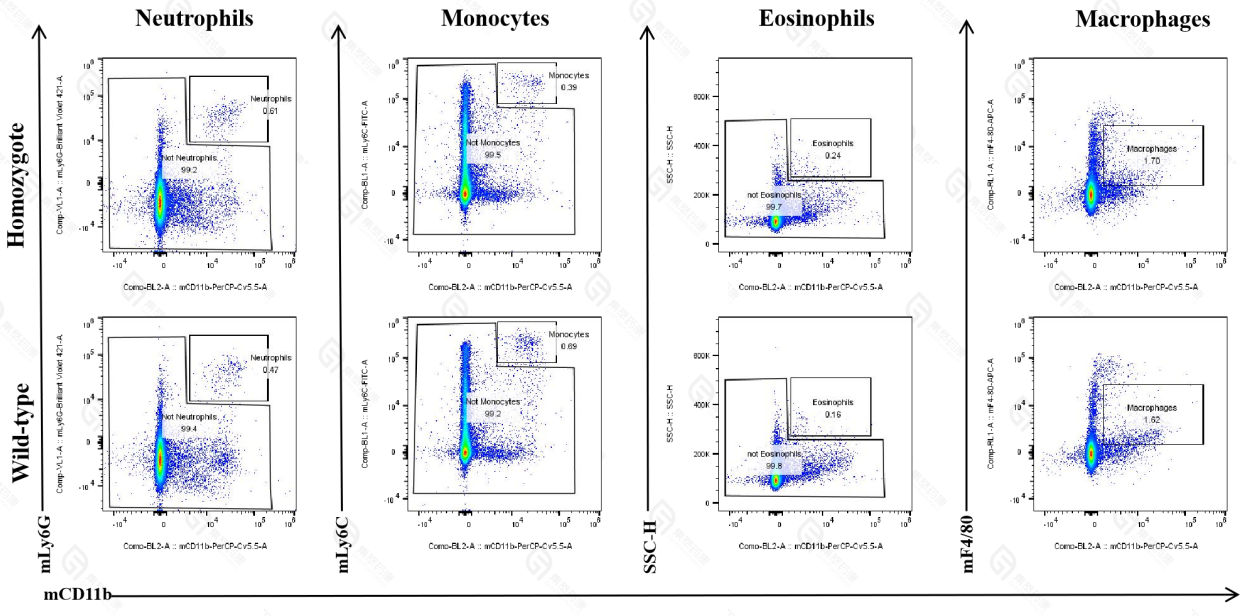


Fig 7. Immune cell subpolulation in blood of B6-hIL18R1/hIL18RAP mice.
Blood was taken from female B6 and B6-hIL18R1/hIL18RAP mice for flow cytometry analysis to assess immune cell subpopulations. The percentages of eosinophils, macrophages, NK cells, B cells, and dendritic cells in B6-hIL18R1/hIL18RAP mice were similar to those in B6, indicating that the replacement of mIL18R1/mIL18RAP by hIL18R1/hIL18RAP did not alter the development, differentiation, and distribution of these cells in blood.

6. Analysis of immune cell subpopulation in spleen



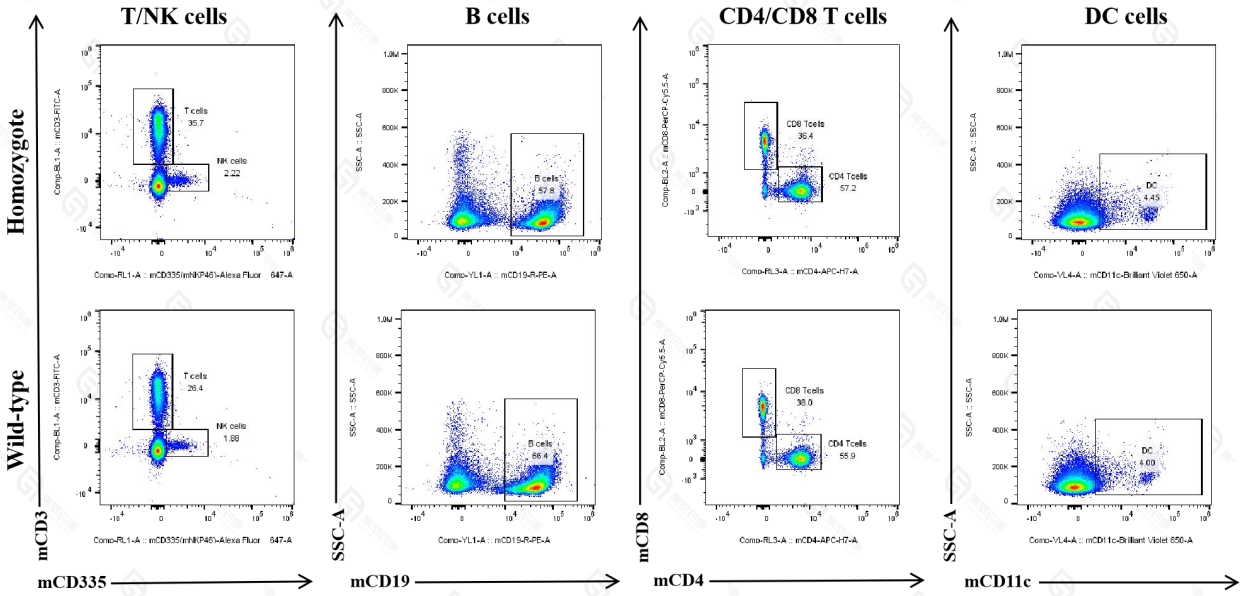


Fig 8. Immune cell subpopulation in spleen of B6-hIL18R1/hIL18RAP mice.
Splenocytes were taken from female B6 and B6-hIL18R1/hIL18RAP mice for flow cytometry analysis to assess immune cell subpopulations. The percentages of neutrophils, monocytes, eosinophils, macrophage, T cells, NK cells, B cells, and dendritic cells in B6-hIL18R1/hIL18RAP mice were similar to those in B6, indicating that the replacement of mIL18R1/mIL18RAP by hIL18R1/hIL18RAP did not alter the development, differentiation, and distribution of these cells in spleen.

References

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