

## C57BL/6JGpt-CD79a-CreERT2

**Strain Name:** C57BL/6JGpt-*CD79a*<sup>em1Cin(CreERT2)</sup>/Gpt

**Strain Type:** Knock-in

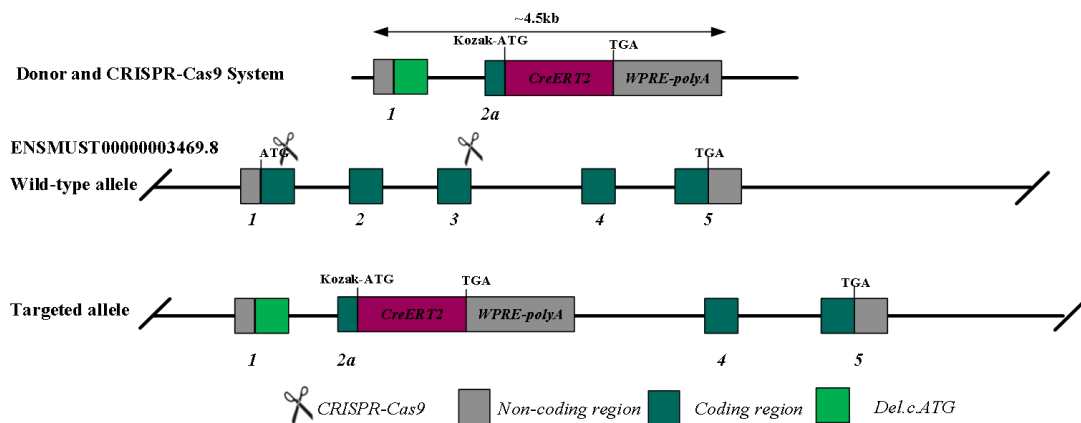
**Strain Number:** T060111

**Background:** C57BL/6JGpt

### Description

This mouse strain expresses CreERT2 inducible recombinase<sup>[1]</sup> under the control of the mouse *CD79a* endogenous promoter, exons 2 and 3 of the *Cd79a* gene have been replaced with CreERT2 and the ATG codon of exon 1 is deleted. When crossed with a strain with loxP site flanked sequence in its genome, Cre-mediated recombination will result in excision of the DNA fragment between the two loxPs in B lymphocytes after tamoxifen administration. Recombinase activity after tamoxifen induction was detected in a proportion of cells in spleen and intestine. Note: mild CreER leaky activity was also observed in some cells in spleen without tamoxifen treatment. Knock-in disrupts the expression of *Cd79a* endogenous genes, and Cre homozygous mice result in a *Cd79a* knockout phenotype, so Cre homozygous mice are not recommended.

### Strategy



**Fig.1 Schematic diagram of C57BL/6JGpt-CD79a-CreERT2 model strategy.**

## Applications

1. Cre tool mice for specific, tamoxifen dependent induction of loxP recombination in B lymphocytes [2].

## Data support

### 1. Validation methods & notes

CD79a-CreERT2 mice was crossed with CAG-loxp-ZsGreen-Stop-loxp-tdTomato mice with ubiquitous reporter expression (hereafter referred as CAG-G/R mice), Cre-mediated recombination will lead to excision of ZsGreen and the stop cassette and expression of tdTomato, thus loss of green fluorescence and gain of red fluorescence will indicate Cre activity. Fluorescence imaging of frozen sections was performed to exhibit Cre activity in various tissues and organs. Imaging sections were performed under a 200x microscopy. For tamoxifen administration, tamoxifen was treated through intraperitoneal injection daily from P44 to P50 (6.3 w~7.1 w).

Note: these results may only represent the activity of CreERT2 in this strain under this certain tamoxifen treatment condition at the identical stage. Recombinase activity may be different at other stages or under different tamoxifen induction conditions in your application.

CD79a-CreERT2 mice exhibited high CreERT2 activity in the spleen and intestine, with little or no CreERT2 activity in the heart, liver, stomach, kidneys, skeletal muscle and sperm after tamoxifen administration. It should be noted that other tissues remain untested, and the presence of CreERT2 in unexamined organs is not guaranteed.

### 2. Timeline of tamoxifen treatment and imaging

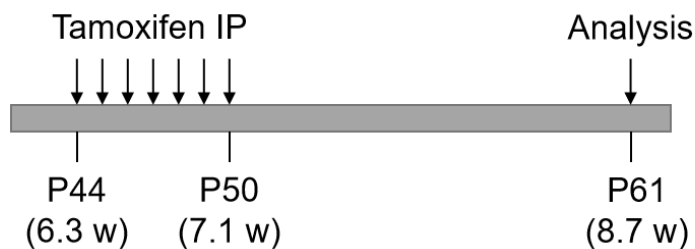
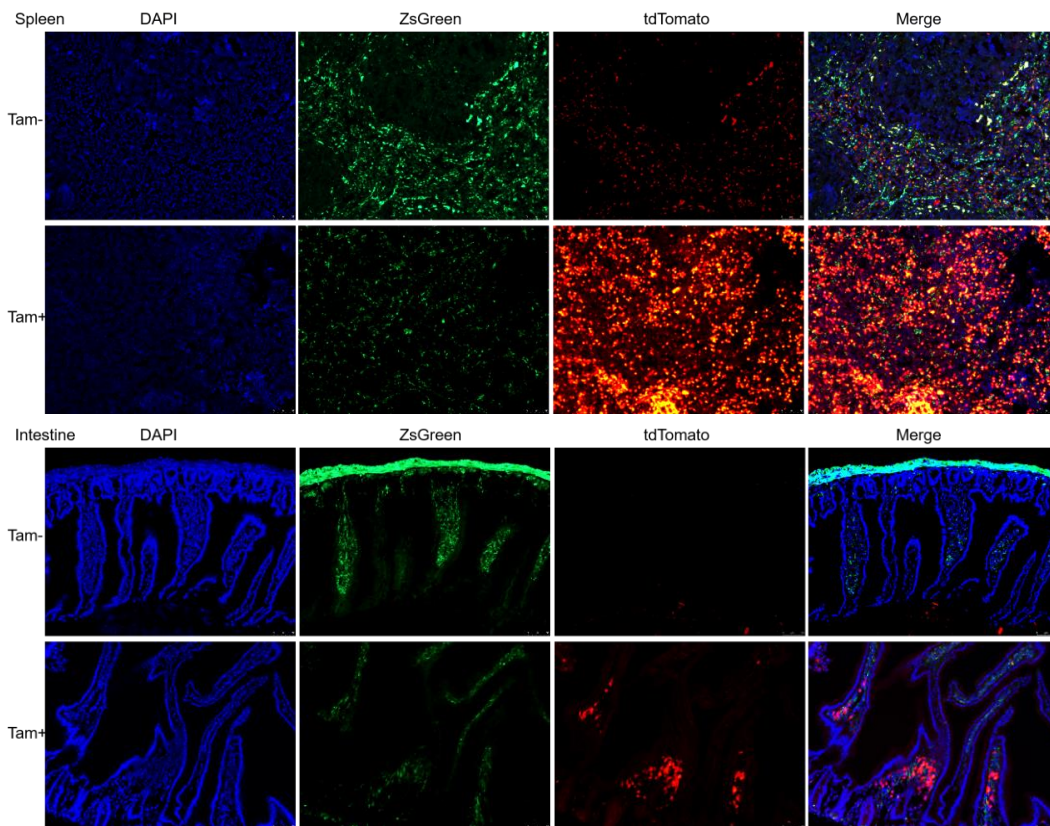


Fig 2. Timeline of tamoxifen treatment and experiment analysis of CD79a-CreERT2 mice.

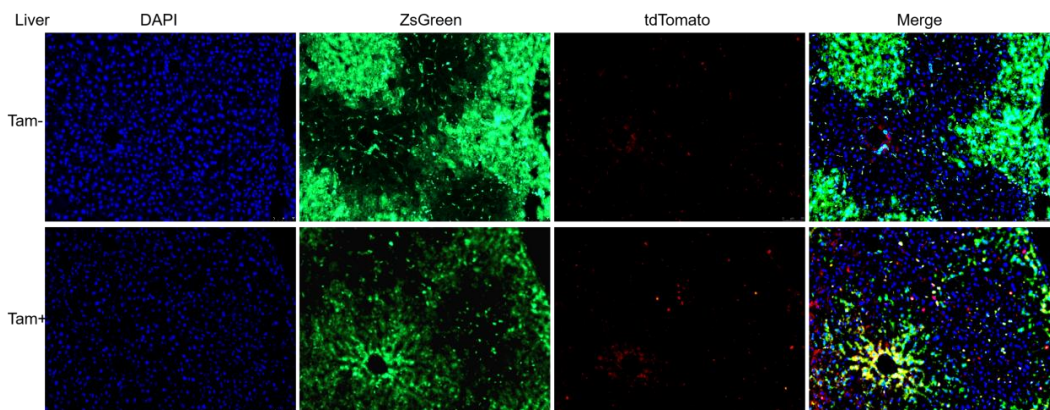
### 3. Images of tissues and organs with obvious Cre activity

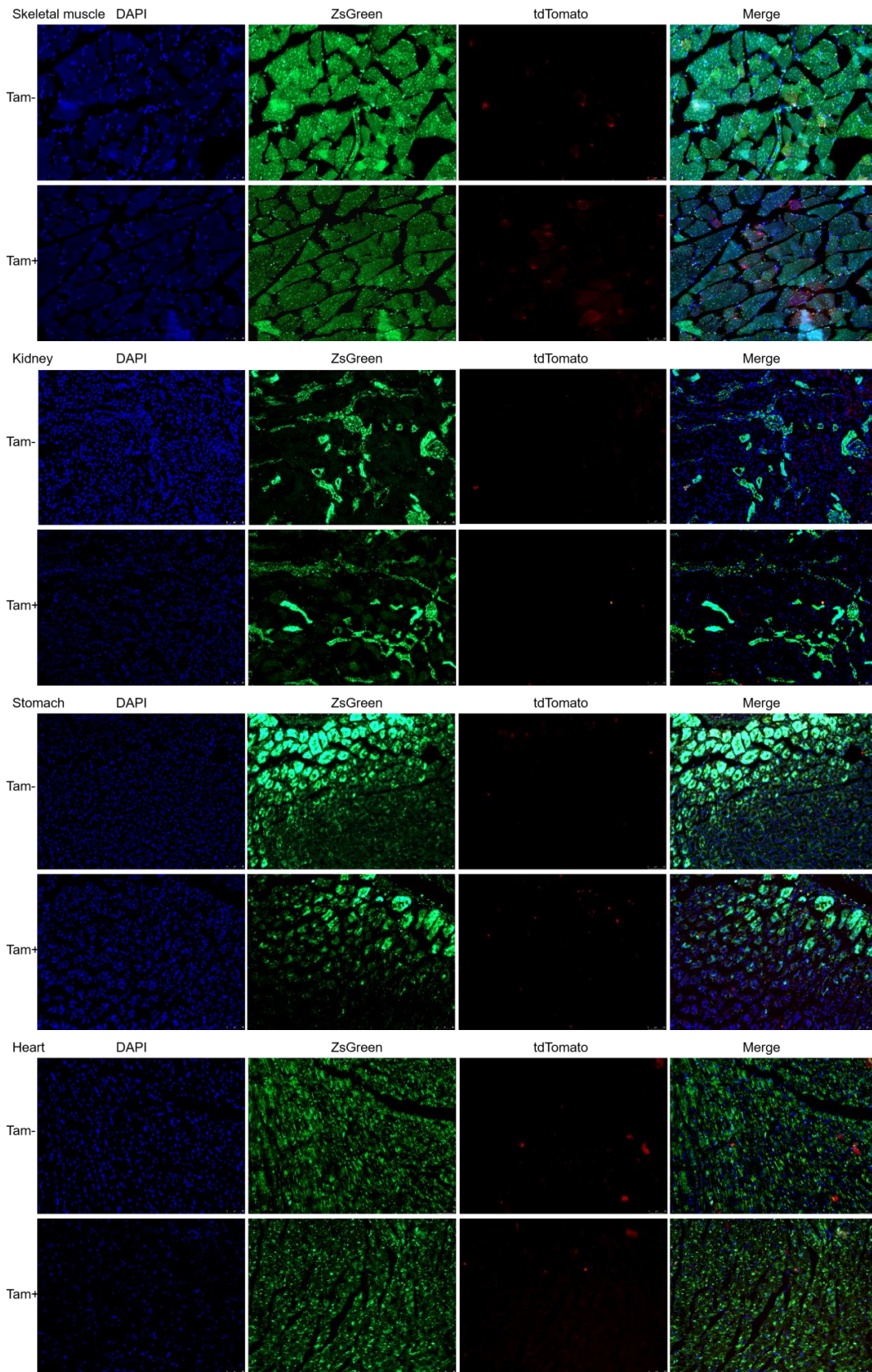


**Fig 3. Fluorescence imaging of tissues and organs with obvious Cre activity.**

Organ name was indicated in the left top of each subfigure group. Tam-: CD79a-CreERT2, CAG-G/R double positive individuals without tamoxifen administration; Tam+: CD79a-CreERT2, CAG-G/R double positive individuals with tamoxifen administration.

#### 4. Images of tissues and organs with little or no Cre activity





**Fig 4. Fluorescence imaging of tissues and organs with little or no Cre activity.**

Organ name was indicated in the left top of each subfigure group. Tam-: CD79a-CreERT2, CAG-G/R double positive individuals without tamoxifen administration; Tam+: CD79a-CreERT2, CAG-G/R double positive individuals with tamoxifen administration.

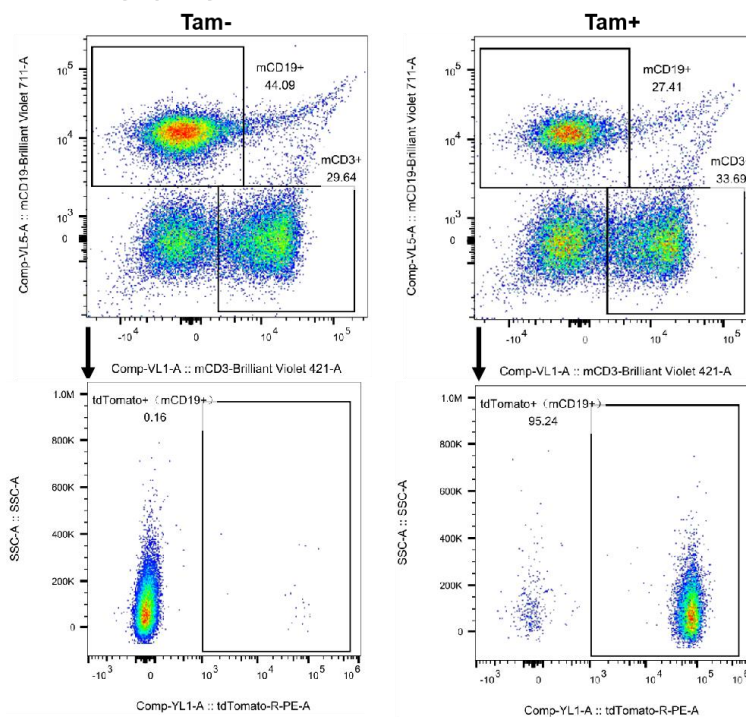
### 5. Gating Strategies for flow Cytometry

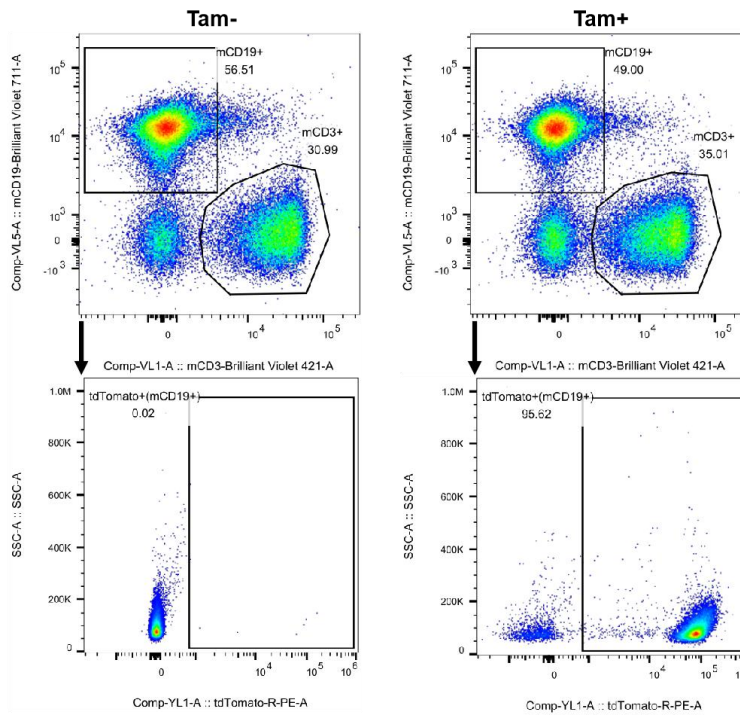
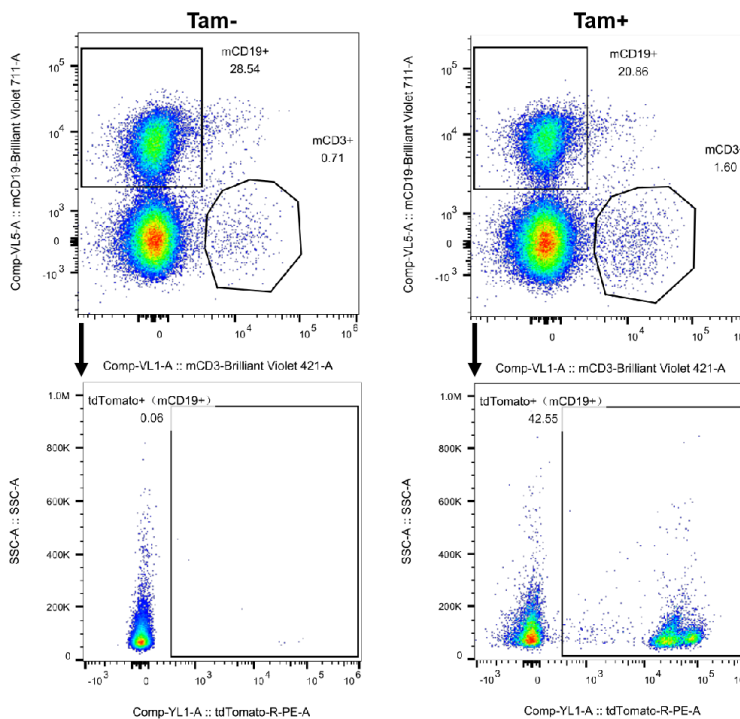
Cell population	Gating		
B lymphocytes	mCD45+	mCD19+mCD3-	B lymphocytes
Total T cells	mCD45+	mCD19-mCD3+	Total T cells

**Table 1. Gating Strategies for flow Cytometry of CD79a-CreERT2 mice.**

### 6. Flow cytometry analysis of cells with Cre activity

Blood: B lymphocytes

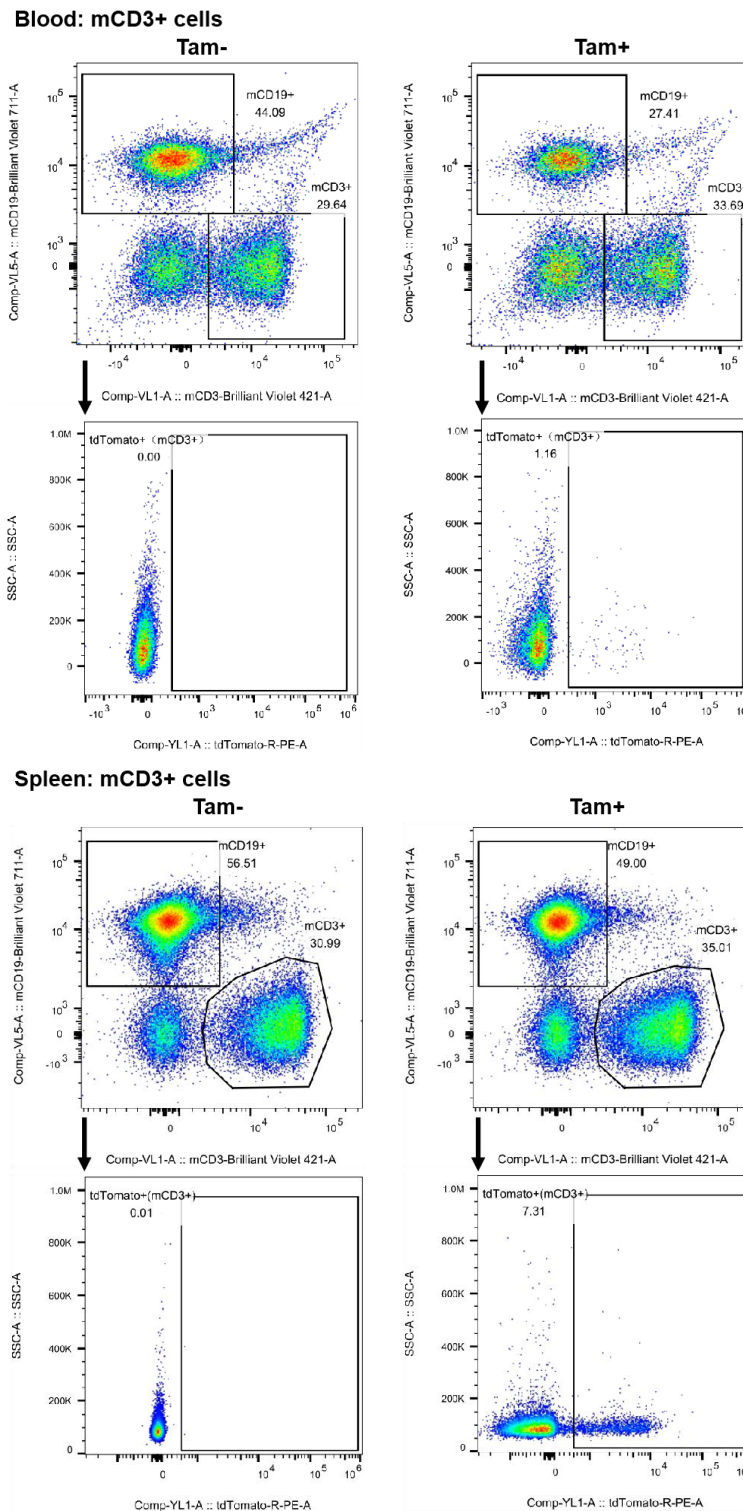


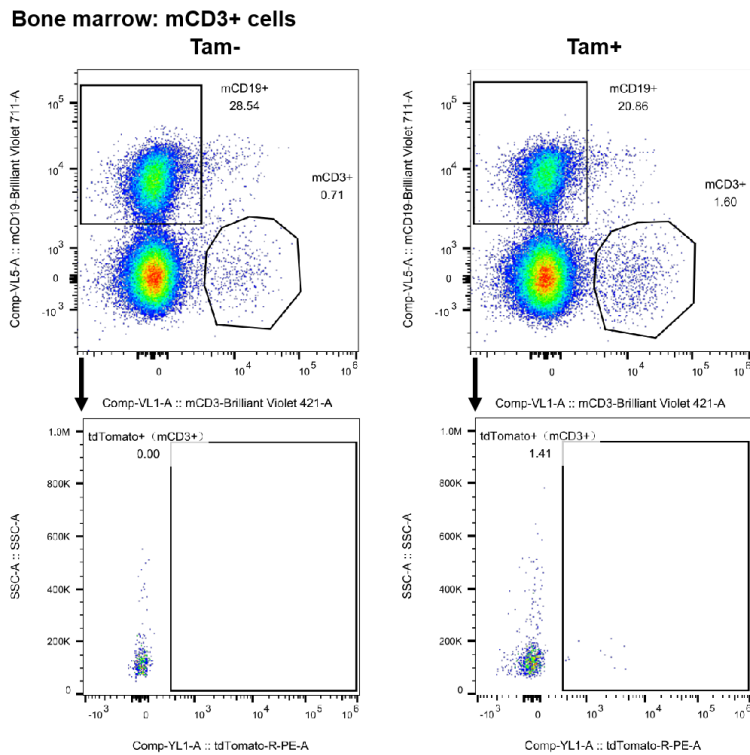
**Spleen: B lymphocytes**

**Bone marrow: B lymphocytes**

**Fig 5. Flow cytometry analysis of cells with Cre activity**

Organ name was indicated in the left top of each subfigure group. Tam-: CD79a-CreERT2, CAG-G/R double positive individuals without tamoxifen administration; Tam+: CD79a-CreERT2, CAG-G/R double positive individuals with

tamoxifen administration. Splenocytes, whole blood cells and bone marrow-derived cells were harvested from Tam- and Tam+ mice and analyzed for tdTomato expression with flow cytometry.

## 7. Flow cytometry analysis of cells with little or no Cre activity





**Fig 6. Flow cytometry analysis of cells little or no Cre activity**

Organ name was indicated in the left top of each subfigure group. Tam-: CD79a-CreERT2, CAG-G/R double positive individuals without tamoxifen administration; Tam+: CD79a-CreERT2, CAG-G/R double positive individuals with tamoxifen administration. Splenocytes, whole blood cells and bone marrow-derived cells were harvested from Tam- and Tam+ mice and analyzed for tdTomato expression with flow cytometry.

## Reference

1. Feil R, Wagner J, Metzger D, et al. "Regulation of Cre recombinase activity by mutated estrogen receptor ligand-binding domains." *Biochem Biophys Res Commun*, 1997, 237(3): 752-757.
2. Hobeika E, Thiemann S, Storch B, et al. Testing gene function early in the B cell lineage in mb1-cre mice. *Proc Natl Acad Sci U S A*, 2006, 103(37): 13789-94.
3. Hobeika E, Levit-Zerdoun E, Anastasopoulou V, et al. CD19 and BAFF-R can signal to promote B-cell survival in the absence of Syk. *EMBO J*, 2015, 34(7): 925-39.