

Supplementary Table 1. Listing of mouse strains employed.

Mouse strain	Description
<i>Foxp3</i> ^{EGFP}	Knock-In strain expressing EGFP, under the control of the <i>Foxp3</i> promoter.
<i>R26</i> ^{YFP}	Knock-In strain carrying a <i>loxP</i> -flanked STOP sequence followed by the Enhanced Yellow Fluorescent Protein gene inserted into the <i>Rosa26</i> locus.
<i>Foxp3</i> ^{EGFPCre}	Strain expressing EGFP and Cre recombinase, under the control of the <i>Foxp3</i> promoter in a bacterial artificial chromosome.
<i>Foxp3</i> ^{EGFPCre} <i>R26</i> ^{YFP}	Strain obtained by crossing <i>Foxp3</i> ^{EGFPCre} and <i>R26</i> ^{YFP} , allowing the expression of YFP in Treg and ex-Treg cells.
<i>Foxp3</i> ^{K276X}	Knock-In strain carrying a targeted mutation of the <i>Foxp3</i> gene that contains the K276X nonsense mutation (A to T substitution in the first base position of codon 276), which creates a stop codon.
<i>Foxp3</i> ^{ΔEGFPCre}	Knock-In strain carrying a bicistronic loss of function <i>Foxp3</i> allele that contained a <i>Frt</i> element inserted between exons 9 and 10 that created an aberrant 3' splice junction site leading to a premature stop codon in the <i>Foxp3</i> transcript and a ribosomal entry sequence inserted in the 3' untranslated region of <i>Foxp3</i> directed the expression of a EGFP protein and codon improved Cre recombinase (iCre).
<i>Foxp3</i> ^{ΔEGFPCre} <i>R26</i> ^{YFP}	Strain obtained by crossing <i>Foxp3</i> ^{ΔEGFPCre} and <i>R26</i> ^{YFP} , allowing the expression of YFP in ΔT _{reg} and ex-ΔT _{reg} cells.
<i>Foxp3</i> ^{ΔEGFPCre/+} <i>R26</i> ^{YFP}	Strain carrying one wild-type and one <i>Foxp3</i> ^{ΔEGFPCre} allele allowing the generation both of wild-type T _{reg} cells (<i>Foxp3</i> ⁺) and ΔT _{reg} cells (YFP ⁺) in female mice.
<i>Foxp3</i> ^{RFP}	Knock-In strain expressing RFP, under the control of the <i>Foxp3</i> promoter.
<i>Foxp3</i> ^{ΔEGFPCre/RFP} <i>R26</i> ^{YFP}	Strain obtained by crossing <i>Foxp3</i> ^{RFP} and <i>Foxp3</i> ^{ΔEGFPCre} <i>R26</i> ^{YFP} allowing the visualization both of <i>Foxp3</i> -sufficient T _{reg} cells (RFP ⁺) and ΔT _{reg} cells (YFP ⁺) in female mice.
<i>Rictor</i> ^{Flox/Flox}	<i>Rictor</i> ^{Flox/Flox} mice possess <i>loxP</i> sites flanking exon 3 of the RPTOR independent companion of MTOR, complex 2 (<i>Rictor</i>) gene.
<i>Foxp3</i> ^{ΔEGFPCre} <i>Rictor</i> ^{Δ/Δ}	Strain obtained by crossing <i>Foxp3</i> ^{ΔEGFPCre} and <i>Rictor</i> ^{Flox/Flox} , allowing the deletion of <i>Rictor</i> in ΔT _{reg} cells.
<i>Foxp3</i> ^{ΔEGFPCre} <i>Rictor</i> ^{ΔWT}	Strain obtained by crossing <i>Foxp3</i> ^{ΔEGFPCre} and one allele <i>Rictor</i> ^{Flox/Flox} , allowing the deletion of only one allele of <i>Rictor</i> in ΔT _{reg} cells.
<i>Foxp3</i> ^{ΔEGFPCre} <i>Rictor</i> ^{Δ/Δ} <i>R26</i> ^{YFP}	Strain obtained by crossing <i>Foxp3</i> ^{ΔEGFPCre} <i>Rictor</i> ^{Δ/Δ} and <i>R26</i> ^{YFP} , allowing the expression of YFP and the deletion of <i>Rictor</i> specifically in ΔT _{reg} cells.
<i>Foxp3</i> ^{ΔEGFPCre/+} <i>Rictor</i> ^{Δ/Δ} <i>R26</i> ^{YFP}	Strain carrying one wild-type and one <i>Foxp3</i> ^{ΔEGFPCre} allele allowing the generation both of wild-type T _{reg} cells (<i>Foxp3</i> ⁺) and <i>Rictor</i> -deficient ΔT _{reg} cells (YFP ⁺) in female mice.
<i>Rptor</i> ^{Flox/Flox}	Knock In strain carrying a <i>loxP</i> sites flanking exon 6 of the targeted <i>Rptor</i> (regulatory associated protein of MTOR, complex 1) gene.
<i>Foxp3</i> ^{ΔEGFPCre} <i>Rptor</i> ^{Δ/Δ}	Strain obtained by crossing <i>Foxp3</i> ^{ΔEGFPCre} and <i>Rptor</i> ^{Flox/Flox} , allowing the deletion of <i>Rptor</i> in ΔT _{reg} cells.
<i>Foxp3</i> ^{ΔEGFPCre} <i>Rptor</i> ^{ΔWT}	Strain obtained by crossing <i>Foxp3</i> ^{ΔEGFPCre} and one allele <i>Rptor</i> ^{Flox/Flox} , allowing the deletion of only one <i>Rptor</i> allele in ΔT _{reg} cells.
<i>Foxp3</i> ^{ΔEGFPCre} <i>Rictor</i> ^{Δ/Δ} <i>Rptor</i> ^{Δ/Δ}	Strain obtained by crossing <i>Foxp3</i> ^{ΔEGFPCre} , <i>Rictor</i> ^{Flox/Flox} and <i>Rptor</i> ^{Flox/Flox} , allowing the deletion of <i>Rictor</i> and <i>Rptor</i> in ΔT _{reg} cells.
<i>Foxo1</i> ^{Flox/Flox}	Knock-In strain carrying a <i>loxP</i> sites flanking exon 2 of the forkhead box O1 (<i>Foxo1</i>) gene.
<i>Foxp3</i> ^{ΔEGFPCre} <i>Foxo1</i> ^{Δ/Δ}	Strain obtained by crossing <i>Foxp3</i> ^{ΔEGFPCre} and <i>Foxo1</i> ^{Flox/Flox} allowing the deletion of <i>Foxo1</i> in ΔT _{reg} cells.
<i>Foxp3</i> ^{ΔEGFPCre} <i>Rictor</i> ^{Δ/Δ} <i>Foxo1</i> ^{Δ/Δ}	Strain obtained by crossing <i>Foxp3</i> ^{ΔEGFPCre} , <i>Rictor</i> ^{Flox/Flox} and <i>Foxo1</i> ^{Flox/Flox} , allowing the deletion of <i>Rictor</i> and <i>Foxo1</i> in ΔT _{reg} cells.
<i>R26</i> ^{Foxo1AAA}	Knock-In strain carrying a <i>loxP</i> -flanked STOP sequence followed by the a modified <i>Foxo1</i> gene inserted into the <i>Rosa26</i> locus, allowing the expression of unsensitized to phosphorylation and constitutively active <i>Foxo1</i> in cells expressing Cre recombinase.
<i>Foxp3</i> ^{ΔEGFPCre} <i>R26</i> ^{Foxo1AAA}	Strain obtained by crossing <i>Foxp3</i> ^{ΔEGFPCre} and <i>R26</i> ^{Foxo1AAA} allowing the expression of the constitutively active <i>Foxo1</i> specifically in ΔTreg cells.
<i>Pfkfb3</i> ^{Flox/Flox}	Knock-In strain carrying a <i>loxP</i> sites flanking exon of the <i>pfkfb3</i> gene, encoding for 6-Phosphofructo-2-Kinase/Fructose-2,6-Biphosphatase 3, a powerful stimulator a glycolysis.
<i>Foxp3</i> ^{ΔEGFPCre} <i>Pfkfb3</i> ^{Δ/Δ}	Strain obtained by crossing <i>Foxp3</i> ^{ΔEGFPCre} and <i>Pfkfb3</i> ^{Flox/Flox} allowing the deletion of <i>Pfkfb3</i> specifically in ΔTreg cells.
<i>Foxp3</i> ^{ΔEGFPCre} <i>Rictor</i> ^{Δ/Δ} <i>Pfkfb3</i> ^{Δ/Δ}	Strain obtained by crossing <i>Foxp3</i> ^{ΔEGFPCre} , <i>Rictor</i> ^{Flox/Flox} and <i>Pfkfb3</i> ^{Flox/Flox} allowing the concurrent deletion of <i>Rictor</i> and <i>Pfkfb3</i> specifically in ΔTreg cells.
<i>Foxp3</i> ^{YFPCre}	Knock-In strain expressing YFP and Cre recombinase under the control of the <i>Foxp3</i> promoter.

<i>Foxp3</i> ^{YFP^{Cre}} <i>Rictor</i> ^{Δ/Δ}	Strain obtained by crossing <i>Foxp3</i> ^{YFP^{Cre}} and <i>Rictor</i> ^{Flox/Flox} allowing the concurrent deletion of Rictor specifically in Foxp3-sufficient Treg cells.
CD45.1 <i>Foxp3</i> ^{EGFP}	Strain expressing the congenic marker CD45.1 in hematopoietic cell lineage and EGFP in T _{reg} cells
<i>CD4</i> ^{Cre}	Knock-In strain expressing Cre recombinase under the control of the CD4 promoter.
<i>CD4</i> ^{Cre} <i>Rictor</i> ^{Δ/Δ}	Strain obtained by crossing <i>CD4</i> ^{Cre} and <i>Rictor</i> ^{Flox/Flox} allowing the deletion of Rictor specifically in CD4 and CD8 cells in periphery.

Supplementary Table 2. Listing of mouse antibodies used.

Marker	Fluorochrome	clone	compagny	dilution	Staining
Foxp3	eF450	FJK-16S	eBioscience	1/500	Intracellular
	PE	NRRF-30	eBioscience	1/500	Intracellular
Gata-3	eF660	TWAJ	eBioscience	1/300	Intracellular
ROR γ t	PE	B2D	BD	1/200	Intracellular
T-Bet	Percp-Cy5.5	eBio4B10	eBioscience	1/300	Intracellular
Blimp-1	PE	5E7	Biologend	1/300	Intracellular
CTLA4	PE	UC10-4B9	eBioscience	1/300	Intracellular
Helios	APC	22F6	eBioscience	1/300	Intracellular
IFN- γ	APC	XMG1.2	Biologend	1/500	Intracellular
IL-17A	PE-Cy7	TC11-18H10.1	Biologend	1/500	Intracellular
	PE	TC11-18H10.1	Biologend	1/500	Intracellular
IL-4	PE	11B11	Biologend	1/300	Intracellular
IL-10	AF700	JES5-16E3	eBioscience	1/300	Intracellular
CD4	BV605	RM4-5	Biologend	1/500	Surface
	PE	RM4-5	Biologend	1/500	Surface
CD8	PE	53-6.7	eBioscience	1/500	Surface
CD62L	APC	MEL-14	Biologend	1/300	Surface
CD44	eF450	IM7	eBioscience	1/300	Surface
CD90.2	APC-Cy7	30-H12	Biologend	1/500	Surface
CD25	PE-Cy7	PC61	Biologend	1/500	Surface
ICOS	PE	7E.17G9	eBioscience	1/300	Surface
Nrp1	PE	3DS304M	eBioscience	1/300	Surface
GITR	PE	DTA-1	eBioscience	1/300	Surface
CD73	APC	TY/11.8	Biologend	1/300	Surface
CD45RB	APC	C363.16A	eBioscience	1/500	Surface
CD45.1	PE-Cy7	A20	Biologend	1/500	Surface
CD45.2	AF700	104	Biologend	1/500	Surface
Viability dye	eFluor506		Biologend	1/1000	Surface
p _{T308} AKT	PE		BD	1/100	Post-Methanol
p _{S473} AKT	PE	D9E	Cell signaling	1/100	Post-Methanol
pS6	PE	cupk43k	eBioscience	1/100	Post-Methanol
p4EBP	eF660	V3NTY24	eBioscience	1/100	Post-Methanol

Supplementary Table 3. Listing of human antibodies used.

Marker	Fluorochrome	clone	compagny	dilution	Staining
FOXP3	BV421	206D	Biologend	1/300	Intracellular
CTLA-4	PE	14D3	eBioscience	1/300	Intracellular
HELIOS	APC	22F6	eBioscience	1/300	Intracellular
IFN- γ	APC	4S.B3	Biologend	1/300	Intracellular
IL-4	PE	8D4-8	Biologend	1/300	Intracellular
CD4	Percp-Cy5.5	RPA-T4	eBioscience	1/300	Surface
CD127	PE-Cy7	A019D5	Biologend	1/500	Surface
CD25	FITC	2A3	BD	1/100	Surface
p473AKT	PE	D9E	Cell signaling	1/100	Post-Methanol

Supplementary Table 4. Listing of immunoblotting antibodies used.

Marker	Source	clone	compagny	dilution
Foxp3	Rabbit	Polyclonal	MBL	1/500
Rictor	Rabbit	D16H9	Cell signaling	1/1000
Pten	Rabbit	D4.3	Cell signaling	1/1000
Phlpp1	Rabbit	Polyclonal	ThermoFisher	1/500
pY ³¹⁹ -ZAP70	Rabbit	Polyclonal	Cell signaling	1/1000
ZAP70	Rabbit	99F2	Cell signaling	1/1000
β -actin	Rabbit	13E5	Cell signaling	1/5000
Rabbit IgG-HRP	Goat	RPA-T4	Cell signaling	1/5000