

SUPPLEMENTAL MATERIAL

Bao et al., <http://dx.doi.org/10.1084/jem.20160438>

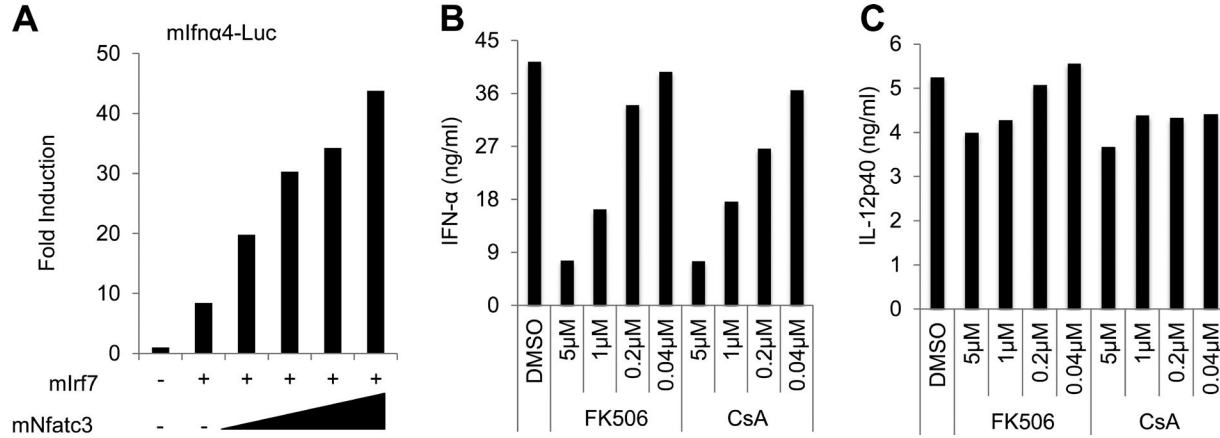


Figure S1. **Mouse Nfatc3 enhances mouse Ifn- α 4 transcriptional activity, and inhibition of Nfatc3 reduces CpG DNA-induced IFN- α in mouse primary pDCs.** (A) HEK-293T cells were transfected with 100 ng mouse IFNA4 luciferase reporter plasmid (mlfna4-Luc) and 0.5 ng mouse Irf7 expression vector together with an increasing amount of mouse NFATC3 expression vector (25, 50, and 100 ng). 0.1 ng renilla luciferase reporter plasmid was transfected simultaneously as an internal control. Results are presented as fold induction relative to the activity of renilla luciferase. (B and C) ELISA of the production of IFN- α and IL-12p40 by mouse bone marrow pDCs stimulated with 1 μ M CpG A for 24 h together with FK506, Cs A, or vehicle (DMSO). Data are representative of two independent experiments.

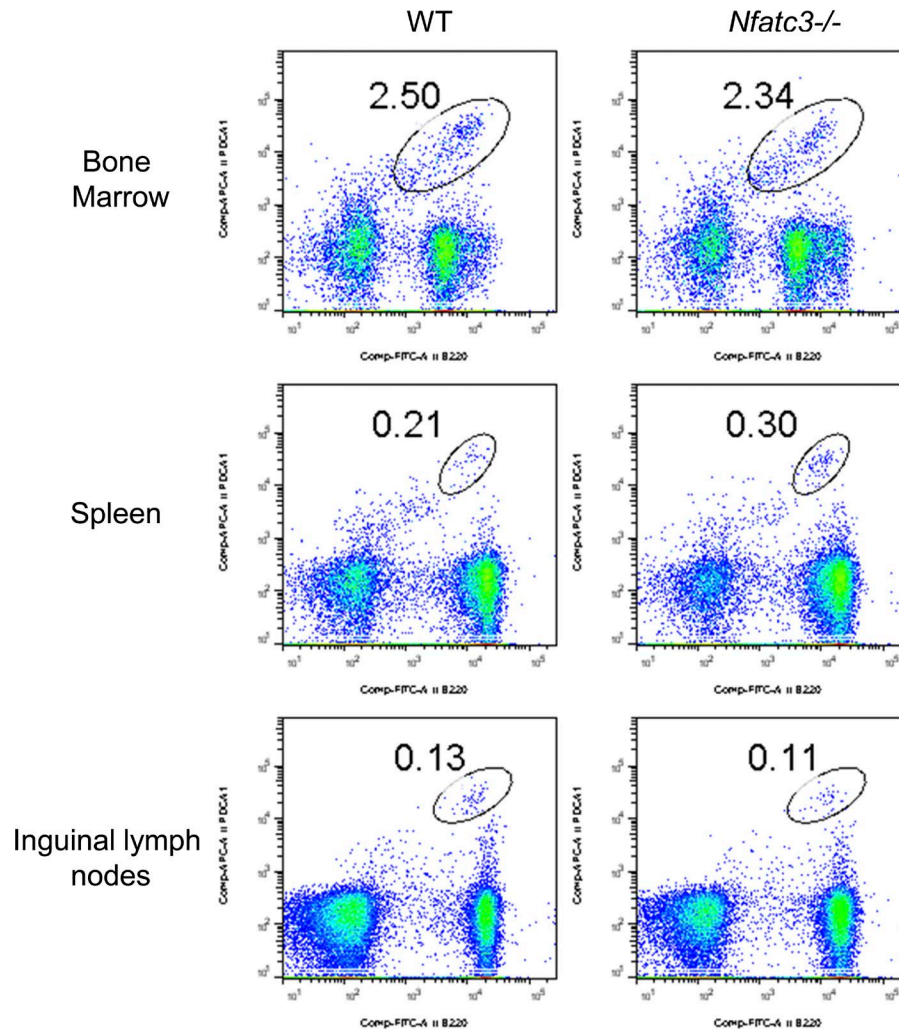


Figure S2. ***Nfatc3* deficiency does not affect pDC differentiation.** Total bone marrow cells, splenocytes, and inguinal lymphocytes were isolated. pDC (B220⁺PDCA1⁺) frequencies were analyzed by FACS.

Table S1. Primers used for real-time PCR

Gene	Sequence (5'-3')
Ifna1 forward	TCAGTCTTCCCAGCACATTG
Ifna1 reverse	GAGAAGAAACACAGCCCTG
Ifna2 forward	CATTCCAAGCAGCAGATGAA
Ifna2 reverse	AGCAGATCCAGAAGGCTCAA
Ifna4 forward	TATGTCCTCACAGCCAGCAG
Ifna4 reverse	TTCTGCAATGACCTCCATCA
Ifna5 forward	TGACCTCAAAGCCTGTGTGATG
Ifna5 reverse	AAGTATTTCTCACAGCCAGCAG
Ifna6 forward	TCAGGGGAAGTGCTGTATC
Ifna6 reverse	TGGAATGCAACCCCTCTAGA
Ifnb1 forward	AGCTCCAAGAAAGGACGAACAT
Ifnb1 reverse	GCCCTGTAGGTGAGTTGATCT
Irf7 forward	ACAGCACAGGCGTTTTATC
Irf7 reverse	TCCCGGCTAAGTTCGTACAC
Il12a forward	GCTTCTCCACAGGAGGTTT
Il12a reverse	CTAGACAAGGCATGCTGGT
Actb forward	GATCATTGCTCCTCCTGAGC
Actb reverse	ACATCTGCTGGAAGGTGGAC

Table S2. Biotin-labeled oligonucleotides used for pull-down assay

Promoter	Sequence (5'-3')
Bio-IFNA1 forward	/5'Biotin/GAAAGCAAAAACAGAAATGGAAAGTGGCCAGAACATTAAAGAAAGTGGAAATCAG
Bio-IFNA1 reverse	/5'Biotin/CTGATTTCCACTTTCTTAATGCTTCTGGCCACTTCCATTCTGTTTTGCTTTC
Bio-IFNA2 forward	/5'Biotin/GAAAGCAAAAAGAGAAGTAGAAAGTAACACAGGGCATTGGAAAAATGT
Bio-IFNA2 reverse	/5'Biotin/ACATTTTCCAAATGCCCTGTGTTACTTTCTACTTCTCTTTTTGCTTTC
Bio-IFNA6 forward	/5'Biotin/GAAAGCAAAAATCAGACGTAGAAAGTAAATTCTGAAAATGAAAACCTAG
Bio-IFNA6 reverse	/5'Biotin/CTAGTTTCCATTTTCAGAATTTACTTTCTACGTCTGATTTTGCTTTC
Bio-IFNA13 forward	/5'Biotin/GAAAGCAAAAACAGAAATGGAAAGTGGTCCAGAACATTAAAGAAAGTGGAAATCAG
Bio-IFNA13 reverse	/5'Biotin/CTGATTTCCACTTTCTTAATGCTTCTGGACCACTTCCATTCTGTTTTGCTTTC
Bio-IFNB1 forward	/5'Biotin/ACATAGGAAAACCTGAAAGGGAGAAGTGAAGTGGGAAATTCCTCTG
Bio-IFNB1 reverse	/5'Biotin/CAGAGGAATTTCCCCTTTCACTTCTCCCTTTCAGTTTTCTATGT
Bio-control forward	/5'Biotin/CTAAGCAAAAACAGAGATACTAAGTACAACCTAGGAATTTAGAAAATCCTTATTAG
Bio-control reverse	/5'Biotin/CTAATAAGGATTTTCTAAATCCCTAGTTGTACTTAGTATCTCTGTTTTGCTTAG