

Fig. S7

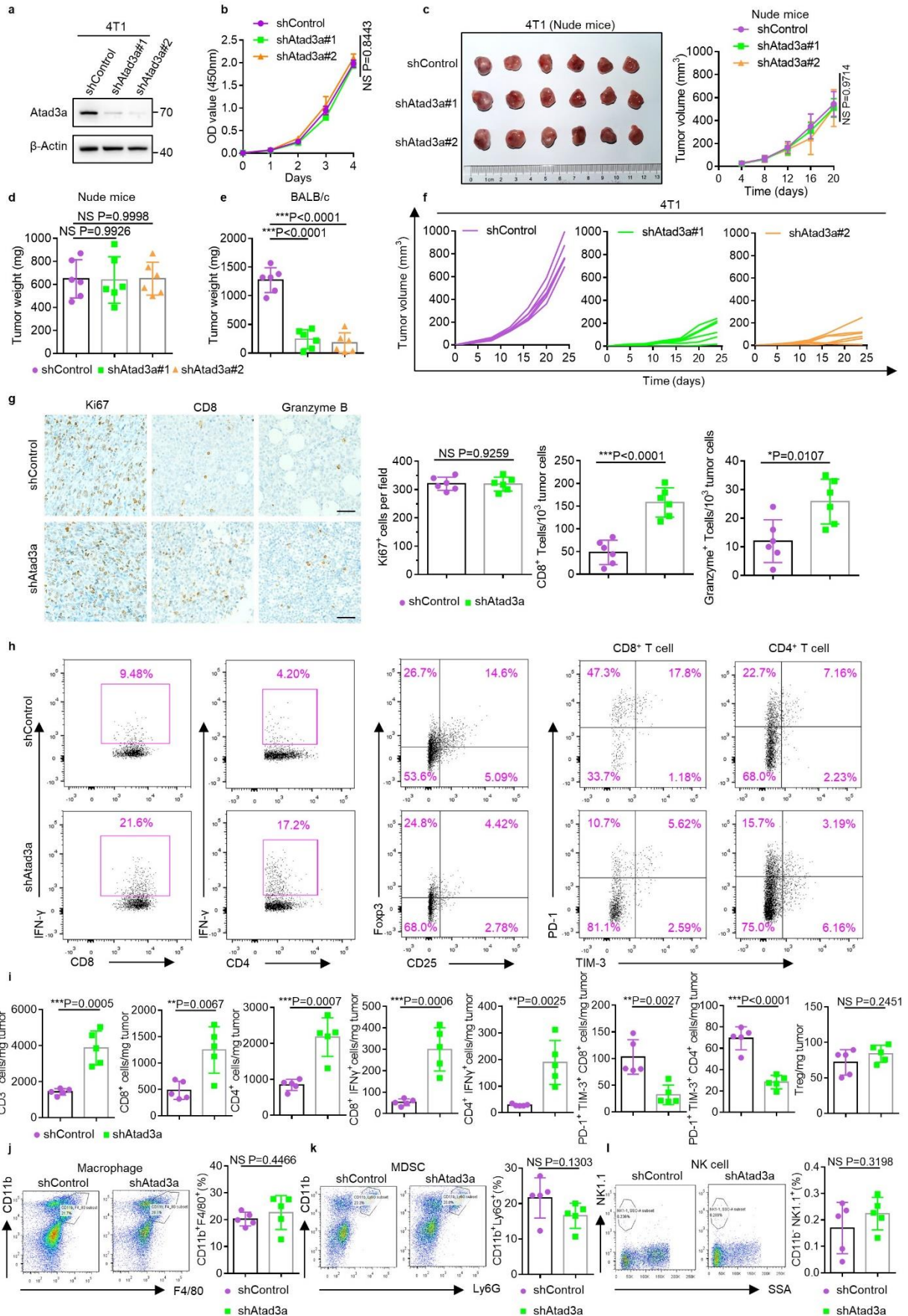


Fig. S7 Depletion of Atad3a in tumor cells suppresses tumor growth in vivo by facilitating anti-tumor immune response.

a 4T1 mouse breast cancer cells were transfected with control shRNA (shControl) or shRNA for Atad3a (shAtad3a#1 and shAtad3a#2). Immunoblot of Atad3a in 4T1 cells. **b** Proliferation of control and Atad3a-knockdown 4T1 cells measured by CCK-8 (Cell Counting Kit-8) assay ($n = 5$, one-way ANOVA). **c** BALB/c nude mice were inoculated orthotopically with 1×10^5 4T1 cells transfected with control shRNA (shControl) or shRNA for Atad3a (shAtad3a#1 and shAtad3a#2). The endpoint images (left) and volume (right) of tumors formed by control and Atad3a-knockdown cells in BALB/c nude mice ($n = 6$, one-way ANOVA). **d** The weight of 4T1 tumors formed by control and Atad3a-knockdown cells in nude mice at the end point ($n = 6$, one-way ANOVA). **e–l** BALB/c mice were inoculated orthotopically with control or Atad3a-knockdown 4T1 tumor cells. **e, f** The weight at the end point (**e**) and growth curves (**f**) of tumors formed by control and Atad3a-knockdown cells ($n = 6$, one-way ANOVA). **g** Left, IHC staining of Ki67, CD8 and granzyme B on serial sections of the tumors formed by control and Atad3a-knockdown cells. Right, quantification of the numbers of Ki67⁺ tumor cells, tumor-infiltrating CD8⁺ T cells and granzyme B secreting cells ($n = 6$, *t*-test). **h** Flow cytometry of tumor-infiltrating IFN γ ⁺CD8⁺, IFN γ ⁺CD4⁺, CD4⁺CD25⁺Foxp3⁺, PD-1⁺TIM-3⁺CD8⁺ and PD-1⁺TIM-3⁺CD4⁺ T cells in tumors formed by control and Atad3a-knockdown 4T1 cells. **i** Quantification of the numbers of tumor-infiltrating CD3⁺, CD8⁺, CD4⁺, IFN γ ⁺CD8⁺, IFN γ ⁺CD4⁺, PD-1⁺TIM-3⁺CD8⁺, PD-1⁺TIM-3⁺CD4⁺ T cells and CD4⁺CD25⁺Foxp3⁺ T_{reg} cells per

milligram of tumor tissue in control and Atad3a-knockdown 4T1 tumors by flow cytometry ($n = 5$, t -test). **j–l** Flow cytometry (left) and quantification (right) of the percentages of tumor-infiltrating macrophages (**j**), myeloid-derived suppressor cells (MDSCs) (**k**) and natural killer cells (NKs) (**l**) in 4T1 tumors formed by control and Atad3a-knockdown cells in BALB/c mice ($n = 5$, t -test). Data are representative of two independent experiments and were shown as means \pm SD.