Figure S1

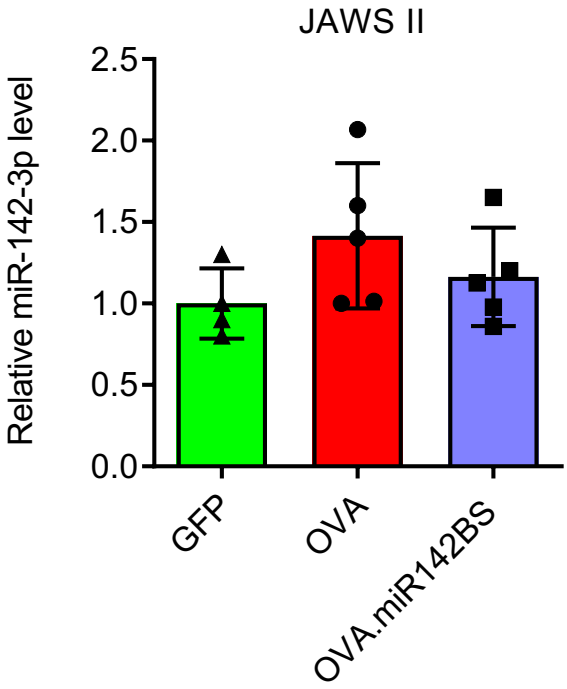

Figure S1. Endogenous miR-142-3p expression levels in JAWS II cells as measured by RT-qPCR. Cells were transiently transfected with pAAV1.EGFP, pAAV1.OVA and pAAV1.OVA.miR142BS plasmids**.** 72 hours post-transfection, cells were harvested for RNA and miR-142-3p was detected by RT-qPCR by ddCT with miR-103-3p transcripts as normalization control. Bar graphs represent mean ± SD (n = 5).

Figure S2

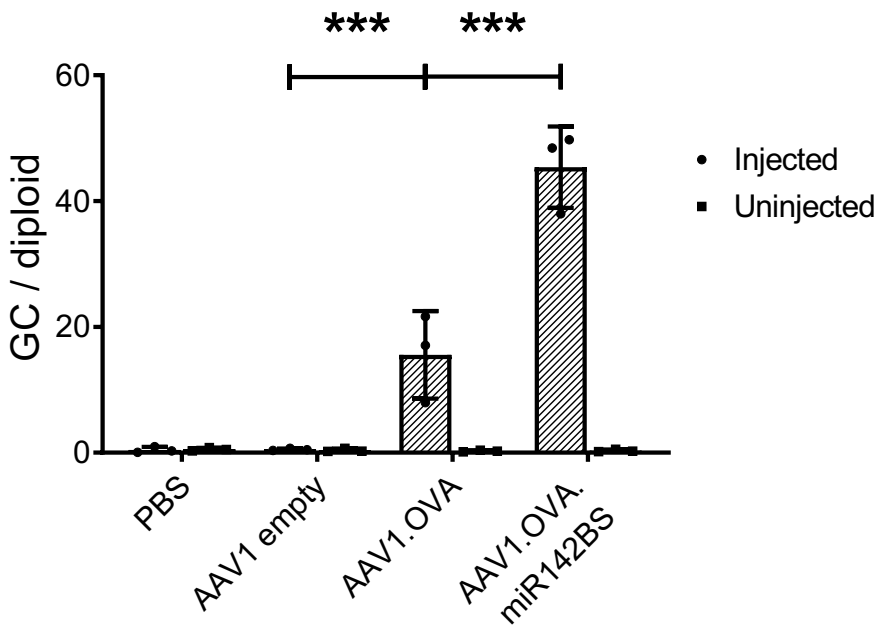

Figure S2. Inclusion of miR142BS elements into the OVA transgene results in high levels of vector genomes detected in treated muscles. qPCR detection of rAAV vector genome copies in injected and contra-lateral uninjected TA muscles, harvested 12 weeks post-injection. Bar graphs represent mean ± SD (n = 3). p values determined by ANOVA with Sidak's post-hoc test. *p < 0.01, **p < 0.01.

Figure S3

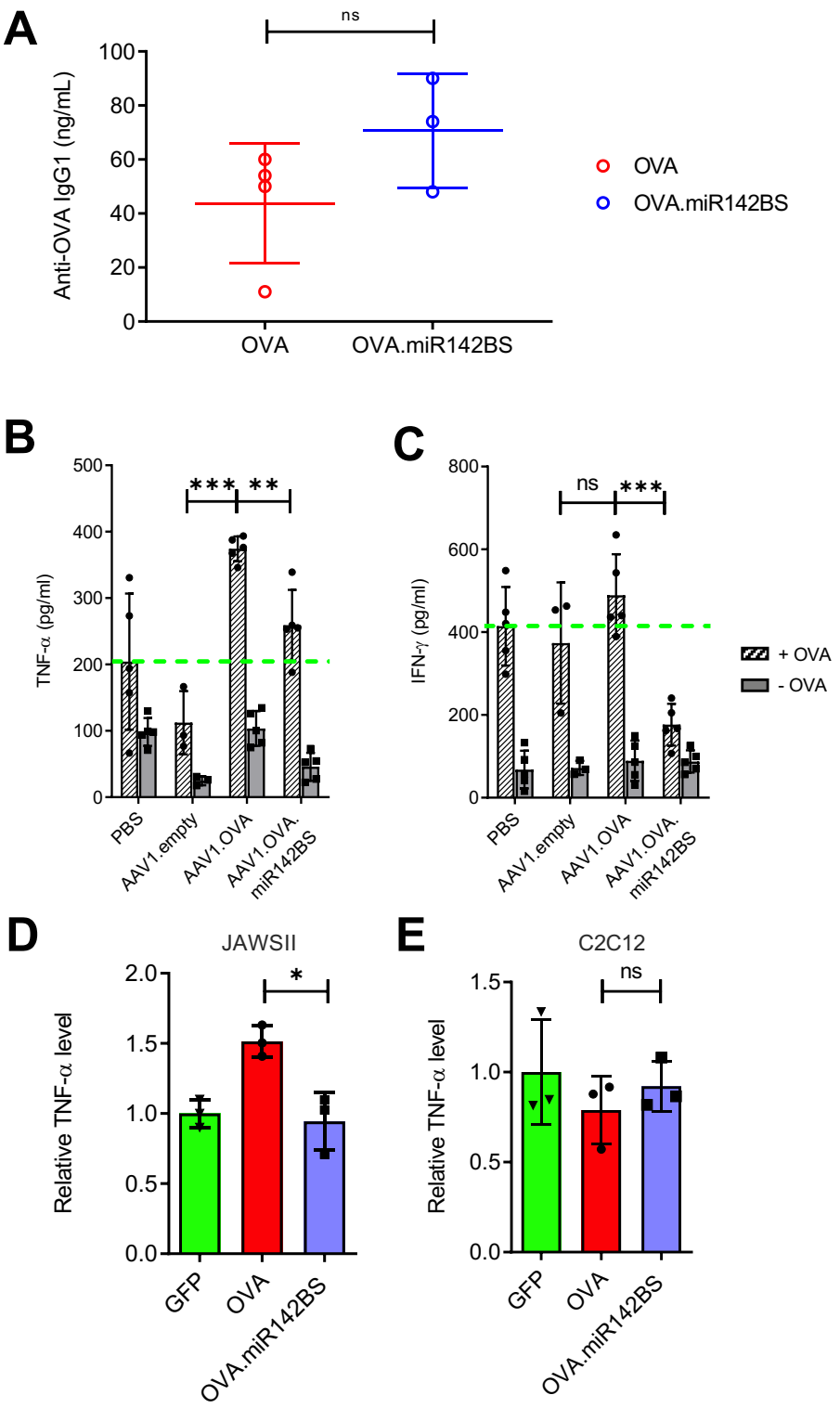

Figure S3. OVA-induced upregulation and miR142BS-mediated attenuation of TNF-α expression is due to dendritic cell populations and not myoblasts. (A) b2m KO mice were intramuscularly injected with OVA or OVA.miR142BS vectors. Five weeks post-treatment, serum was collected and assayed for anti-OVA IgG1 by ELISA. No difference between treatment groups were observed; unpaired t test; n = 4, for each group. Mean ± SD. **(B-C)** Assessment of IFN-γ and TNF-α response to OVA protein (+OVA, 5 µg/mL) by splenocytes isolated from mice four weeks post-vector injection. Three days after treatment, supernatants were collected and quantitated by ELISA (mean ± SD, n = 5). p values determined by ANOVA with Tukey's post-hoc test) -OVA, mock treatment. **D-E)** JAWS II DCs and C2C12 myoblasts were transfected with plasmids encoding GFP, OVA, or OVA.miR142BS. Two days after transfection, the cells were harvested for total RNA isolation. TNF-α mRNA levels in JAWS II **(D)** and C2C12 **(E)** cells were determined by RT-qPCR. Dashed green lines indicate baseline activation values. Bar graphs represent mean ± SD (n = 3). p values determined by ANOVA with Tukey's post-hoc test *p < 0.05, **p < 0.01, ***p < 0.001.

Figure S4

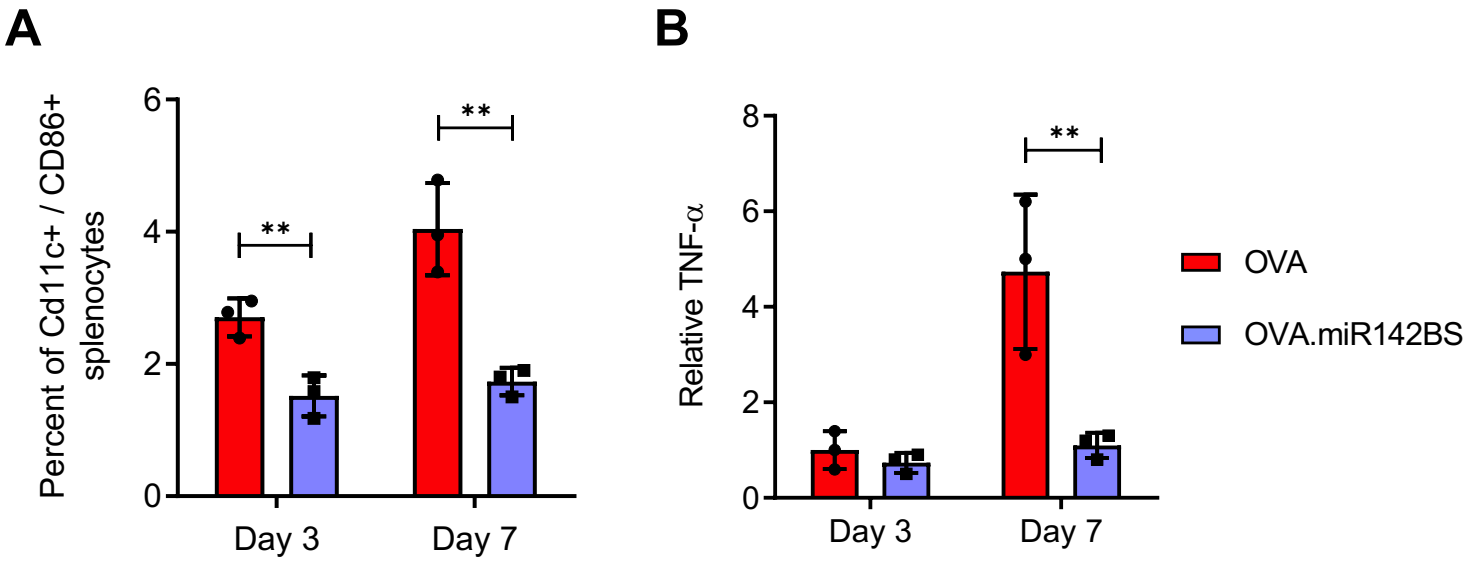

Figure S4. miR142BS incorporation suppresses transgene immunity by reduction of activated T cells and inflammatory cytokines. (A) Six-week-old C57BL/6 male mice were injected intramuscularly with PBS, rAAV1.OVA, rAAV1.OVA.miR142BS, or AAV1 empty vector (1×10^{11} GC/mouse, n = 3). Quantification of CD11c+/CD86+ splenocytes harvested 4 or 12 weeks after vector administration by flow cytometry. **(B)** Quantitation of TNF-α mRNA levels in whole spleens by RT-qPCR. **p < 0.01, unpaired t test (n = 3).