Supplementary Material

Supplemental Figure 1. Normal histology of salivary glands in poly(I:C) treated mice. Representative photomicrographs showing histology of submandibular salivary glands from mice treated with four injections of (A) poly(I:C) and (B) PBS. Mice were euthanized on day 8 and submandibular glands were collected in 10% buffered formalin. Tissue was processed for histology using standard techniques and embedded in paraffin and sections stained with H & E. Note salivary glands from both groups show normal glandular architecture. Inflammatory foci were not seen in any of the mice (magnification 20X).

Supplemental Figure 2. Poly(I:C) treatment does not cause AQP5 redistribution within the submandibular glands. Representative picture of immunohistochemical staining with polyclonal antibody to AQP5 in salivary glands from an NZB/W F1 mouse treated with poly(I:C). (A) The distribution of AQP5 appeared normal with positive staining of lateral and apical regions of acinar cells (indicated by arrows). The cells of the granular convoluted tubules and ducts did not stain. (B) An adjacent section was stained with rabbit polyclonal antibody and served as a negative control (magnification 40X).

Supplemental Figure 3. Autoantibodies do not seem to play a critical role in salivary gland hypofunction caused by poly(I:C) treatment. Mice (5 per group) were treated either with poly(I:C) or PBS every other day for 2 weeks. Mice were bled at different time points and sera analyzed for presence of autoantibodies by western blot.

WEHI 7.1 cell extract was separated by SDS-PAGE and transferred to nitrocellulose paper. Strips of nitrocellulose paper were blocked in PBS containing 5% milk for 2 hours. Strips were probed with sera diluted (1:100) in PBS containing 0.1% Tween-20 (PBST) and 5% milk protein, overnight at 4°C. Strips were incubated with HRP conjugated goat anti-mouse IgG antibody diluted in PBST containing 5% milk for 1 hour at room temperature. Bound antibodies were revealed by Enhanced Chemiluminescence. Between all steps, nitrocellulose strips were washed with PBST. Note antibody response increased in the poly(I:C) injected mice even after termination of treatment. Despite recovery in glandular function, higher antibody titers are observed in poly(I:C) treated mice.