

## Supplemental Data:

**Figure S1.** No difference between T cell responses to TSHR A-subunit protein or mitogen in wild-type and Aire defective mice. Splenocytes from Aire  $+/+$  (n=7), Aire  $+/-$  (n=8) and Aire  $-/-$  (n=7) mice immunized three times with A-subunit-Ad were incubated in medium alone or with A-subunit protein (panel A) or Concanavalin A (panel B) for 6 days and culture supernatants were analyzed for IFN- $\gamma$  by ELISA (Methods). Controls were splenocytes from Aire  $+/+$  and  $+/-$  mice (n=5) immunized with Con-Ad. The data are shown as the mean  $\pm$  SEM ng/ml IFN- $\gamma$  for each group. In panel B, the shaded area represents the mean  $\pm$  SEM for splenocytes from all groups cultured in medium only. Significant differences between splenocytes incubated with A-subunit protein versus medium alone: \* p=0.038 for Aire  $-/-$ , p=0.015 for Aire  $+/-$  and p=0.003 for Aire  $+/+$  mice (paired t tests).

**Figure S2.** Similar recognition of TSHR peptides by wild-type and Aire defective mice. Splenocytes from Aire  $+/+$  (n=7), Aire  $+/-$  (n=8) and Aire  $-/-$  (n=7) mice immunized three times with A-subunit-Ad were challenged for 6 days with a panel of TSHR peptides (A to Z, EC1, EC2, EC3; see Methods) or cultured in medium alone (Med) and responses measured as IFN- $\gamma$  generated. Responses of splenocytes from  $+/+$  mice immunized with Con-Ad (n=5) were used as controls. The data are shown as the mean  $\pm$  SEM (ng/ml IFN- $\gamma$ ) for responses to each peptide. The dominant peptides recognized by splenocytes for each group are indicated by capital letters.