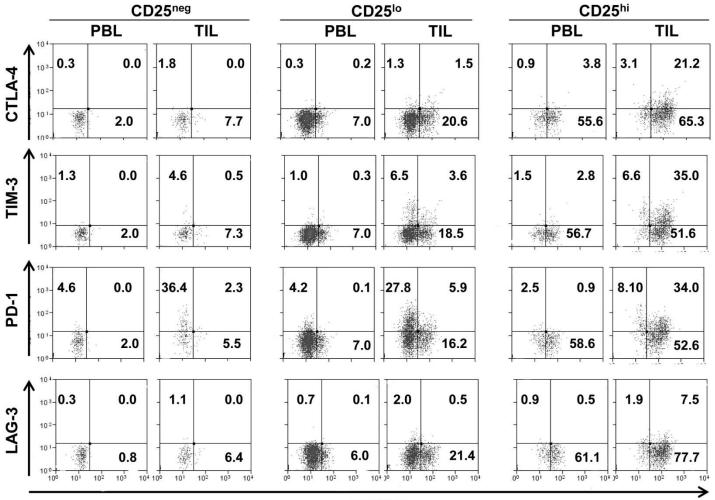
## **Supplemental Figure Legends**

**Supplemental Figure 1.** Flow cytometry analysis of CTLA-4, TIM-3, PD-1, and LAG-3 in CD4<sup>+</sup> T cell subsets. PBL and TIL CD4<sup>+</sup> T cells isolated from HNSCC patients were gated at CD25 negative (CD25<sup>neg</sup>), CD25 low (CD25<sup>lo</sup>), and CD25 high (CD25<sup>hi</sup>). (A) The numbers represent the percentages of CTLA-4<sup>+</sup>, TIM-3<sup>+</sup>, PD-1<sup>+</sup>, LAG3<sup>+</sup> cells in CD4<sup>+</sup> PBL and TIL. (B) LAG-3 expression on T cell subsets (CD25<sup>hi</sup>, CD25<sup>lo</sup>, and CD25<sup>neg</sup>) in PBL CD4<sup>+</sup> T cells of 6 HNSCC patients was analyzed and compared to that in TIL.

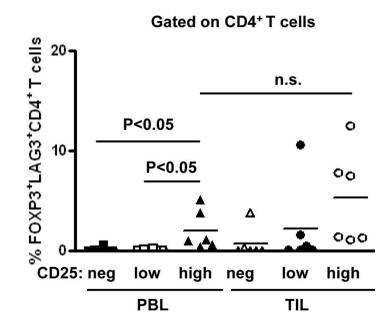
## Supplemental Figure 2. CD73 expression is not significantly upregulated by intratumoral FOXP3<sup>+</sup> Treg.

(A) CD73<sup>+</sup> cells among FOXP3<sup>+</sup>CD4<sup>+</sup> PBL were compared to those in FOXP3<sup>+</sup>CD4<sup>+</sup> TIL isolated from 7 HNSCC
(B) patients. (B) Flow cytometry analysis of CD73 expression on the FOXP3<sup>+</sup> and FOXP3<sup>-</sup> cells in CD4<sup>+</sup> TIL
(C) and PBL isolated from one of the HNSCC patients. The percentage values represent the fraction of CD73<sup>+</sup> cells
(D) in each FOXP3<sup>+</sup> and FOXP3<sup>-</sup> CD4<sup>+</sup> T cells in upper quadrants.

## Gated on CD4<sup>+</sup> T cells



FOXP3



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