



Distinct characteristics of novel immunoregulatory canine non-conventional TCRαβ⁺CD4⁻CD8α⁻ double-negative T cells

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Introduction

Potential immunosuppressive capacity of canine non-conventional TCRαβ⁺CD4⁻CD8α⁻ double-negative (dn) T cells

- Conventional CD4⁺FoxP3⁺ regulatory T cells (Treg) are crucial for controlling immune responses, thereby maintaining homeostasis and self-tolerance ¹
- Recently, within the substantial population of non-conventional TCRαβ⁺CD4⁻CD8α⁻ double-negative (dn) T cells of dogs, a novel FoxP3⁺ Treg-like subset was described ²
- Similar to conventional Treg cells, canine Treg-like dn T cells are characterized by high expression of the surface molecule CD25 ²
- The increase of dn T cells after allergen desensitization of dogs with adverse food reactions points to a regulatory role of canine dn T cells *in vivo* ³
- Despite the lack of FoxP3 expression, the small population of human and murine TCRαβ⁺ dn T cells have suppressive potential ^{4,5}

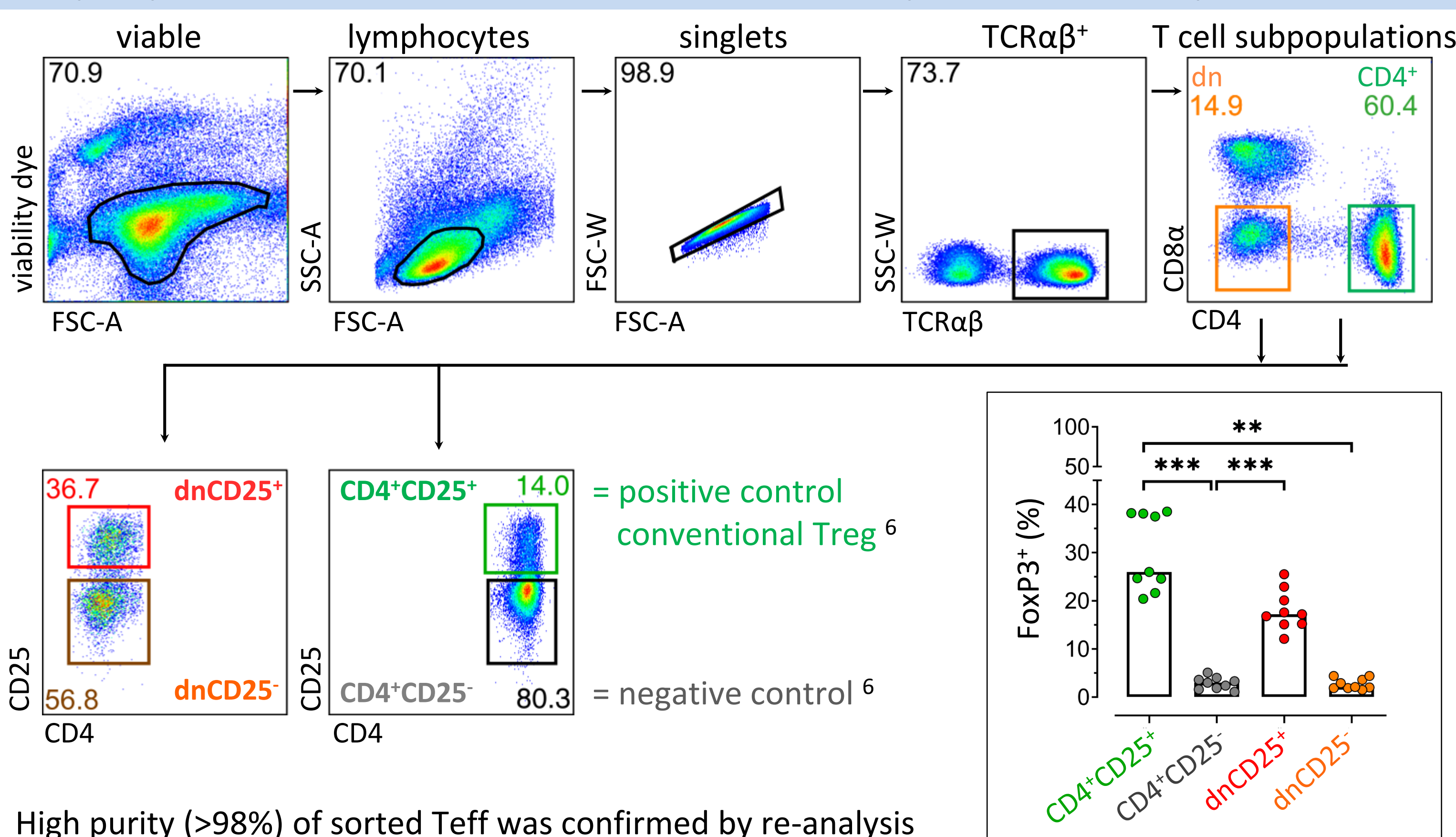
Aim of the study

Study the putative regulatory role of canine non-conventional FoxP3⁺ and FoxP3⁻ TCRαβ⁺ dn T cells by a functional *in-vitro* assay

Methods & Results

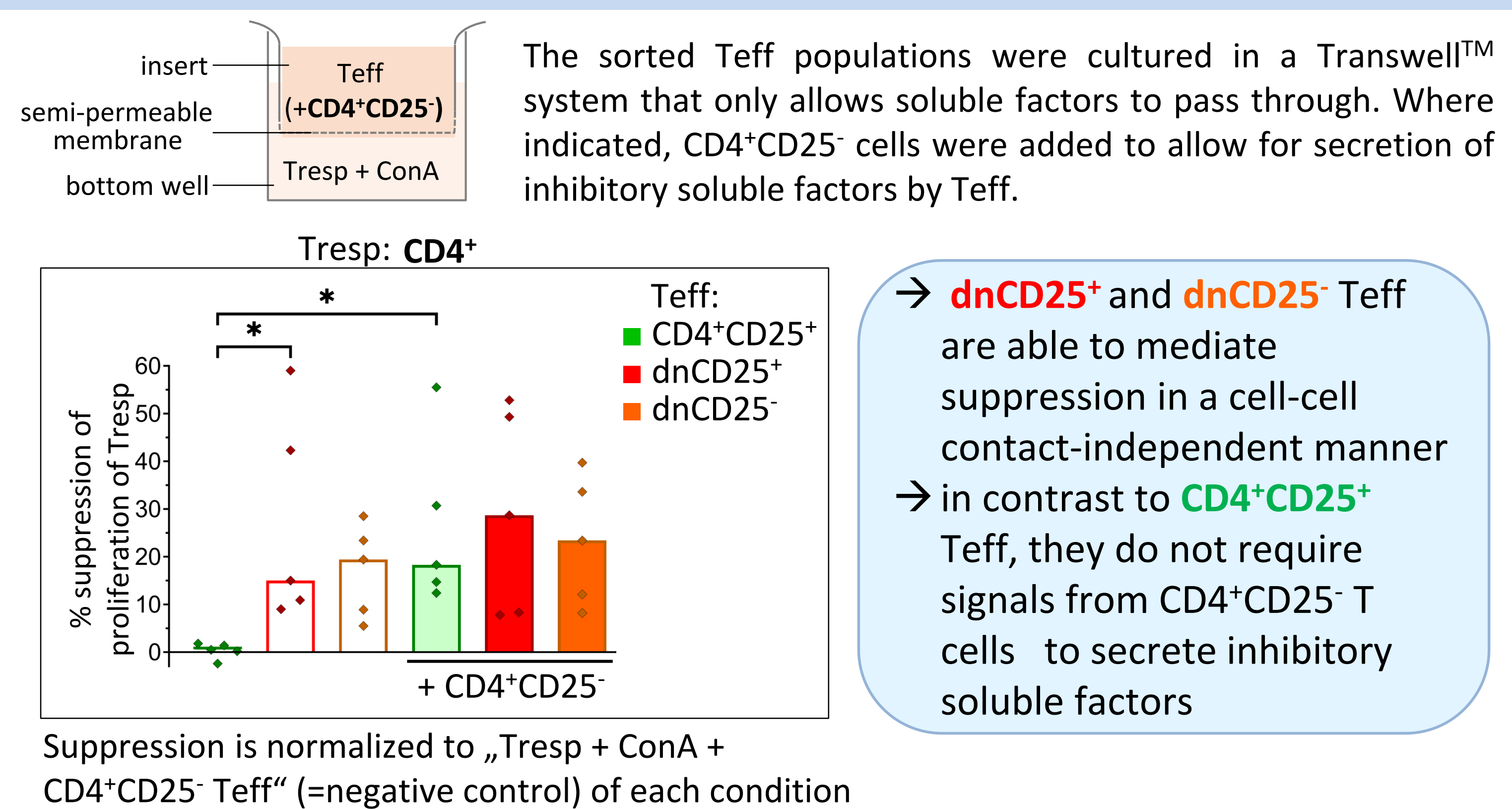
In-vitro suppression assay

1. Isolation of different effector T cell (Teff) populations from peripheral blood mononuclear cells and analysis of FoxP3 expression

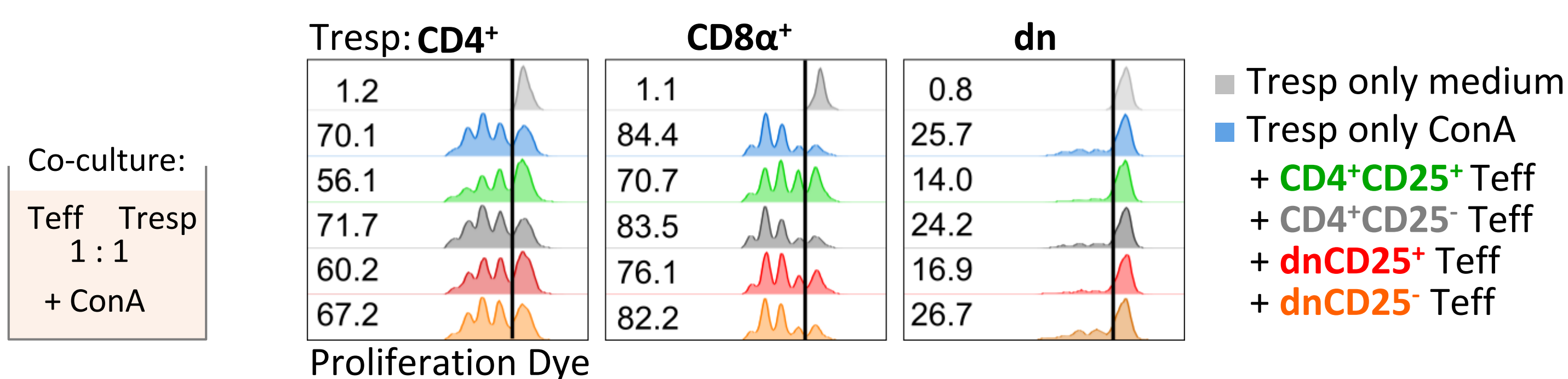


Transwell™ suppression assay: How do they suppress?

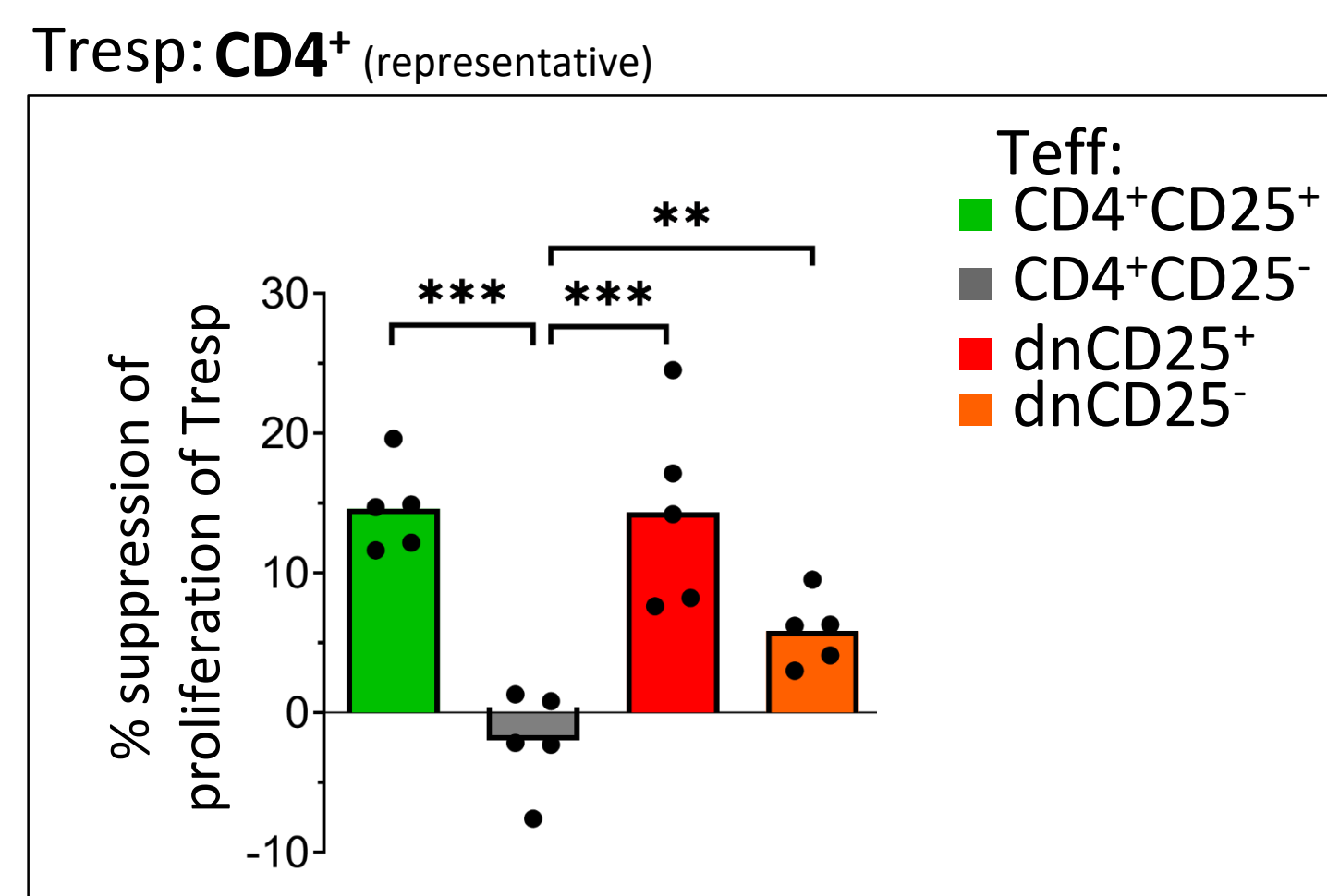
3. Cell-cell contact dependency ?



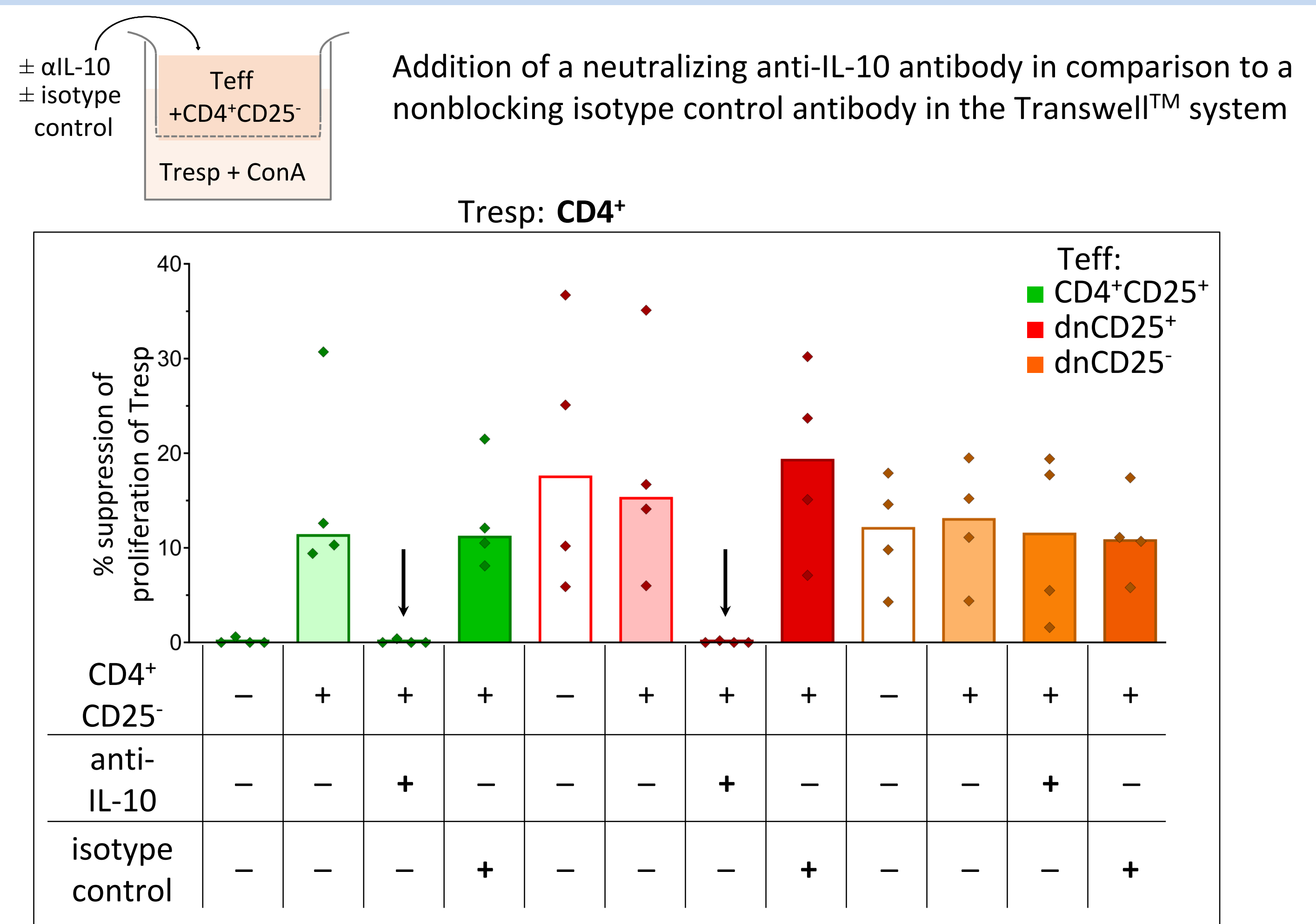
2. Suppressive effect of Teff on the proliferation of mitogen-stimulated responder T cells (Tresp) ?



Suppression is normalized to „Tresp only ConA“:



4. IL-10 dependency ?



Summary and conclusion

Our results demonstrate that canine non-conventional TCRαβ⁺ dn T cells contain unique and potent suppressive subpopulations *in vitro*. This is of high relevance, given the immunotherapeutic potential of manipulating regulatory T cell responses *in vivo*.

