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場　所：実習館 2 階総合歯科医学研究所セミナールーム

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タイトル：新規抗原取り込み受容体を標的とした抗体型ワクチンの研究

Dendritic cells (DCs) are professional antigen presenting cells (APCs) that can control innate and adaptive immune responses. Several DC subpopulations are known, and they typically express distinct receptors for antigen (Ag) uptake and capacities for Ag processing. A new way to assess receptor function, including in vivo, is to deliver Ags within anti-receptor monoclonal antibodies (mAbs), which selectively bind to the receptor and initiate Ag uptake, processing, and presentation on MHC products. The role of DC subsets expressing select receptors, as well as the role of the receptors themselves in Ag presentation, might therefore be determined by using specific mAbs as Ag carriers.
Triggering receptor expressed on myeloid cells-like 4 (Treml4) has been cloned in our Lab using microarray analysis as a molecule expressed CD8-positive DCs but not CD8-negative DCs in spleen. The functions of Treml4 are not known, but it has been shown that Treml4-Ig fusion protein binds to necrotic cells, and Treml4 associates with DAP12 in vitro. The aims of this study are to examine the expression of Treml4 on APCs, and to determine whether Ags can be presented via Treml4 using hybrid antibody of anti-Treml4: OVA.
We first examined the cell types that express Treml4 by FACS analysis. In mouse spleen and peripheral lymph nodes (pLNs), Treml4 was more abundantly expressed on CD8-positive DCs than CD8-negative DCs, which was consistent with results from microarray. However, CD3-epsilon positive T cells and B220-positive B cells do not express Treml4. We also found that F4/80-positive macrophages highly expressed Treml4 in spleen. Further analysis using tissue section staining reveal that Treml4 is abundant on F4/80-positive (red pulp) and CD169-positive (marginal metallophilic) macrophages in spleen. Because macrophages are found in many tissues, we further investigated the tissue distribution of Treml4 by western blotting. Endogenous Treml4 protein was readily detected only in spleen lysate, even though other macrophage markers were observed in all tissues investigated (spleen, pLNs, lung, liver, bone marrow and peritoneal lavage). These results indicate that Treml4 is primarily expressed in spleen, and in spleen, Treml4 is predominantly expressed on some DCs and macrophages.
We next determine whether antigens can be presented via Treml4 using hybrid mAbs (anti-Treml4: OVA), which was genetically introduced OVA protein into the C terminus of anti-Treml4 mAb H chains. C57BL/6 mice were injected with anti-Treml4:OVA and isotype control (ISO:OVA) 1 day after inoculating CFSE-labeled OT-I or OT-II T cells. After 3 days, pLNs and spleen were evaluated for T cell proliferation by CFSE dilution and the total number of expanded CFSE-low cells. The injection of anti-Treml4:OVA but not isotype control Ig:OVA induced proliferation of MHC I restricted CD8-positive OT-I cells, as well as MHC II-restricted CD4-positive OT-II cells, in both spleen and pLNs. The observations with OT-II cells in C57BL/6 mice were confirmed on DO11.10 T cells in BALB/c mice. These data suggest that antigens can be taken up via Treml4 and presented on MHC I and II products in vivo.
In conclusion, Treml4 is expressed on some DCs and macrophages in spleen, and antigens can be presented via Treml4 using anti-Treml4: OVA

担当：硬組織疾患制御再建学講座　宇 田 川 信 之