CD56 (NCAM1) in rainbow trout – molecular and functional characterization

Dang, Huong Thi; Korytar, Tomas; He, Xiaocui and Köllner, Bernd

Neural Cell Adhesion Molecule (CD56 or NCAM1) is a multifunctional member of immunoglobulin (Ig) supergene family. It is a marker of the natural killer (NK) cells and plays important roles in brain and muscle. However, very limited information about CD56 is available in fish. Here we describe the cloning and characterization of CD56 in trout. Sequence analysis showed that they possess all conserved CD56 characteristics such as signal peptide (SP) domain, five Ig domains, two fibronectin repeats (FNIII), either GPI anchor domain or transmembrane (TM) domain with intracellular cytoplasmic (IC) domain and CD56 functional motifs. Trout CD56 is derived from two genes composed of 20 constitutive exons leading to six isoforms CD56-120a, CD56-120b, CD56-140a, CD56-140b, CCD56-180a and CD56-180b. Two domains, the diverse alternative small VASE element and the muscle specific domain (MSD) create a very high variability of CD56 by alternative splicing (AS) events. Phylogenetic analysis revealed that trout CD56 molecules belong to the CD56 group and close to zebrafish CD56a. Expression pattern analysis in tissues obtained from healthy fish shows a specific pattern: CD56-180s were detected exclusively in brain while CD56-120s and CD56-140s are highly expressed both in brain and muscle. CD56-140 transcripts are detected in leukocytes excluding IgM$^+$ B cells and thrombocytes. Detailed analysis using sorted leukocyte subpopulations from primary, peripheral and mucosal lymphoid organs revealed the expression of CD56 in NK as well as NK-T-cell in trout. Furthermore, an up-regulation of CD56-140 upon xenogeneic stimulation suggests that CD56-140s+ Leukocytes are responding. Finally, an unexpected high variability of CD56-140s was detected in this cell that is changing after xenogeneic stimulation. These finding helps to identify NK cells in trout and opens field for future functional studies of these cells.