THE ROLE OF *cyyr1* GENE DURING ZEBRAFISH DEVELOPMENT IN HH-MEDIATED MYOGENESIS AND NEUROMASTS DIFFERENTIATION

F.Pizzetti¹, G.Deflorian², A.Pistocchi³, L.Ferrari², M.C.Pelleri¹, R.Casadei⁴ and F.Frabetti¹

¹ DIMES Dep. of Experimental, Diagnostic and Specialty Medicine, University of Bologna, ² The FIRC Institute of Molecular Oncology (IFOM) Foundation, Milan, ³ Dep. of Medical Biotechnology and Translational Medicine, University of Milan, ⁴ Dep. for Life Quality Studies, University of Bologna; Italy E-mail: fabrizio.pizzetti2@unibo.it

CYYR1 (Cysteine/tyrosine-rich 1) cloned on human chromosome 21 defines a new family of highly conserved vertebrate-specific genes^{1,2}. The analysis of the human locus revealed the presence of a multitranscript-system that includes alternative spliced isoforms and one ncRNA gene overlapping *CYYR1* in antisense orientation³. Original results suggest the need of further investigations in order to verify a putative role of *CYYR1* in the tumorigenic process, caused by dysfunction of cell differentiation and possibly related to the Hh pathway⁴; to date, the specific function of the CYYR1 product is still unknown.

The zebrafish *cyyr1* is present in single copy and maintains almost 58% of identity with human protein therefore, we decided to perform a full characterization of *cyyr1* expression and function using zebrafish as model system.

WISH approach defined a broad expression in central nervous system (CNS), somites and muscles during somitogenesis and at 24-48 hpf. The *cyyr1* knock-down with two different MOs targetting the ATG and the first splice-site of the transcript, affected both CNS and muscle development with a significant rescue in embryo co-injected with *cyyr1* mRNA. Defects were also evident in ciliated cells of neuromast of the lateral line.

Morphologically, the *cyyr1*-MOs injected embryos display some features of embryos inhibited for the Hh pathway through injection of the *lefty* mRNA and *cyyr1* expression was significantly inhibited following Hh inhibition. Interestingly, the injection of *cyyr1* mRNA was able to partially rescue Hh-defective phenotype in embryos at 24 hpf.

Results obtained through immunofluorescent staining, qPCR and western blotting, support a role for *cyyr1* in primary myogenesis probably downstream of Hh pathway.

- 1. Vitale L et al. Gene 2002, 290:141-51.
- 2. Casadei R et al. Gene Expr Patterns 2011, 11: 271-6.
- 3. Casadei R et al. Mol Biol Rep 2014, 41:6025-38.
- 4. Xu J et al. Genetics 2006, 174:735-52.