Molecular lesions in right ventricular infundibulum in Tetralogy of Fallot

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Introduction
Narrowing of the sub pulmonic infundibulum of right ventricle is characteristics of Tetralogy of Fallot (TOF). Transcription factors, signaling molecules, and structural proteins involved in embryonic development of right ventricular outflow tract (RVOT) could be key candidates in the pathogenesis of the infundibular stenosis from increased muscle mass in the region.

Objective
To elucidate the causal factors for regional muscle growth the gene expression pattern of cardiac transcription factors such as Mef2c, HAND2, GATA4 and ISL1 and proteomic profile of RVOT myocardium of patients with TOF were studied.

Methods
Sub pulmonic infundibular muscle tissue were obtained from six patients with TOF who underwent corrective surgery (age: 1 to 19 years) and four donor healthy human hearts harvested for transplantation (age: 11 to 40 years). The expression of candidate genes were analyzed and validated by semi quantitative RT-PCR (Applied Biosystems). Tissue proteins were subjected to proteomics protein expression analysis by 2D nano-LC MS/MS (Waters). Immunoblot experiments were performed to substantiate differential expression of selected proteins.

Results
An increased expression of Islet-1 (ISL1) was observed in all patients. ISL1 is a member of LIM-homeodomain transcription factor family and a marker of resident cardiac progenitor cells that are derived from second heart field (SHF) region. Mef2c, GATA4 and HAND2 had a decreased expression pattern in patients with TOF. In LC MS/MS analysis, 1500 proteins were identified in the infundibular muscle tissue among which 113 proteins were differentially expressed in patients and normal individuals. Among them, Retinaldehyde dehydrogenase 2 (ALDH1A2), an upstream element required for the downregulation of ISL1 was decreased while there was increased expression of proteins such as MAP4, required for cellular growth and division.

Conclusions
Decreased expression of transcription factors such as Mef2c, HAND2, GATA4 in the myocardium of patients with TOF suggest the underdevelopment of RVOT. Increased expression of ISL1 together with proteins for cell proliferation in RVOT indicate the presence of progenitor cells in the region, their inadequate differentiation and proliferative potential, all of which could contribute to the infundibular stenosis.