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COMMENTARY

GATA transcription factors in congenital heart defects: A Commentary on a novel *GATA6* mutation in patients with tetralogy of Fallot or atrial septal defect

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Congenital heart defects (CHDs) occur in nearly 1% of all live births and are the major cause of infant mortality and morbidity. In addition, about 3 per every 1000 live births will require some intervention during the first year of life.[1] Despite its clinical importance, the underlying genetic etiology of most CHD remains unknown. Previous studies succeeded in revealing genetic causes of some syndromic or familial CHD, however, there are limited numbers of such cases, and it is still difficult to approach the etiology of the majority of CHD, that manifest non-syndromic and non-familial phenotype, because of their multi-factorial nature.

In this issue, Lin *et al.* report on a novel mutation of a gene encoding a transcription factor, GATA6, as a possible cause of CHD.[2] The authors screened 270 individuals with non-syndromic CHD for the *GATA6* gene by direct sequencing, and identified a missense mutation (S184N) in three individuals, one with tetralogy of Fallot and the others with atrial septal defect. Although the incomplete penetrance of the phenotype by this mutation was observed, the mutation was not found in 500 ethnically matched healthy controls. And their subsequent biological analysis revealed the decreased transcriptional activity of GATA6 with the S184N mutation for the gene regulation of several important cardiac factors, suggesting that the *GATA6* S184N mutation may have an important role in the pathogenesis of CHD.

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The first identification of disease-associated mutations of *GATA6* in CHD was originally reported by us.[3] We screened mutations of cardiac transcription factors in patients with selected non-syndromic CHD, namely persistent truncus arteriosus, representing the most severe cardiac outflow tract (OFT) defects. Two different *GATA6* mutations were identified in two probands, but not in 182 unrelated controls with no CHD. Our subsequent biological analyses revealed that genes encoding the neurovascular guiding molecule semaphorin 3C and its receptor plexin A2 were directly regulated by GATA6, and both GATA6 mutant proteins failed to transactivate these genes. Transgenic analysis further suggested that the expression of semaphorin 3C and plexin A2 in the OFT was dependent on GATA transcription factors during the heart development. Together, our data implicate mutations in *GATA6* as novel genetic causes of CHD involving the OFT development, as a result of the disruption of the direct regulation of semaphorin-plexin signaling. Another recent study by Maitra *et al.* showed two novel sequence variations in *GATA6* (A178V and L198V) from the screening of 310 individuals with CHD.[4] These variants were identified in two individuals, one with tetralogy of Fallot and another with atrioventricular septal defect, but not in 288 ethnically matched healthy controls. Biochemical analysis demonstrated that the GATA6 A178V mutant protein resulted in increased transactivation ability for cardiac genes compared with the wildtype. Our first report and subsequent reports by Maitra *et al.*[4] and Lin *et al.*[2] in this issue provide an approach for the etiology of non-syndromic or 'multi-factorial' CHD in the post-genomic era, and together indicate that mutations in *GATA6* cause CHD implicated in the cardiac OFT and septal development.

GATA6 belongs to a family of transcription factors that bind to a GATA consensus motif (A/TGATAA/G) through a highly conserved zinc finger domain. Six members (GATA1–GATA6) of this family have been identified, all showing distinctive tissue-specific expression, and having an essential role during vertebrate development.[5] Members of GATA transcription factors are classified into two groups: GATA1–3 are expressed predominantly in haematopoietic cells and heterozygous mutations of *GATA1* and *GATA3* cause blood disorders and organ malformations in human, respectively, whereas GATA4–6 are expressed in the developing heart, as well as endodermal lineages, including gastrointestinal tract. To date, implication of GATA4 and GATA6 in CHD has been suggested (Figure 1). Homozygous *Gata4* knockout mice die *in utero* and develop two symmetric promyocardial primordial that fail to migrate ventrally and form the heart tube. Mice with heterozygous *Gata4* mutations exhibit septal defects and endocardial cushion defects. In human, mutations in the *GATA4* gene have been reported in familial cases of atrial septal defect and in a minority (1–4%) of sporadic patients with septal or OFT defects. Gene disruption of *Gata6* in mice results in early embryonic lethality from defects of the endodermal differentiation. Conditional inactivation of *Gata6* specifically in the cardiac neural crest cells that give rise to the septum of the OFT causes persistent truncus arteriosus, suggesting an essential role of

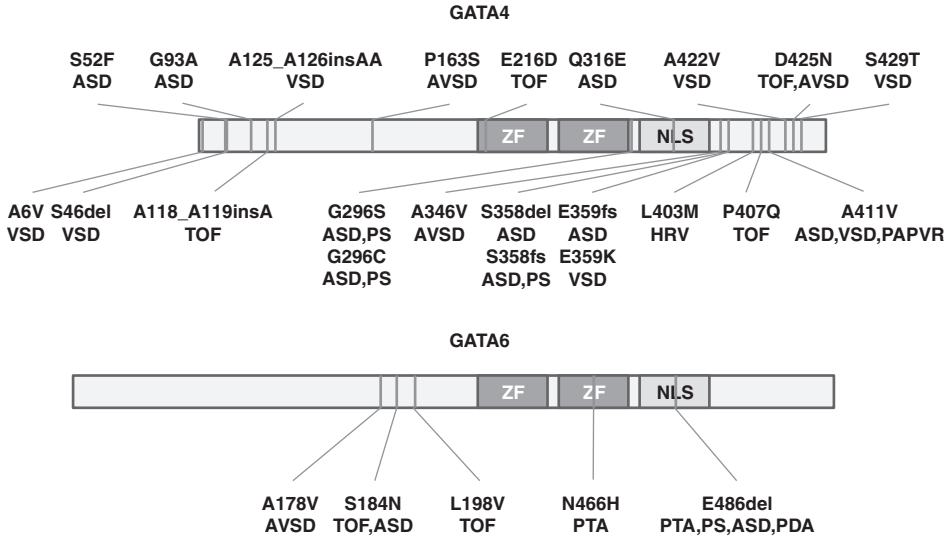

Figure 1 Schematic of GATA4 and GATA6 protein indicating the location of mutations and phenotypes of congenital heart defects reported to date ZF, zinc finger domain; NLS, nuclear localization signal; ASD, atrial septal defect; AVSD, atrioventricular septal defect; HRV, hypoplastic right ventricle; PAPVR, partial anomalous pulmonary venous return; PDA, patent ductus arteriosus; PS, pulmonary valve stenosis; PTA, persistent truncus arteriosus; TOF, tetralogy of Fallot; VSD, ventricular septal defect.

Gata6 during the OFT development.[6] Mice compound heterozygous for *Gata4* and *Gata6* null alleles die *in utero* and exhibit a spectrum of CHD, including septal and OFT defects, demonstrating a genetic interaction between these two GATA factors. It has also been shown that GATA4 and GATA6 regulate their expression each other during development and that GATA6 may function in concert with GATA4 to direct tissue-specific gene expression essential for formation of the mammalian heart. It is of note that *GATA6* mutations identified in human are predominantly associated with OFT defects, whereas *GATA4* mutations are commonly associated with septal defects, although there are some phenotypic overlap, probably, result from a redundant role between Gata4 and Gata6. OFT defects are significantly associated with the DiGeorge/22q11.2 deletion syndrome, and TBX1 on chromosome 22q11.2 has been proposed as a major genetic determinant of the extensive clinical features of this syndrome.[7] Commonly, the clinical phenotype of individuals with *GATA6* mutations involves the OFT, but is distinct from that of DiGeorge/22q11.2 deletion syndrome and, rather, manifests as non-syndromic CHD. To date, no molecular link has been demonstrated between GATA6 and TBX1,[8,9] however, a recent study showed that the expression of Sema3c in the OFT was down-regulated in mouse embryos deficient for *Tbx1*,[10] suggesting that GATA6 may share, at least in part, a common molecular pathway with TBX1 during OFT development.

Finally, Lin *et al.* in this issue identified the same heterozygous, non-synonymous mutation of *GATA6* in three patients with different clinical manifestations, one with tetralogy of Fallot and the other two with atrial septal defect.[2] The same mutation was also observed in their parents, two fathers with no CHD and a mother with bicuspid aortic valve. In previous reports, *GATA4* missense mutations identified in patients with cardiac septal defects were also found in some of their non-affected parents or family members, indicating the reduced penetrance of the phenotype. Either the reason for the incomplete penetrance or the underlying mechanism for the phenotypic difference among individuals with the identified *GATA6* mutations is unclear. Environmental factors, epigenetic factors and/or other genetic modifiers may also be responsible for the final phenotype of CHD in addition to the *GATA6* mutations in such individuals. There should be necessary further studies for more delineated genotype–phenotype correlations between *GATA6* mutations and cardiac OFT and/or septal defects, and the next challenge for identifying such factors that modify or influence the phenotype.

1 Gruber, P. J. & Epstein, J. A. Development gone awry: congenital heart disease. *Circ. Res.* **94,** 273–283 (2004).
2 Lin, X., Huo, Z., Liu, X., Zhang, Y., Li, L., Zhao, H. *et al.* A novel *GATA6* mutation in patients with tetralogy of Fallot or atrial septal defect. *J. Hum. Genet.* **55,** 662–667 (2010).
3 Kodo, K., Nishizawa, T., Furutani, M., Arai, S., Yamamura, E., Joo, K. *et al.* GATA6 mutations cause human cardiac outflow tract defects by disrupting semaphorin-plexin signaling. *Proc. Natl Acad. Sci. USA* **106,** 13933–13938 (2009).
4 Maitra, M., Koenig, S. N., Srivastava, D. & Garg, V. Identification of GATA6 sequence variants in patients with congenital heart defects. *Pediatr. Res.* (e-pub ahead of print 24 June 2010).
5 Maeda, M., Kubo, K., Nishi, T. & Futai, M. Roles of gastric GATA DNA-binding proteins. *J. Exp. Biol.* **199,** 513–520 (1996).
6 Lepore, J. J., Mericko, P. A., Cheng, L., Lu, M. M., Morrisey, E. E. & Parmacek, M. S. GATA-6 regulates semaphorin 3C and is required in cardiac neural crest for cardiovascular morphogenesis. *J. Clin. Invest.* **116,** 929–939 (2006).
7 Yamagishi, H. & Srivastava, D. Unraveling the genetic and developmental mysteries of 22q11 deletion syndrome. *Trends Mol. Med.* **9,** 383–389 (2003).
8 Yamagishi, H., Maeda, J., Hu, T., McAnally, J., Conway, S. J., Kume, T. *et al.* Tbx1 is regulated by tissue-specific forkhead proteins through a common Sonic hedgehog-responsive enhancer. *Genes Dev.* **17,** 269–281 (2003).
9 Yamagishi, H., Maeda, J., Uchida, K., Tsuchihashi, T., Nakazawa, M., Aramaki, M. *et al.* Molecular embryology for an understanding of congenital heart diseases. *Anat. Sci. Int.* **84,** 88–94 (2009).
10 Théveniau-Ruissy, M., Dandonneau, M., Mesbah, K., Ghez, O., Mattei, M. G., Miquerol, L. *et al.* The del22q11.2 candidate gene Tbx1 controls regional outflow tract identity and coronary artery patterning. *Circ. Res.* **103,** 142–148 (2008).