SEARCHING FOR POINT MUTATIONS IN GENES SELECTED BY CGH ARRAY IN PATIENTS WITH THYROID DYSGENESIS (TD) THROUGH WHOLE EXOME SEQUENCING (WES) ANALYSIS

Kizys, M.M.L.¹; Mitne-Neto, M.²,³; Furuzawa, G.K.¹; Nesi-França, S.⁴; Dias-da-Silva, M.R.¹; Maciel, R.M.B.¹

¹Medicine, Laboratory of Molecular and Translational Endocrinology, Escola Paulista de Medicina, Universidade Federal de São Paulo, São Paulo, São Paulo, Brazil
²Research and Development, Fleury Group, São Paulo, São Paulo, Brazil
³Human Genome and Stem Cell Research Center, Biosciences Institute, University of São Paulo, São Paulo, Brazil
⁴Pediatrics, Universidade Federal do Parana, Curitiba, Paraná, Brazil

Introduction: Thyroid dysgenesis (TD) is the most cause of Congenital Hypothyroidism and is characterized by alterations during thyroid embryogenesis: absence of the tissue (agenesis), diminished size (hypoplasia), misplaced gland (ectopic) and absence of bilobation (hemiagenesis). Thorwarth and cols (2010) have performed Array Comparative Genomic Hybridization (CGH) and identified novel and exclusive DNA copy number variations (CNVs) in 8.75% of 80 patients with TD. Objective: To search for point mutations in genes with CNVs previously described in TD patients. Material and Methods: Peripheral leucocyte DNA was isolated from 3 thyroid hemiagenesis, 3 hypoplasia, 3 ectopic, 5 agenesis cases and 14 controls for Whole Exome Sequencing (WES). Exons were captured by SeqCap EZ Human Exome v3 kit, sequenced in Illumina HiSeq2000 and mapping and variant calling done in Genomic Workbench 6.5. To filter the variations identified the following filters were used: minor allele frequency (MAF) ≤ 0.05, impact, coverage, prediction of effect and comparison to control cases. Results: in comparison with 6 control individuals, we have identified point mutations in PRODH in all TD types. LRBA and GGT2 have presented variations in agenesis, hypoplasia and hemiagenesis groups. We have identified variations in CLTCL1, USP9Y and ZDHHC8 genes in ectopic cases. For agenesis, variations were found in CLTCL1, COMT, DDX18, SERPIND1, SH3D19. In hypoplasia, DGCR14, and SH3D19 presented variations on the coding region. For hemiagenesis, EN1, MACROD2, USP9Y and ZNF74. No mutations were found in classical genes related to TD: NKX2.1, NKX2.5, PAX8, FOXE1, HHEX and TSHR. Conclusions: Herein, we first report the preliminary analysis of point mutations in genes with CNVs in patients with TD through WES analysis and suggest a possible complex genomic mechanism and/or signature of alterations involved in TD development. Further analysis including additional healthy thyroid individuals may strengthen the possible role of these new genes in thyroid development.

Acknowledgement: FAPESP
Key Words: Comparative Genomic Hybridization, Exome Sequencing, Thyroid Dysgenesis