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New mechanism of *DNMT3L* revealed in a Down syndrome study

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INTRODUCTION

Down syndrome (DS) is the most common genetic disorder affecting various organs and systems due to developmental defects. In nervous system, it manifests as intellectual delay in early age and early onset of Alzheimer disease [1]. Although the general cause is known as the trisomy of chromosome 21, it is not clear the overdoses of which genes are responsible for the neurological phenotypes. Furthermore, the involvement of epigenetic regulation during individual growth makes it even complicated [2]. These issues could potentially be addressed by a better mechanistic understanding of the effects of genes on chromosome 21, and DS is an ideal human model for understanding genetic-epigenetic interactions. Recently, we reported a novel molecular mechanism of *DNMT3L*, a gene located on chromosome 21 and involved in DNA methylation [3].

DESCRIPTION

In this study, we overexpressed *DNMT3L* gene in early-differentiated human neuroprogenitors and observed modest but significant changes in DNA methylation and more striking changes of RNA expressions in pathways involved in type I interferon signaling, cytokines, and cell adhesion. We further found *DNMT3L* bound the transcription factors (TFs) STAT1 and STAT3 and stimulated their tyrosine-phosphorylation, which in turn activated the expressions of several TFs, causing pro-differentiation of neuroprogenitors. This DNA methylation-independent pathway was further confirmed in *DNMT3L* conditional knock in mice, and the abnormal phenotypes was rescued by STATs phosphorylation inhibitors. These results suggest *DNMT3L* plays an important role in neurodevelopment and is a key contributor of neurological phenotypes in DS. This is a thorough and potentially important study of the roles of *DNMT3L* gene overexpression in mediating epigenetic and functional changes in developing DS neurons.

Previous studies with fetal DS cortices have shown global DNA hypermethylation (increased DNA methylation level compared with diploid controls) changes [4-8]. Since *DNMT3L* is known to facilitate the DNA methylation function of DNMT3A and DNMT3B, one hypothesis is that DS chromosome 21-linked *DNMT3L* causes these hypermethylation changes, affecting the downstream targeting genes for neurodevelopment. Thus, we tested this hypothesis in human fetal neuroprogenitors

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by comparing the DNA methylation and RNA profiles of control and *DNMT3L* overexpressing neurons. Out of our expectation, the methylation changes did not overlap with those in DS fetal cortices and were not correlated with the RNA expression changes. It suggests that *DNMT3L* causes some of the methylation changes in DS, but most changes in DS are not directly affected by *DNMT3L*. Alternatively, the temporal window of DS cortices did not match that of the *DNMT3L* overexpressing neuroprogenitors, and the dynamically modulated methylation during cortical development might cause the discrepancy. The non-correlation between methylation and gene expression suggests *DNMT3L* has a mechanism independent of DNA methylation.

Next, we examined the function of *DNMT3L* overexpression in human neuroprogenitors, and found it inhibited the proliferation and increased the differentiation of these cells, where the expression of several pro-differentiation TFs was upregulated. These results were consistent with the RNAseq data, where significant numbers of genes controlling neural differentiation or cell fate were altered to different extents. To connect *DNMT3L* with these TFs, we made use of proteomics data to look for *DNMT3L* binding proteins, among which we found STAT1 and STAT3 could modulate the transcription of pro-differentiation TFs and *DNMT3L* could promote STATs tyrosine phosphorylation. Thus, the molecular causal relationship between *DNMT3L* and early differentiation of neuroprogenitors was established.

Could the TFs changes cause the DNA methylation changes we observed? An alternative hypothesis is that the altered DNA methylation patterns in DS might be due to changes in the occupancy of TF binding sites in DS cells [9]. With HOMER software, we tested whether STAT-motif binding sites might be enriched among differentially methylated loci in human DS fetal brains and in the *DNMT3L*-overexpressing neuroprogenitors. Although there were TF binding sites motif enrichments of STAT3 around the hypermethylation CG sites of some genes, the enrichments in DS were completely different from those in *DNMT3L*-overexpressing neuroprogenitors, which could not be explained in the current study.

A limitation of this study is that we still don't know how much *DNMT3L* overdose contributes to the DS neuronal phenotypes. To answer this question, we need rescue experiments in DS neurons and mouse models by using genetic approaches such as CRISPR inhibition to titrate the expression levels in the DS neuroprogenitors to tune down *DNMT3L* expression to diploid levels and then to examine the epigenetic profile and neuronal phenotypes *in vivo*.

CONCLUSION

It's the first report that *DNMT3L* functions as a cofactor of transcription factor for transcriptional activation that does not affect DNA methylation or epigenetic modifiers. It is also noteworthy that *DNMT3L* dysregulation occurs not only in Down syndrome but also in autism, stress, cancer, and inflammatory diseases, where it may function as the bridge between environment and genes (our ongoing studies). We believe that this article will be of broad interest to the researchers and clinicians in terms of understanding mechanisms of *DNMT3L* in regulating the neurodevelopment in Down syndrome and providing clues for its function in other related disorders.

REFERENCES

1. **Lu J, Sheen V.** Combinatorial gene effects on the neural progenitor pool in Down syndrome. In: Down syndrome Book 1 Genetics and Etiology of Down Syndrome. 2011;pp:37-64.
2. **Lu J, Sheen V.** Genetic and epigenetic mechanisms in Down Syndrome brain. In: Down Syndrome. 2013;pp:237-261.
3. **Qin L, Qiao C, Sheen V, et al.** DNMT3L promotes neural differentiation by enhancing STAT1 and STAT3 phosphorylation independent of DNA methylation. *Prog Neurobiol*. 2021;201:102028.
4. **El Hajj N, Dittrich M, Bock J, et al.** Epigenetic dysregulation in the developing Down syndrome cortex. *Epigenetics*. 2016;11(8):563-578.
5. **Horvath S, Garagnani P, Bacalini MG, et al.** Accelerated epigenetic aging in Down syndrome. *Aging Cell*. 2015;14(3):491-495.
6. **Laufer BI, Hwang H, Ciernia AV, et al.** Whole genome bisulfite sequencing of Down syndrome brain reveals regional DNA hypermethylation and novel disorder insights. *Epigenetics*. 2019;14(7):672-684.
7. **Lu J, McCarter M, Lian G, et al.** Global hypermethylation in fetal cortex of Down syndrome due to DNMT3L overexpression. *Hum Mol Genet*. 2016;25(9):1714-1727.
8. **Mendioroz M, Do C, Jiang X, et al.** Trans effects of chromosome aneuploidies on DNA methylation patterns in human Down syndrome and mouse models. *Genome Biol*. 2015;16:263.
9. **Do C, Xing Z, Yu YE, et al.** Trans-acting epigenetic effects of chromosomal aneuploidies: Lessons from Down syndrome and mouse models. *Epigenomics*. 2017;9(2):189-207.