DOI 10.18699/SBB-2022-244

Pluripotent stem cell lines from two patients with *COH1* gene mutations as the valuable *in vitro* model of Cohen syndrome

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Key words: COH1, VPS13B, intellectual disability, autism, induced pluripotent stem cells, Cohen syndrome, Golgi apparatus, vesicular transport

Cohen's syndrome (CS) is an autosomal recessive disorder caused by mutations in the *COH1 (VPS13B)* gene. CS is characterized by growth and mental retardation, microcephaly, and the development of autism spectrum disorders.

The *COH1* gene has 62 exons and covers a region of approximately 864 kb on the human chromosome 8 [2]. For the moment, several hundred patients with CS have been described in the literature [1, 4], and according to the Cohen Syndrome Association (http://www.cohensyndrome.org/), there are about a thousand of CS patients all around the world.

The COH1 protein is one of the key participants of the intracellular membrane transport system, the central part of which is the Golgi apparatus (GA). Recent studies have shown that mutations affecting GA proteins also result in the postnatal microcephaly associated diseases. GA is extremely important for the differentiation and functioning of postmitotic neurons. It plays a major role in determining and creating the cellular polarity necessary for the formation of highly specialized neurites – axons and dendrites.

The product of the *COH1* gene is a peripheral membrane protein localized in GA and contributes to the structural maintenance and functioning of this complex. The supposed function of the COH1 protein is the regulation of vesicular transport and intracellular protein sorting [2, 5, 6]. Meanwhile, the molecular mechanisms of the *COH1* effects are still poorly understood.

Despite the similarity of the early stages of neurogenesis in all mammals, some of them cannot be reproduced in laboratory animals due to species-specific diversity. However, now the technologies of patient-specific induced pluripotent stem cells (iPSCs) have been developed and established the methods of their directed differentiation into neural cells. These technologies allow to overcome such limitations and carry investigations directly on the human cells. These approaches are already widely used for studying and modeling of the human neurodevelopmental and neurodegenerative disorders. However, in the literature there is only one report of CS iPSCs [3].

At the moment, we have obtained iPSC lines from the biopsy material of two patients with compound heterozygous mutations in the *COH1* gene:

Patient 1, (biopsy material – blood mononuclear cells) is a carrier of a compound heterozygous mutation (*COH1*-/-; chr8:g.100108653dup/chr8:g.100494031G > T).

Patient 2, (biopsy material – skin fibroblasts) is a carrier of a compound heterozygous mutation (*COH1*-/-; chr8:g.100514033T > C/ chr8:g.100844663_100844664del). For the skin fibroblasts of this patient, the following was revealed: swelling and vacuolization of GA cisternas, as well as GA fragmentation, which is typical for the CS patients. In addition, in the CS fibroblast cytoplasm, there are a large number of electron-dense granules, vesicles, autophago- and lysosomes, as well as an arrangement of dense bundles of microfilaments associated with the nucleus.

The iPSC lines from both patients have morphology of pluripotent cell, normal karyotype (46, XX), express pluripotency markers (OCT4, NANOG), and surface markers SSEA-1 and TRA-1-60. During differentiation in embryoid bodies, they contribute to three germ layers (ectoderm, mesoderm, and endoderm).

iPSC lines with a compound-heterozygous mutation in the *COH1* gene will serve as the basis for studying the pathology of cortical neurons formation in CS patients. There using will allow us to study the later stages of neurogenesis, in particular neuritogenesis, axons and dendrites formation, as well as the features of the structural organization of the GA and vesicular transport in the cell. By studying the effect of mutations in the *COH1* gene that cause CS, we will be able to establish the role of this gene in the process of normal human neurogenesis.

Acknowledgements: This study was supported by the Russian Foundation for Basic Research (grant No. 19-29-04067). We also thank the Multiple-access Center for Microscopy of Biological Subjects (Institute of Cytology and Genetics, Novosibirsk, Russia), state project No. FWNR-2022-0019 for granting access to microscopic equipment.

We are grateful to Pomerantseva E., Kotov I. and Musatova E., from the Center of Genetics and Reproductive Medicine GENETICO, JSC, Moscow, Russia and Lagarkova M. and Anufrieva K. from the Federal Research and Clinical Center of Physical-Chemical Medicine of Federal Medical Biological Agency, Moscow, Russia, for the invaluable help in obtaining biopsy material from patients and fibroblast cell.

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