

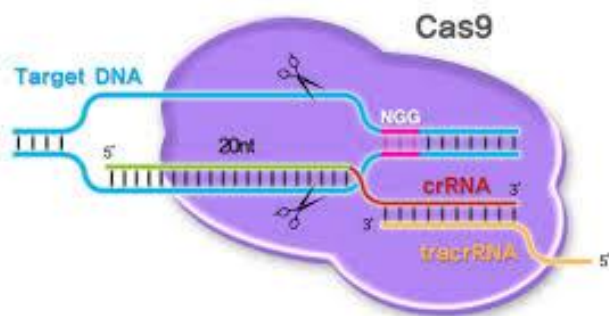


NATIONAL BIOETHICS COMMISSION

OPINION

Recent developments in human genome modification: Genome Editing

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The Hellenic National Bioethics Commission met repeatedly in order to consider the ethical issues within its jurisdiction regarding “human gene editing”. This has been occasioned by the announcements about the new CRISPR/Cas9 technology, which is characterized by simplicity, effectiveness and flexibility and thus, greatly facilitates genome editing in various organisms, including humans. This method has been the most remarkable achievement of the last decade on a road of continuous progress in genetic engineering technology.

Even though this new technology has far-reaching applications in all species, the present Opinion focuses on humans as this is where the bioethical issues appear to be most acute.

I. The data

The new genome editing technologies are based on Zinc Finger Nucleases (ZFNs), TALEN type nucleases (Transcription Activator-Like Effector Nucleases) and CRISPR/Cas9 system nucleases (Clustered Regularly Interspaced Short Palindromic Repeats/associated protein-9 nuclease). These nucleases’ systems have the ability to excise DNA by generating double-strand breaks at specific and targeted sites in the genome leading to the repair of the specific site, either with addition, removal or replacement of specific DNA sequences.

These technologies apply first and foremost to the editing of gene mutations which lead to human diseases. The repair may apply to somatic cells, germ line cells (gametes), stem cells and embryonic cells as well. Furthermore, new technologies may be used in the editing and repair of chromosomes and in treatments against infection.

Zinc Finger Nucleases (ZFNs) have been applied to human cell lines,¹ and indeed have already been used in a Phase I clinical study for patients infected by the

¹ Carroll D. Progress and prospects: zinc-finger nucleases as gene therapy agents. *Gene Ther.* 2008 Nov;15(22):1463-8.

Human Immunodeficiency Virus (HIV).^{2,3} The results of this study are expected to be announced.

The CRISPR/Cas9 is the most widely used system for genome editing because of its simplicity and flexibility. However, its main drawback so far has been that it also acts in unintended targets in the genome and creates cleavages in locations other than the ones of the intended gene or site (off-target effects), which present common sequences.

As of now the CRISPR/Cas9 technology has been applied to human cell lines.^{4,5,6} Specifically, it has been used in intestinal stem cells of cystic fibrosis patients to edit a CFTR gene mutation, which causes the monogenic disease of cystic fibrosis,⁷ but also in the simultaneous repair of several DNA sites,⁴ with a potential to edit mutations leading to polygenic diseases too.

At the heart of the concerns about the new genome editing technologies has been the recent application of the CRISPR/Cas9 system to human embryos by a scientific team in China. The application aimed to repair a β -globin gene mutation,

² Phase 1 Dose Escalation Study of Autologous T-cells Genetically Modified at the CCR5 Gene by Zinc Finger Nucleases in HIV-Infected Patients. <https://www.clinicaltrials.gov/ct2/show/NCT01044654?term=hiv+and+sangamo&rank=1>.

³ Tebas P, Stein D, Tang WW, Frank I, Wang SQ, Lee G, Spratt SK, Surosky RT, Giedlin MA, Nichol G, Holmes MC, Gregory PD, Ando DG, Kalos M, Collman RG, Binder-Scholl G, Plesa G, Hwang WT, Levine BL, June CH. Gene editing of CCR5 in autologous CD4 T cells of persons infected with HIV. *N Engl J Med*. 2014 Mar 6;370(10):901-10.

⁴ Cong L, Ran FA, Cox D, Lin S, Barretto R, Habib N, Hsu PD, Wu X, Jiang W, Marraffini LA, Zhang F. Multiplex genome engineering using CRISPR/Cas systems. *Science*. 2013 Feb 15;339(6121):819-23.

⁵ Mali P, Yang L, Esvelt KM, Aach J, Guell M, DiCarlo JE, Norville JE, Church GM. RNA-guided human genome engineering via Cas9. *Science*. 2013 Feb 15;339(6121):823-6.

⁶ Hou Z, Zhang Y, Propson NE, Howden SE, Chu LF, Sontheimer EJ, Thomson JA. Efficient genome engineering in human pluripotent stem cells using Cas9 from *Neisseria meningitidis*. *Proc Natl Acad Sci U S A*. 2013 Sep 24;110(39):15644-9.

⁷ Schwank G, Koo BK, Sasselli V, Dekkers JF, Heo I, Demircan T, Sasaki N, Boymans S, Cuppen E, van der Ent CK, Nieuwenhuis EE, Beekman JM, Clevers H. Functional repair of CFTR by CRISPR/Cas9 in intestinal stem cell organoids of cystic fibrosis patients. *Cell Stem Cell*. 2013 Dec 5;13(6):653-8

which causes β -thalassemia.⁸ The results of the study indicated mosaicism in embryos, i.e. the mutation was not fully repaired in the genome of all cells, and that there were off-target cleavages in unintended genes, suggesting that the method still has ample room for optimization.

II. Bioethical and law Issues

The Commission considers that interventions in the human genome can make a decisive contribution to health protection as long as their safety and effectiveness have been experimentally confirmed, especially in the field of prevention of major genetic diseases. However, the Commission notes that the application of genetic engineering in humans, even under safety conditions, bears the danger of the ability to predetermine genetic traits, something which ultimately leads to positive eugenics. In particular:

1. The issue of safety

Human genome editing still presents with considerable uncertainty concerning the results due to our limited knowledge of the exact gene functions and their interactions. Despite the increasing predictability that the new technologies may promise (especially the CRISPR/Cas9 method), the possibilities of unintended outcomes are still strong.

It should be noted that the degree of uncertainty as to the results increases when the genomic changes are transferred to offsprings through reproduction. Therefore, the prohibition of targeted gene editing in gametic cells, according to the

⁸ Liang P, Xu Y, Zhang X, Ding C, Huang R, Zhang Z, Lv J, Xie X, Chen Y, Li Y, Sun Y, Bai Y, Songyang Z, Ma W, Zhou C, Huang J. CRISPR/Cas9-mediated gene editing in human triprounuclear zygotes. *Protein Cell*. 2015 May;6(5):363-72.

explicit provisions of our legislation (Article 13 of the Oviedo Convention), is justified.⁹

In addition, studying the consequences of gene editing on embryo development *in vivo* prerequisites that the risks of the technology have been precisely identified. As long as this risk identification has yet to be reached, and thus the degree of uncertainty remains high, the approval of relevant research protocols by responsible state or academic authorities remains unlawful since the consequences on the health of the new organism are unknown.

2. *The issue of use under safety conditions*

Besides the safety issues there is an additional ethical issue as to the use of the modification methods on the human genome. What is being questioned is whether any use is lawful –either for health reasons or not– provided that the specific genome editing intervention has been proved to be safe.

The Commission states that, according to the law, the genome editing interventions that are acceptable are those being made for “*preventive, diagnostic or therapeutic purposes*”. However, it is still unclear whether this expression also covers genetic enhancement interventions, especially if we take into consideration the widely defined notion of “health” according to the World Health Organization (WHO), which includes “enhancement” too.

Recalling past the Opinions on “Human Enhancement”, in which the relevant issues emerged– the Commission considers that the genome editing interventions for enhancement reasons may be lawful only if they do not abet discriminations based on genetic, predetermined external features of persons. This is because:

a) the predetermination of external features –also known as “positive eugenics”– may place significant burdens on the autonomy of the future person,

⁹ In the same direction, see the recent Statement of the International Summit On Human Gene Editing: International Summit on Gene-Editing 2015. <http://www.nationalacademies.org/gene-editing/Gene-Edit-Summit/index.htm>.

fostering the impression that he/she is actually his/her parents' "design" product, without the possibility of free differentiation, and

b) this predetermination may have wider social consequences if it is considered that some characteristics are generally desirable while others are not, and respective "models" of unfair discriminations against certain population groups are being fostered.

Finally, the Commission highlights that possible mutation editing in the genome in order to avoid late onset disease, will be lawful (in persons or embryos), as long as they do not raise issues of positive eugenics, such as the above mentioned.

III. Suggestions

Taking into consideration a wider application of the "precautionary principle", the Commission deems that the only way to address the issue of safety concerning human genome editing is to persist in basic and pre-clinical research, with the aim to gain understanding of the human embryo's biology and gametes, and continuously assess the potential benefits and risks of the suggested clinical applications.

In light of these:

1. It is imperative to make efforts on the precise identification of unintended effects particularly regarding the practical support of research initiatives, in order to be able to design experimental clinical protocols that will guarantee a higher degree of safety for the participants. This safety measure should be at least of the same extent with the one applied to clinical trials of conventional pharmaceutical drugs. First of all, the safety measure and the benefit regarding genome editing technologies as a means of gene therapy in somatic cells, must be precisely specified and authoritatively established by the scientific community. The application of genome editing technologies in somatic cells may predate due to the fact that

genome interventions affect the persons themselves and are not inherited by their offsprings. Consequently, the evaluation of applications in somatic cells will be expedited by the existing regulatory framework.

Our country should not be absent from the important international effort in this crucial field of basic biomedical research. It is imperative for the relevant Ministry to take this into consideration for the national research planning and encourage the application of relevant research proposals by our research bodies.

2. According to the Commission, genetic interventions in human gametes, as well as in embryos *in vitro*, are lawful solely and exclusively for the purpose of basic research; that is as long as gametes and embryos are not intended for reproduction but are being used in order to assess the dangers, the potential benefits and the alternative approaches of genetic editing. This is imposed not only for safety reasons, but also for the need to avoid any possible implementations of genetic discriminations and positive eugenics in the future.

In this context, the Commission suggests a special legislative clarification of the provision of the Article 13 of the Oviedo Convention, so that the application of genome editing would explicitly be excluded for purposes of positive eugenics. A relevant addendum could be incorporated in Law 3305/2005 (“Enforcement of Medically Assisted Reproduction”).

Finally, the Commission emphasizes the need for a continuous participation of the Greek scientific community, the concerned State bodies and the patients’ associations in the continuing consultation process in national and international fora, aiming to fostering relevant guidelines, which will be subjected to continuous updating.

Dissenting Opinion

The Metropolitan of Samos and Ikaria, Efsevios Pistolis, member of the Commission, expressed his disagreement with the opinion of the majority, as follows:

“The beginning of life and the embryo’s value

The Fathers and the great theologians of the Church speak of a psychosomatic being from the first moment of conception and refer to a being that has all the human characteristics, which, nevertheless, need a specific procedure so they can be completed and manifested. The fact that this biological procedure has not been completed does not, in any way, mean that there is no life. For this reason, the term “embryo” is replaced by the term “human” in many of their literary works; because the soul coexists with the body in life since the very moment of conception.

All the other viewpoints upon this matter, seem to be without God, and that is why the embryo is treated as a biological tissue, as a mass of undifferentiated cells or as a biological material without rights in life. Some scientists are looking for grounds to distance as far as possible the beginning of life from the first moment of conception, creating a morality based on a purely utilitarian nature. This enables them to use the embryo as research material, to accept abortions, to indiscriminately endeavor cloning, and to speak of treatments for incurable diseases with the use of stem cells, which destroy the embryo.

For the Greek Orthodox Church “to destroy an embryo” means “to destroy a person”. The disruption of an embryo’s life, in whatever stage of development it might be, is equivalent to murder and the taking of human life. Even if this destruction is taking place in order to save the lives of others, it cannot be acceptable by the Greek Orthodox Christian Morals because no one has the right to take even his/her own life, let alone the life of another person; even if the person is in an embryonic state. However, if someone wishes to sacrifice something for the good of the humankind, he/she can offer him/herself by obeying to the teaching of our Lord, Jesus Christ, who emphasizes in John, Chapter 15, verse 13: “Greater love has no one than this, that one lay down his life for his friends”, something which, of course, He first realized with his voluntary passion”.

Athens, 27 April 2016