Nuffield Council for Bioethics: Genome Editing call for evidence

Submission from the Biotechnology and Biological Sciences Research Council and the Medical Research Council *(February 2016)*

Summary

- Genome Editing (GE) techniques such as CRISPR[1]-Cas9 are having a game-changing effect on research. We encourage an open and widespread debate about the future application of GE techniques, and therefore strongly welcome the breadth and scale of the Nuffield Council for Bioethics' current project.

- In the UK our strong ethical and regulatory framework has allowed us to find ways forward through complex ethical issues. We support the continued assessment, refinement and use of CRISPR-Cas9 and other GE techniques alongside open debate and responsible and robust regulation. This includes the use of these technologies in preclinical research, including in human reproductive cells and early embryos, where fully justified scientifically, ethically and legally.

- There are risks common to all applications of GE that must be considered by all researchers, including off-target changes and significant genetic drift. There is a clear obligation on researchers we fund to consider the wider potential impacts of their research, and follow best practice as outlined in our guidance and conditions of their award. Experimental design is quality assured through BBSRC and MRC's high standards of peer review.

- We believe that a system based primarily upon self-governance by the scientific community, but drawing on the inputs of other key stakeholders, will ultimately provide the most effective means of managing risks of misuse.

- Particular applications of GE – including plant, animal and microorganism - have additional unique considerations, as detailed further in our response.

Introduction

1. The Research Councils welcome the Nuffield Council on Bioethics project on Genome Editing and the opportunity to respond to the call for evidence.

2. The submission represents the views of the Biotechnology and Biological Sciences Research Council (BBSRC) and the Medical Research Council (MRC) as the two Research Councils which fund research utilising a range of different GE techniques. The Research Councils have previously contributed to the public debate around these technologies. In September 2015 we issued a joint statement[2] with the Academy of Medical Sciences, the Association of Medical Research Charities and the Wellcome Trust in support of the continued use of CRISPR-Cas9 and other GE techniques in preclinical research. This includes the use of the technology for research purposes in human reproductive cells and early embryos, where this is fully justified, scientifically and ethically, and within the confines of the law. The statement called for an open and widespread debate involving scientists, ethicists and the public about how GE techniques could be applied in the future. We therefore strongly support the breadth and scale of the current project as an important contribution to the discussion. BBSRC also issued a position statement in October 2014 on the new techniques for genetic crop improvement[3], which referenced the contemporary landscape for GE in plants; it recognises that the techniques are already used widely in research and regulatory processes for new crops will need to be able to accommodate them.

3. In our response to this call for evidence we have aimed to provide BBSRC and MRC's perspective on some of the indicative questions, we have crossed referenced our responses across sections as appropriate. We have also encouraged our investments to respond directly and separately on how these issues impact on their own work. The Research Councils collectively will follow the progress of the working group and would

[1] CRISPR: Clustered Regularly Interspaced Short Palindromic Repeat
[2] http://www.wellcome.ac.uk/About-us/Policy/Spotlight-issues/Genome-editing/WTP059704.htm
[3] http://www.bbsrc.ac.uk/documents/genetic-crop-improvement-position-statement-pdf/

be happy to comment on issues relating to other aspects of research in GE as they emerge from the project.

4. The MRC, along with the Wellcome Trust and the Nuffield Foundation, funds the Nuffield Council for Bioethics. Several Working Group and Council members also receive BBSRC and MRC research funding.

Perspectives on genome editing

5. GE is not an entirely new idea or capability. For decades researchers have been developing tools for genetic modification, some of which have offered potential gene therapy applications to correct missing or faulty DNA and restore healthy gene expression in cells, or modifying genes. Many of the ethical challenges of GE techniques such as CRISPR-Cas9 are similar to those presented by conventional gene therapy however there are important considerations around issues such as traceability which we address below (see 12).

6. Notwithstanding the above, rapid developments in GE technologies are having a game-changing effect on research. The emergence of techniques that can edit the genome in a much more efficient and targeted fashion, such as CRISPR-Cas9, has accelerated the field to such an extent that researchers can now make precise edits to the genome in a relatively easy, speedy, precise way. Such a capability has a myriad of potential uses, from lab tool to explore the mechanisms of disease to a way to develop novel therapeutic treatments for a range of diseases through gene or cell therapy. Researchers who may previously not have attempted to use GE technologies are now doing so, including many funded by the BBSRC and the MRC. As an illustration the number of applications to the MRC proposing to utilise CRISPR for gene editing rose from less than 10 in 2013/14 to 80 in 2015/16; the number of applications to the BBSRC citing CRISPR in their experimental approach narrative more than doubled from February 2015 (41 applications) to February 2016 (83 applications).

7. When discussing GE it is important to differentiate between different applications and outcomes, which are, briefly:
   - In laboratories as a further research tool to, for example, effectively introduce changes to the genomes of cell lines;
   - In laboratories as a research tool to induce changes to somatic or germline cells in experimental non-human animal models, such as mice;
   - In laboratories as a research tool to induce changes to somatic or germline human cells;
   - In laboratories as a research tool to induce changes to human embryos;
   - Its clinical application to produce non-heritable changes to ameliorate or prevent genetic disease;
   - Its clinical application to produce heritable changes to germline or embryonic cells to ameliorate or prevent genetic disease (currently not permissible within the UK);
   - Its application to induce changes to agriculturally important traits (crop plants and livestock; no applications yet realised).

8. There are risks common to all applications of GE that must be considered by all researchers. While the likes of CRISPR-Cas9 offer more precise targeting than previously available, off-target changes still occur. Furthermore, clonal selection of edited cell lines remains necessary to ensure that all cells in the experimental population carry the required change. As more laboratories implement GE technology researchers will need to continue to mitigate the risk of off-target effects and significant genetic drift through stringent validation and the careful use of standardised background cell or animal lines. Funders and research publishers will continue to engage with the research community to refine protocols and experimental design, areas quality assured through the Research Council's robust peer review mechanism. Funders may also see increased researcher need for facilities able to advise and carry out high-standard GE techniques on their behalf (for example mouse line genome editing services provided by the Mary Lyon Centre at MRC Harwell).

9. Before GE methodologies can be introduced at the clinical level, basic and pre-clinical research is needed to assess the wider impact of such therapeutic interventions on the edited cell and daughter cells and in the human context. For example, research may be needed to explore the long-term impact of a cell's response to the introduction of a modification (e.g. in terms of gene expression levels) and any associated stress or immune responses that may be induced by this or the GE process. Sustainability/stability of an engineered change across a cell and its progeny will also need to be explored.

10. In the UK our strong ethical and regulatory framework allows us to find ways forward through complex ethical issues. For example, in recent years, the public, researchers and regulatory systems have come

together to find a way forward both in research using human embryonic stem cells and in the use of mitochondrial donation. An important marker of the debate in the UK is the recent approval by HFEA for the application of CRISPR technology by Dr Karen Niaken's group at The Francis Crick Institute to investigate healthy human embryo development during the first seven days of life[4]. The Working Group's current project, along with other activities such as the work of the Hinxton Group[5], will play an important role in moving the debate forward. We fully support the continued assessment, refinement and use of GE technologies to unravel the complexity of disease and find new ways to treat it. For all GE-based research, we believe that the responsible and (robustly) regulated use of these technologies, while their future is openly debated, is the most effective way to push the frontiers of science. Consequently, we do not support a moratorium on the use of GE technologies within the UK.

11. **Distinctive significance of genome interventions:** All approaches to the breeding of plants and animals, whether biotechnological or by other methods, result in changes to the organism's genome. Some interventions can make this process more selective and controlled, while some can increase the chances of random genetic changes. Biotechnological processes enable researchers to exert control over changes to the genome, rather than it being a random process controlled by evolution.

12. **Extent to which genome editing techniques can be regarded as distinct from or continuous with existing techniques:** GE approaches are on the continuum of existing techniques however, there are a number of important distinctions that greatly expand the research toolkit and accelerate the pace such as: (1) ability to target a very precise location for change (inserting a change at a specified genetic location[6] or targeting a single gene sequence[7]); (2) possibility of creating "markerless" changes, for example single point mutations or small indels that may be indistinguishable from naturally-occurring variation. In these respects, GE techniques are more precise than chemical or UV mutagenesis techniques, which have long been accepted as "traditional" approaches to breeding; they are also less dependent upon wider regions of insertion and have a reduced need for selectable markers such as antibiotic resistance genes.

13. **Scientists' obligations, freedoms, distinctiveness from other scientists:** Researchers should be free, within legal and regulatory limits, to undertake fundamental discovery research judged as excellent by peer review. The Research Councils expectations for Research Organisations and researchers in ensuring they adhere to all legal, regulatory and ethical requirements are outlined clearly in our guidance on good research conduct and conditions of award, as well as the clear obligation to consider the wider potential impacts of their research and, where appropriate, to engage with stakeholders around potential benefits and dis-benefits.

14. **Value of GE as a pure research tool vs. envisaged practical applications:** GE techniques have huge potential as research tools and for practical applications. In 2015 CRISPR was described as "the biggest game-changer to hit biology since PCR"[8]. As GE is still at an early stage technologically (although seeing rapid new developments), its full value, and the potential of the increased precision, efficiency and pace it offers is still to be fully realised.

15. **Other issues to note on genome modification:** Genetic changes, however they are produced in an organism[9], can add beneficial characteristics or remove undesirable ones. Benefits and dis-benefits are associated with the particular changes introduced, rather than the method by which they were introduced.

16. In some cases it will be impossible to tell what method was used to produce an altered organism, (e.g. a new crop variety as noted at 12 above), because a range of conventional breeding or GE techniques could produce the same genetic changes. The boundaries between established genetic modification (GM) and non-GM techniques will also become increasingly blurred as GE techniques develop. This raises questions about how organisms altered by any means should be regulated. Regulation based on the characteristics of novel organisms, however produced, would provide more effective, robust and future-proofed regulation than considerations based on the method used to generate them.

[4] http://www.mrc.ac.uk/news/browse/new-gene-editing-techniques-approved/
[5] http://www.hinxtongroup.org/. The Hinxton Group is funded by the MRC, the Wellcome Trust and others.
[6] Notwithstanding any off-target effects. The nature and likelihood of off-target effects is the subject of current research, for example: http://www.cell.com/trends/biotechnology/abstract/S0167-7799(14)00240-6 and research is continuing on making gene editing highly specific (for example see http://www.nature.com/news/enzyme-tweak-boosts-precision-of-crispr-genome-edits-1.19114)
[7] For example in the CRISPR-Cas9 system, using specific guide RNA.
[8] http://www.nature.com/news/crispr-the-disruptor-1.17673
[9] Either via biotechnology, conventional mutagenesis by humans, or natural selection 'in the wild'

Paragraph 17 to 36 outline the BBSRC and MRC's perspective on indicative questions relating to plant science, animals, microorganisms and human applications and there is some overlap between sections.

Genome editing in plant science

17. **State of the art; direction of travel; envisaged endpoints:** GE offers great potential to generate new plants varieties and animal breeds more rapidly, at reduced cost, without marker DNA insertions[10], to deliver benefits that would otherwise be too slow, costly or not possible currently, to develop by conventional means. GE could improve crops by enhancing disease resistance, nutritional content, harvest storage performance, or processing properties for food and non-food uses.

18. **Gene drives:** GE offers the possibility of stimulating biased inheritance of particular genes to alter entire populations of sexually-reproducing organisms, enabling population control. In plants, gene drive could contribute potentially to sustainable agriculture by reversing pesticide and herbicide resistance[11]. More widely, it holds promise for the control of insect pests and vectors of disease – something already being tested in the field for controlling malaria-bearing mosquito species[12].

19. **Drivers and obstacles towards endpoints:** Both plant research and plant breeding can be achieved in some way by current techniques, but GE is likely to prove more efficient and cost effective, and potentially open up new possibilities for R&D that have not yet been anticipated. Limitations might include research funding, time needed to tailor GE techniques for specific aims, and training researchers to use GE effectively. Regulation based on genetic technique rather than end-product could also be a barrier towards achieving endpoints.

20. **Historical GM public dialogue; potential dialogue on GE:** The extent to which a public dialogue around GE is needed or desirable depends on a broad range of factors including (but not limited to): scope for influence; buy-in of those with the power to effect change; the extent to which GE differs substantially from existing technologies. A Sciencewise-commissioned review of public dialogue on GM crops and food[13] concluded that dialogue is more useful when challenges rather than technologies are discussed, e.g. how can we produce food sustainably?

21. **Costs; benefits:** GE is still evolving; it is difficult to predict accurately all the costs and benefits. On initial indications, it will deliver greater value for both public research investment and commercial biotechnological R&D by enabling faster and cheaper experiments. It could also reduce unwanted genes in the environment[14].

22. **Particular issues for GE – biodiversity; shared benefits:** GE techniques used to generate new plants (and animals) will not necessarily affect biodiversity; introduction and management of resulting varieties and their interaction with the wider environment will be key. GE has potential to improve biodiversity within species, e.g., re-introducing traits from crop wild relatives lost through conventional breeding[15]. As a form of genetic R&D, GE would fall also within existing access and benefits sharing frameworks where these apply in law[16].

Genome editing in animals

23. **State of the art/direction of travel:** We are seeing increasing number of applications and awards proposing the use of CRISPR methodology due to its ease of application, very diverse uses (e.g. knockout and site-directed mutagenesis) and the precise targeting that it offers.

24. GE techniques are relatively simple, however screening for the correct change and checking for off-target effects remain the most time-consuming aspects of this research. Consistent use of standardised lines for GE is also critical to prevent genetic drift impacting upon results. Research Council investments stand to play an important role in mitigating risks of genetic drift and incomplete validation. For example, MRC

[10] For example antibiotic resistance genes, as used in conventional GM
[11] http://wyss.harvard.edu/staticfiles/newsroom/pressreleases/Gene%20drives%20FAQ%20FINAL.pdf
[12] http://www.oxitec.com/health/our-products/aedes-agypti-ox513a/ongoing-field-trials-of-ox513a-aedes-aegypti/
[13] 2011; http://www.sciencewise-erc.org.uk/cms/assets/Uploads/Talking-about-GM.pdf
[14] For example, the aforementioned antibiotic resistance markers (see paras. 2 & 6)
[15] The importance of wild relative traits is described here: http://www.cwrdiversity.org/ , cited as of strategic importance to food security.
[16] For example, where nations are signatories to the Nagoya Protocol – in the EU, enshrined in law via the ABS Regulation

investments at Harwell[17] have the capability to deliver complex GE mice that have been appropriately validated and phenotyped. Harwell is a major contributor to the International Mouse Phenotyping Consortium (IMPC) and is making a globally significant contribution to the production of well-characterised knockout mice which will continue to serve as a key resource to the international research community. An IMPC working group has, over the last 3 years, worked to progress and refine GE tools and protocols in the mouse. This working group will continue to work internationally to refine high-throughput protocols for (1) NHEJ[18]-mediated indels, (2) precise sequence deletions and (3) point mutation insertion via HDR[19]. They have developed a Gold Standard allele QC including the assessment of the risk of off-target integration

25. **Endpoints:** It is not yet clear how many entirely new opportunities may be opened up. Many of the aims, e.g., new breeds of livestock, will be the same as for conventional breeding or older GM techniques but GE should allow researchers to get there faster, at lower cost, with greater precision.

26. **Animal welfare; 3Rs; special considerations:** The same welfare and ethical considerations should apply to animals altered through GE approaches as those produced by conventional breeding. However the increased specificity of new GE methodologies and improvements in GE techniques should reduce the welfare and economic costs of animal experiments. Less time, and importantly fewer animals, would be needed to generate lines.

27. **Direct or indirect influence of public/current level and focus of public debate:** BBSRC and MRC have clear policies on the use of animals in medical research, informed by public consultation. We view increased efficiency and precision in GE as positives, since both will act to reduce the numbers of animals required to generate the lines used in experiments.

Genome editing in microorganisms

28. Endpoints; continuity with synthetic biology: As with plants and animals, GE is being applied mainly in research at present, but there are examples of microorganisms produced by GE technology being used in industrial biotechnology and synthetic biology. In these instances they can replace conventional GMOs and have the potential to produce fuels and higher value products such as chemicals and flavourings[20][21]. Accurate circuit design and metabolic pathway engineering are synthetic biology aims: by providing 'designer nucleases' for engineering (alongside current highly advanced DNA synthesis capabilities), GE has enabled precision engineering of cells with novel pathways and properties. Potential end-points would be those envisaged for synthetic biology[22].

29. **Biosafety; biosecurity:** Considerations are unlikely to be significantly different in degree or in kind from other R&D using microorganisms, however the consequence of factors such as reduced traceability should be explored.

Biomedical research and human applications

30. **State of the art/direction of travel:** We welcome innovation in GE as an established means of manipulating genomes for biomedical research and also its potential for therapeutic treatment of diseases with their origin in the genome, particularly Mendelian diseases (those linked to mutations in a single gene). Much currently funded research involves the use of GE techniques to model disease, understand gene function in normal and pathophysiological settings, and to understand if gene correction in a model system suggests this therapeutic approach may be viable.

31. Researchers are now contemplating how they could use the potentially higher degree of specificity/precision of intervention to cure human genetic disease - either through non-hereditable intervention with somatic cell therapy, or through hereditable genome fixes to prevent onward transmission of genetic risks (be they absolute or possible). The latter brings with it the concept that we will have introduced a man-made genetic fix into the human gene pool, even if that corrected sequence is based on what might otherwise be a natural

[17] MRC Mammalian Genetics Unit and MRC Mary Lyon Centre: https://www.har.mrc.ac.uk/
[18] NHEJ: Non-Homologous End-Joining
[19] HDR: Homology Directed Repair
[20] https://amyris.com/
[21] http://oxfordbiotrans.com/products/
[22] https://connect.innovateuk.org/documents/2826135/3815409/Synthetic+Biology+Roadmap+-+Report.pdf/fa8a1e8e-cbf4-4464-87ce-b3b033f04eaa

sequence (i.e. it is outside of a natural sequence in context). There are some genetic diseases that "conventional" gene therapy will struggle to address for technical reasons for example Duchene Muscular Dystrophy, in which the size of the Dystrophin gene makes it difficult to express using the currently available gene therapy vector systems. As with all gene therapies there are technical hurdles that need to be overcome before they can be trialled in humans. The MRC Translation funding schemes are yet to see a large number of applications attempting to use GE for human therapeutic interventions which reflects the stage of the science.

32. The challenges of getting to this position as a human therapeutic approach are many-fold and are highlighted in the Hinxton Group statement. The ease and rapidity of making (specific) change has shifted, bringing human application closer, but there are significant challenges to the existing regulatory framework and a need for a societal debate around the ethical concerns before implementation of such biologically complex interventions could be considered. As with other advanced therapies, the costs will be high in the first instances and many of the challenges found in regenerative and stratified medicine, such as access and cost, will need to be played out.

33. **'Drivers' and 'obstacles'/ endpoints:** As outlined above (see 24) validation is time-consuming but essential. Drivers remain the ability to produce increasingly accurate models of human disease.

34. **Direct or indirect influence of public/current level and focus of public debate:** There appear to be two foci for current debate: modification of embryos for research and modification of germ cells and/or embryos as a therapeutic intervention to combat Mendelian and other diseases. Another point for debate is the extent to which the gene-therapy endpoints of GE research can be of benefit to countries with relatively low national healthcare investment and limited infrastructure and consequently the extent to which the UK should allocate its investments to maximise benefit internationally.

35. **Impact on animal lives/ expected contribution/inhibition to the '3Rs':** As outlined above (see 26) we consider that GE technologies may make a contribution to the 3Rs in a number of ways, these may include:
    - Less need for crossing in lines to generate mice of interest, which would reduce animal numbers
    - More accurate targeting, which should reduce numbers of animals required to generate lines of interest
    - Fewer off-target effects, which should improve accuracy and reliability of knockout models

36. However, it is important to consider that the ability to create single point mutations as well as make knockouts may also increase the potential for use of animal models to accurately model human disease.

Military and security considerations

37. BBSRC and the MRC recognise that across the breadth of research we support, there is a risk that the results of some types of research could potentially be misused and pose a threat to public health and safety, agricultural crops and other plants, the environment and security. The Research Councils already have robust governance procedures for the research that we support and existing regulatory frameworks could also cover this technology. We believe that a system based primarily upon self-governance by the scientific community, but drawing on the inputs of other key stakeholders, will ultimately provide the most effective means of managing risks of misuse. To support researchers in identifying and managing the potential risk we have agreed a common joint position[23], with the Wellcome Trust, on managing risks that the outputs, (knowledge, products and/or technologies) resulting from research could be misused for harmful purposes.

38. Central to our position is the need for an inclusive and open discussion throughout the lifecycle of a study, with researchers actively considering the potential risk at the outset and funders addressing risks as part of the peer review and assessment process. An ongoing dialogue is essential between researchers, funders and other relevant authorities, to consider any change in the status of an identified dual use risk or any new risks that may emerge during the course the research project that were not anticipated in the original application.

39. Identification of areas of GE research where the risk of misuse may be particularly high could be helpful in supporting researchers in considering risk. However, due to the rapid pace of change we would recommend that any areas the working group may wish to highlight should be presented as illustrative.

[23] http://www.mrc.ac.uk/research/research-policy-ethics/managing-risks-of-research-misuse/