Blurring the Germline: Genome Editing, Transgenerational Epigenetic Inheritance and Urban Planning.

Tim Lewens
University of Cambridge
Department of History and Philosophy of Science
Email: tml1000@cam.ac.uk
Blurring the Germline: Genome Editing, Transgenerational Epigenetic Inheritance and Urban Planning.

Abstract

Until now, bioethical discussion of germline interventions has focused more or less exclusively on changes to the genome. But sperm, eggs and embryos are made up of more than genes, and there are indications that changes to non-genetic structures in these elements of the germline can also be inherited. It is, therefore, a mistake to treat phrases like ‘germline inheritance’ and ‘genetic inheritance’ as simple synonyms. This error is given additional weight by the recent advent of technological approaches that are described as forms of ‘epigenome editing’, which could potentially attract just the same forms of ethical concern as germline genome editing. Moreover, additional research on non-genetic inheritance draws attention to a variety of means whereby adults can transmit traits to their offspring that bypass the germline altogether. How, then, should bioethical discussion be updated to take account of these forms of non-genetic inheritance? This article argues that research on various forms of non-DNA-sequence based inheritance undermines the notion that there is some special form of ethical concern that falls on germline interventions in general, and on interventions to the nuclear genome within the germline in particular.

Keywords

Genome Editing, Mitochondrial Donation, Transgenerational Epigenetic Inheritance, Germ Cells, Germline Inheritance, Epigenome Editing.

1. Epigenetics and the Germline

Bioethicists are beginning to pay attention to research on non-genetic forms of inheritance. Thus far, they have tended to examine how epigenetic inheritance may complicate public health ethics. In particular, they have pointed to ways in which traditional public health concerns may need to address the impacts of a variety of nutritional and environmental factors on the health of multiple generations (e.g. Loi et al 2013, Juengst et al 2014, Dupras
et al 2014, Del Savio et al 2015). This article addresses a related question, which has not yet received sustained ethical discussion, but which also concerns the relationship between epigenetics and inheritance. Do we need to rethink our understanding of the ethical significance of germline interventions, in the light of work that draws attention to a variety of non-genetic mechanisms that influence how traits reappear across generations? This introductory section sketches, in very brief terms, some reasons for thinking the answer may be yes. The remainder of the article argues in more detail that the very idea of the ‘germline’ is one whose ethical, but not scientific, significance is being eroded in the light of empirical and conceptual work on non-genetic inheritance.

A recent review of non-genetic forms of inheritance opened with the claim that, ‘It is now clear that inheritance not based on DNA sequence exists in multiple organisms, with examples found in microbes, plants, and invertebrate and vertebrate animals’ (Miska and Ferguson Smith 2016: 59). This means that if we want to understand the processes by which differences between organisms of one generation are reflected in similar differences between their offspring, we must sometimes look beyond DNA. The same reviewers, in spite of the admirable caution they urged with respect to our knowledge of the mechanisms and significance of non-genetic inheritance in mammals, also indicated the potential relevance of non-DNA-sequence-based forms of inheritance for our own species. These forms of inheritance, they say, ‘have major implications for human health’ (63).

The catch-all terms ‘non-genetic inheritance’ and ‘non-DNA-sequence-based inheritance’ encompass all non-genetic processes by which differences between adults reliably reappear in their offspring. One example of a form of non-DNA-sequence-based inheritance with obvious importance in our own species is learning. Children observe their parents, and in so doing they come to resemble them in various ways. This example of behaviourally-mediated inheritance works independently of what is packaged into sperm and egg cells, but there are also non-DNA-sequence-based forms of inheritance that do operate via the contents of sperm and egg, hence which developmental biologists class as cases of non-genetic germline inheritance.
A different pair of scientific reviewers, a little more bullish than Miska and Ferguson-Smith, have claimed that, ‘Evidence for germline-dependent non-genetic inheritance of acquired traits in mammals has accumulated in neuroscience, behavioural neuroendocrinology, environmental toxicology and nutritional science’ (Bohacek and Mansuy 2015: 641). The work they review draws attention to the possibility, perhaps the probability, that changes to organisms that occur across their lives can be passed onto offspring via changes to gametes, yet not via changes to DNA sequence.

This type of work raises interesting questions about how ethicists are supposed to understand the ethical significant of germline interventions. Until now, discussion of such interventions has focused more or less exclusively on changes to the genome within the germline. This is hardly surprising, because in recent years these discussions have largely focused on the ethics of genome editing technologies. But sperm, eggs and embryos are made up of more than genes, and there are indications that non-genetic changes in these elements of the germline can also be inherited. In spite of increasing scientific attention to such forms of non-genetic germline inheritance, we will see in this article that bioethical discussion typically presupposes that phrases like ‘germline inheritance’ and ‘genetic inheritance’ are simple synonyms.

At the same time as researchers from developmental biology and genetics are drawing attention to instances of non-DNA-sequence-based inheritance, DNA sequence has remained the paramount focus of attention when reproduction is discussed in ethical and regulatory circles. In the UK, for example, only what are called ‘permitted eggs’ and ‘permitted sperm’ may be used for reproductive purposes. The HFEA Act (2008)—one of the primary legal tools regulating reproductive technologies in the UK—defines a permitted egg as one:

(a) which has been produced by or extracted from the ovaries of a woman, and
(b) whose nuclear or mitochondrial DNA has not been altered. (Human Fertilisation and Embryology Act 2008)

It gives a parallel definition for ‘permitted sperm’.
The HFEA Act tells us that it is alteration of DNA—rather than other cellular structures within the egg—that makes that an egg impermissible for reproductive purposes. This regulatory tool has the function of preventing germline alterations. But work on epigenetic inheritance suggests that alterations to inheritable structures within sperm and eggs other than DNA sequence might occur as downstream consequences of changes that occur during the lifetimes of humans as they develop. Changes to inheritable non-DNA-sequence structures of sperm and egg fit the general scientific understanding of what it means to affect the ‘germ line’, research indicates they may be quite easily induced by a range of environmental changes, and yet they are not explicitly prohibited by legislation. This presents bioethicists with a question: if forms of human intervention on developmental and environmental processes turn out regularly to induce alterations to the germline, how should this inform the general ethical disapproval that falls on the prospect of germline modification?

This question is rendered especially pressing by the recent advent of technological approaches that are described as forms of ‘epigenome editing’. Liu et al (2018) describe a modification of the CRISPR/Cas9 genome editing approach, which allows the ‘editing’ not of DNA sequence itself, but of the methylation patterns that affect gene action. They develop a proof-of-concept for the therapeutic use of a form of epigenome editing to combat Fragile X syndrome, a common form of intellectual disability in males. Liu et al applied their techniques to cell lines in vitro, which were then transplanted into the brains of mice. Their work did not, then, directly influence the mouse germ line; yet there is no reason in principle that prevents the germline application of this form of epigenome editing technology.

In October 2015, the UNESCO International Bioethics Committee (IBC) produced an ‘update’ to its reflections on the human genome and human rights (IBC 2015). It noted that, ‘We are human because of the interplay of many biological, historical, and cultural determinants, which preserve the feeling of our fundamental unity and nourish the richness of our diversity’ (4). This interactive, multi-faceted conception of what makes us human is, in the IBC’s view, ‘why the human genome is one of the premises of freedom itself and not simply
raw material to manipulate at leisure’ (ibid.). In a section entitled ‘Germline Modification’, the IBC went on to argue that ‘interventions on the human genome should be admitted only for preventive, diagnostic or therapeutic reasons and without enacting modifications for descendants’ (14). The research we have briefly alluded to on epigenetic inheritance is, of course, consistent with the IBC’s insistence that we are made by processes that are ‘biological, historical and cultural’. Moreover, we will see over the course of this article that the same epigenetic work reminds us that the ‘biological’ itself involves far more than simply nuclear DNA. Indeed, this work draws attention to forms of inheritance that are biological, historical and cultural all at once. But the very lines of reasoning that reinforce this rich interactive conception of growth and inheritance also undermine the notion that interventions to the nuclear genome within the germline should attract unique forms of ethical opprobrium, or so this article will argue.

Thus far we have only seen a flavour of the issues raised by work on non-genetic inheritance for our ethical appraisal of germline interventions. The remainder of the article proceeds as follows. Section two gives a reminder of the bad ethical odour that continues to be hang around the prospect of interventions to the human germline. Section three begins the work of asking what scientists mean by ‘the germline’. It argues that it is a mistake simply to equate ‘the germline’ with transmitted genetic material. Within scientific communities, the notion of the germline is instead understood by reference to lineages of germ cells. Section four briefly surveys research indicating the importance of forms of non-genetic inheritance which sometimes are, and sometimes are not, mediated by germ cells. Section five establishes that bioethical discussion often understands both the notion of the germline, and the notion of germ cells, in ways that depart significantly from the mainstream scientific understanding of those terms. There is, then, a tension between a comparatively narrow understanding of ‘the germline’ in bioethical circles, where ‘germline’ inheritance is equated with ‘genetic’ inheritance, and a broader understanding of ‘the germline’ in scientific circles, where forms of non-genetic germline inheritance are widely recognised. Section six then raises the question of how this tension should be resolved. It recommends that we look in detail at the background reasons given for subjecting genetic germline interventions to heightened ethical scrutiny, in order to assess whether there are salient ethical features that apply to these interventions, which do not arise in the context of
inheritable changes to non-genetic structures within the germline. It makes the case for thinking there are no such salient differences. Sections seven and eight then consider, and reject, two potential responses to this effort to blur the ethical significance of germline genetic interventions, one based on the alleged impact of genomic changes on ‘identity’, the other on the irreversibility of genomic alterations.

2. Ethics and the Germline

High-profile international agencies, as well as groups of leading scientists, have regularly expressed ethical concern at the prospect of interventions to the human germline. This section does nothing more than exhibit a few examples of such disapproval.

In April 2015 the NIH announced that it:

...will not fund any use of gene-editing technologies in human embryos. The concept of altering the human germline in embryos for clinical purposes has been debated over many years from many different perspectives, and has been viewed almost universally as a line that should not be crossed... [T]he strong arguments against engaging in this activity remain. (NIH 2015)

This form of ethical disapproval is indeed old. Article 24 of UNESCO’s original 1997 Declaration on the Human Genome and Human Rights recommended the ‘identification of practices that could be contrary to human dignity, such as germline interventions’ (UNESCO 1997). We have already seen that when the UNESCO IBC updated this declaration, it again expressed concerns about ‘interventions on the human genome’ that might result in ‘modifications for descendants’. Here the IBC followed the Oviedo convention, article 13 of which states that:

An intervention seeking to modify the human genome may only be undertaken for preventive, diagnostic or therapeutic purposes and only if its aim is not to introduce any modification in the genome of any descendants. (Council of Europe 1997)
The statements exhibited above indicate comparatively rigid stances against germline interventions. Other more recent discussions take milder positions which hint that, were a favourable risk assessment to be forthcoming, some germline interventions might be acceptable. In 2015 a prominent group of scientists working with genome editing asserted a fairly strong opposition to germline modification:

Many oppose germline modification on the grounds that permitting even unambiguously therapeutic interventions could start us down a path towards non-therapeutic genetic enhancement. We share these concerns. (Lanphier et al. 2015: 411)

Earlier in the same article, however, they had noted that:

In our view, genome editing in human embryos using current technologies could have unpredictable effects on future generations. This makes it dangerous and ethically unacceptable. (410)

The unstated implication is that genome editing in embryos might eventually become acceptable as the technologies improve and the uncertainty associated with them decreases.

The Hinxton Group also discussed the regulation of genome editing technologies when used at early stages in human development, a time when ‘any such intervention might reasonably be assumed to integrate into the germline, and therefore have the opportunity of being passed down to future generations’ (Hinxton Group 2015). Its conclusion was that, ‘when all safety, efficacy and governance needs are met, there may be morally acceptable uses of this technology in human reproduction’. A joint statement of September 2015 on genome editing by a number of important UK research institutes and research funders also noted, in a similarly flexible manner, that the use of genome editing in ‘human germ cells or embryos’ was unlikely to be permissible ‘at present’ in the UK and Europe, but that the ethical issues it raises ‘need to be anticipated and explored in a timely and inclusive manner as the basic research proceeds and prior to any decisions about clinical application’
(Academy of Medical Sciences et al 2015). These various groups all indicate that while germ-cell and embryonic interventions are currently prohibited by law, we may in time we may be able to justify a lifting of these prohibitions.

As the Nuffield Council on Bioethics has also noted in its ethical review of genome editing (NCOB 2016), the language of these statements is highly divergent (see also Bonnickson 1998 for much earlier reflections on such diversity, and Scott and Wilkinson 2017 for incisive analysis). They sometimes discuss interventions in the inheritable genome, they sometimes look more generally at interventions in the germline, and they sometimes write in even more general ways of interventions that can affect an individual’s descendants. For example, the US National Academies’ report on what they called ‘mitochondrial replacement technologies’ notes that the FDA, in its statement of task provided to the committee, defined ‘germline modification’ as ‘human inheritable genetic modification’ (National Academies of Sciences, Engineering and Medicine 2016). The US National Academies themselves, in their report, also equated ‘germline modification’ with ‘human inheritable genetic modification’. But we have already seen indications that, within the scientific community, there is considerable debate over the question of whether there can be inherited modifications—including inherited modifications to germline cells—that do not involve changes to genes.

The American Association for the Advancement of Science’s (2000) discussion of ‘Human Inheritable Genetic Modifications’ begins by avowing concern with the ‘the scientific, ethical, religious, and policy issues associated with interventions in the human germ line.’ Yet this comparatively broad brief quickly narrows:

This report assesses the scientific prospects for inducing controlled inheritable genetic changes in human beings and explores the ethical, religious, and social implications of developing and introducing technologies that would change the genetic inheritance of future generations. (Frankel and Chapman 2000)
In other words, the report’s authors assume that there is no difference between investigating interventions in ‘the human germ line’, and assessing ‘inheritable genetic changes’.

As the next section makes clear, these notions are not synonymous. We need to distinguish three increasingly narrow concepts: first, there is a very general notion of (to borrow the UNESCO IBC’s language) interventions that might result in ‘modifications for descendants’; second, there is a narrower notion of achieving this via alterations to the germline; and third, there is an even narrower notion of changing the germline via the specific method of inducing genetic changes.

3. The Germline in Scientific Work

What is meant by phrases like ‘germline engineering’, ‘germline intervention’ or just ‘the germline’? In a helpful background document prepared for the Nuffield Council on Bioethics, Frankel and Hagen state that, ‘The term “germline” refers to genetic material that is hereditable from parent to child’ (Frankel and Hagen 2011). The Oxford English Dictionary instead defines the ‘germ line’ as ‘a series of germ cells each descended from earlier cells in the series, regarded as continuing through successive generations of an organism’ (OED online 2018). The OED definition differs importantly from Franken and Hagen’s definition, because germ cells evidently contain far more than just genetic material. Defining ‘the germline’ in terms of ‘genetic material’ characterises it in a narrower way than if we define it as a series of germ cells.

In the face of such disagreement over what ‘the germline’ is, it makes sense to look directly at work in fields like developmental biology and genetics for clarification. A detailed position paper on the ethics of genome editing from the American Society of Human Genetics gives no explicit definition of ‘germline’, but it does define ‘germline genome editing’ as ‘genome editing that occurs in a germ cell or embryo and results in changes that are theoretically present in all cells of the embryo and that could also potentially be passed from the modified individual to the offspring’ (Ormond et al 2017: 169). This supports the OED’s
suggestion that we need to understand the germline in terms of germ cells, rather than just genes.

Unlike the term ‘germline’, the term ‘germ cell’ often receives an explicit definition in technical biological contexts. Germ cells have a special role in reproduction. One of the leading labs working on germ cells explains on its website that:

Germ cells, the precursors of sperm and eggs, are immortal in the sense that they generate a whole organism upon fertilisation and through them provide an enduring link between all generations, while the body cells perish with each individual. (Surani 2018)

It is also easy to find statements that germ cells are involved in the transmission to future generations of more than DNA. As Schedl puts it, ‘Germ cells are a central component of sexual reproduction in animals. They are the route by which the genome and cytoplasmic components are transferred to the next generation’ (Schedl 2001: 837; emphasis added).

The function of germ cells, then, is reproductive. They are frequently characterised in exceptionally rich terms, by spelling out when and how they arise as the early embryo develops. Wylie and Anderson, in a collection on mouse development, tell us that ‘Germ cells are the embryonic precursors of the gametes. They are set aside from the somatic cell lineages early in the development of most species’ (Wylie and Anderson 2002). Similarly, Schedl defines them as the cells that ‘will form gametes’ (Schedl 2001). These domains of biology are full of detailed descriptions of the point in development at which germ cells appear, and how this occurs.¹

¹ The following is fairly typical: ‘In the mouse, the germ cells, once they have formed, migrate through the tissues of the embryo to the gonad primordia...where they coassemble with somatic gonadal cells to form the sex cords’ (Wylie and Anderson 2002). Consider also Surani et al’s claim that ‘Germ cells are highly specialized cells established by a specific transcriptional program that includes repression of the somatic fate’ (Surani et al 2007: 747).
The AAAS’s paper on ‘Human Inheritable Genetic Modifications’ defines germ cells as ‘The reproductive cells of an organism, i.e., the sperm and egg cells’ (Frankel and Hagen 2000). The American Society for Human Geneticists also seems to take the view that while gametes are germ cells, the cells of undifferentiated early embryos are not: only this interpretation can explain why it equates germline genetic engineering with genetic intervention in ‘germ cells or embryos’ (Ormond et al 2017). However, from the strict perspective of developmental biology, gametes are not germ cells. As Surani’s lab describes the matter on its website, germ cells are ‘the precursors of sperm and eggs’ (Surani 2018). The zygote, and the cells of the very early embryo, are not germ cells either. At this very early developmental stage germ cells have not yet been formed.

When, in the early 2000s, Martin Johnson summarised the state of research in this area he remarked that, ‘In mammals, it is thought that the totipotentiality which characterises germ cells emerges anew with each generation at some point during the embryogenic process, which is when the first evidence of mammalian germ cells can be found...’ (Johnson 2001). In other words, germ cells (in mammals at least) are re-established via early developmental processes in embryos. In mammalian species there is no uninterrupted lineage of germ cells descending across generations.

We have seen, then, that developmental biologists and geneticists do not usually think of embryos and gametes as germ cells. They do, however, describe them as elements of the germline. Gapp and Bohacek, for example, comment that ‘life experiences can induce epigenetic changes in the germline (sperm and eggs)’ (2017: 1). Moreover, while the germline is evidently not equated with an uninterrupted sequence of germ cells that descends down generations—for there is no such uninterrupted sequence in mammals—there is a link between the concept of the germline and the concept of a germ cell. An inherited germline intervention—whether it is wrought directly on germ cells, or on sperm, eggs or the cells of early embryos—is one that is passed across generations via corresponding changes to germ cells sensu stricto. That is why, in animals that can learn from their parents, changes to learned traditions do not count as germline modifications, even though they may be inherited.
4. Non-genetic germline interventions

There is disagreement among scientists regarding the nature and significance of epigenetic inheritance, and there is also a widespread sense that its significance is frequently exaggerated in communications to non-specialist audiences (Grossniklaus et al 2013, Meloni 2014, 2015, Meloni and Testa 2014). In 2014, Heard and Martienssen concluded a review in sceptical fashion, arguing that ‘although much attention has been drawn to the potential implications of transgenerational [epigenetic] inheritance for human health, so far there is little support’ (106). An older review from Whitelaw and Whitelaw (2008) noted in a rather undecided way that, ‘In some cases germline epimutations in humans appear to be inherited from the parent, raising the possibility of transgenerational epigenetic inheritance via the gametes.’ Miska and Ferguson-Smith, in their review, take a cautious line noting that while there is a clear case for non-genetic inheritance in many plant and animal organisms, ‘In mammals, the molecular mechanisms have been challenging to elucidate’ (2016: 59). They are confident of a variety of phenomena in humans whereby alterations to epigenetic states result in the inheritance of phenotypes over a small number of generations. They are not convinced, as yet, of the strength of evidence that allows us to categorise how this occurs.

Others seem far surer of both the existence of some important forms of epigenetic inheritance in mammals, and of the mechanisms that explain it. In a 2017 review Gapp and Bohacek argued that:

Life experiences can induce epigenetic changes in mammalian germ cells, which can influence the developmental trajectory of the offspring and impact health and disease across generations. While this concept of epigenetic germline inheritance has long been met with skepticism, evidence in support of this route of information transfer is now overwhelming, and some key mechanisms underlying germline transmission of acquired information are emerging. (Gapp and Bohacek 2017: 1)

When considering the nature of non-genetic inheritance, and the apparent disputes over its significance that characterise the remarks quoted above, it is useful to bear in mind a
threelfold distinction between different types of non-genetic inheritance process (Miska and Ferguson-Smith 2016). Two involve inheritance mediated by germ cells, and a third involves inheritance that bypasses germ cells. We will begin with the third type.

Non-Germ Cell-based Inheritance

It is possible for parents to affect their offspring in ways that result in the reconstruction of resembling traits in the new generation, even when the germ cells they transmit to those offspring play no role in explaining this aspect of parent/offspring inheritance. For example, Bohacek and Mansuy suggest that, ‘The composition of the seminal fluid [rather than the gametes contained within it], which is transferred to the female with sperm during mating, can change with the environment and influence the offspring independently of sperm’ (2015). Whitelaw and Whitelaw give a very different example in rats: ‘The offspring of greying mothers exhibit greying but the offspring of greying fathers do not. This maternal effect was initially thought to be controlled by a dominant ‘greying’ gene...but an elegant study involving caesarean sections and foster nursing showed that it was the result of transmission from the mother to newborns of a murine leukaemia virus (probably via the milk) that causes greying’ (Whitelaw and Whitelaw 2008: 276).

In both of these cases, when offspring resemble their parents it is not because of what is passed on via genes, nor is it because of what is passed within gametes more generally. Some of the most obvious and uncontroversial cases of inheritance that bypasses germ cells involve learning and other forms of social transmission (Lewens 2015a). Here, too, offspring end up possessing the same traits as their parents—we see a ‘like resembles like’ phenomenon—albeit mediated by culture. Evidently traditions of many kinds are transmitted in humans in this way, but there is also considerable evidence for the existence of learned tradition in animals, and work on animal culture is burgeoning (Avital and Jablonka 2000).

There is, then, an enormous diversity of non-genetic processes whereby offspring develop in such a way that they acquire traits similar to those of their parents. This diversity explains why there is no agreed-upon definition of ‘epigenetic inheritance’. Cultural inheritance is
rarely labelled as ‘epigenetic inheritance’—the term is more usually reserved for inheritance that is explained via alterations in such things as methyl groups, histones, small RNAs and other molecules packed into gametes—but there is no special bright line that justifies the exclusion of cultural inheritance from the domain of epigenetics. Consider, for example, that one of the most widely disseminated examples of apparent epigenetic inheritance—namely Meaney’s work on inheritance in rats—seems to rely on a hybrid mechanism that involves both epigenetic marks and social behaviour. (It is, to reinvoke the UBESCO IBC’s update statement, simultaneously ‘biological, historical and cultural’.) Meaney’s work is contentious, but it appears that methylation on DNA, which affects stress responses in maturing pups, is itself affected by the postnatal grooming behaviours of dams (Champagne and Meaney 2007). Germ cells are not involved here: instead, a pattern of methylation that leads to a behavioural response to stress is reconstructed in offspring via the social behaviour of mothers.

Germline Epigenetic Inheritance

Biologists have distinguished two ways in which effects on germ cells can be inherited in offspring via non-DNA-sequence structures; transgenerational inheritance, and intergenerational inheritance (Miska and Ferguson-Smith 2016). Transgenerational inheritance involves changes to germline epigenetic structures persisting over multiple reproducing generations, in a way that continues beyond the initial event that first caused an epigenetic alteration. This would potentially give epigenetic systems of inheritance a number of similar properties—in terms of fidelity and longevity—to the genetic inheritance system. Intergenerational inheritance instead occurs when an environmental effect on (for example) a pregnant female directly affects not only the phenotype of the maturing embryo within the parent, but it also has an influence on the germ cells within that very embryo; that is, it also affects what the embryo will bequeath to its own progeny. The result is that an effect can potentially be observed in three generations—parent, offspring and grand-offspring—even if there is no tendency for the epigenetic alterations in question to be preserved faithfully across further reproductive cycles. This is why an important test for true transgenerational inheritance, as opposed to intergenerational inheritance, is to ask
whether epigenetic modifications persist into a fourth generation following an environmental insult to the organism.

There is debate over the extent to which transgenerational epigenetic inheritance has been demonstrated in mammals, although the effect seems well confirmed over many generations in plants. Scientists have tended to assume, for example, that epigenetic marks are ‘reset’ or ‘reprogrammed’ with the establishment of each new mammalian generation, with the result that in the case of methylation, for example, differences are not inherited across generations (Heard and Martienssen 2014). However, there are also indications that this resetting may not apply to all areas of the genome. There are also considerable problems in establishing with confidence that alterations to epigenetic marks, rather than alterations in genes, ground the explanation of trans-generational inheritance. One problem that Ferguson-Smith draws our attention to is, ‘the fact that DNA methylation is a mutagen that contributes to C to T transitions if not repaired’ (Grossniklaus et al 2013: 233). In other words, an environmental intervention that causes an alteration in an epigenetic methylation state might have, as a knock-on effect, a further alteration in the genome which could then persist over several generations. The upshot is that it is hard to tell, even if we observe a sequence that begins with an environmental change causing an epigenetic alteration, and which results in phenotypic changes that are transgenerationally stable, that we are dealing with non-genetic inheritance.

The question of how important transgenerational epigenetic inheritance is for our own species, and for other mammalian species, is consequently unclear. Yet it is abundantly clear that there is an important question to be asked about whether such transgenerational epigenetic inheritance is real, hence whether there are potential ways of affecting the germline, with effects across multiple generations, that do not require alterations to DNA.

5. The Germline in Bioethics

In bioethical literature we often find that definitions of key terms like ‘germline’ are not drawn directly from scientific work, but instead they are tailored to the specific context of ethical discussion. One clear example of tailoring a definition of the germline to suit the
needs of ethical discussion can be seen in the context of genome editing. Baltimore et al, for example, reported on a meeting in Napa, California, which was convened to determine how to use genome editing technologies, such as CRISPR, in a responsible way. They explained their understanding of ‘germline engineering’ in the following way:

The Napa discussion did not address mitochondrial transfer, a technique that does not use CRISPR-Cas9. Although characterized by some as another form of ‘germline’ engineering, mitochondrial transfer raises different issues and has already been approved by the Human Fertilisation and Embryology Authority and by Parliament in the United Kingdom and is being considered by the Institute of Medicine and the Food and Drug Administration in the United States. At the Napa meeting, ‘genome modification’ and ‘germline engineering’ referred to changes in the DNA of the nucleus of a germ cell. (Baltimore et al 2015: 37)

It seems that they used the facts that (i) the UK Parliament had legalised what they here call ‘mitochondrial transfer’ and (ii) their own meeting had the purpose of addressing a different technology, namely CRISPR, to justify defining ‘germline engineering’ by fiat as changes in the DNA of a cell nucleus. Yet these mitochondrial transfer techniques, if they work as intended, will allow women who would otherwise have had children with a high chance of being affected by mitochondrial disease to have not just healthy children, but healthy grandchildren and great-grandchildren, too. Moreover, the interventions affect cells in embryos or gametes, in such a way that their transgenerational effect is mediated via changes to germ cells. So by any normal scientific definition they are uncontroversial cases of interventions to the germline, even though they do not alter nuclear DNA.

Baltimore et al chose to exclude mitochondrial ‘transfer’ from the scope of ‘germline engineering’, in part because they took the view that it raised different ethical issues to

---

2 I use the term ‘mitochondrial transfer’ in scare quotes. This term gives the impression that only healthy mitochondria are transferred from the donated eggs of a healthy woman to the intended mother. In fact the donor contributes all cellular structures with the sole exception of nuclear material that comes from the intended mother (Lewens 2015b).
genome engineering. Some of the ethical issues prompted by mitochondrial transfer are indeed different to those raised by CRISPR, and the two techniques are entirely different. For example, one specific set of issues raised by mitochondrial transfer turns on the involvement of three, rather than the usual two, providers of genetic material for babies born from these techniques. But many of the issues raised are the same: in both cases we are dealing with interventions that carry uncertain levels of risk, whose clinical rationales are disputed, and whose effects may persist across multiple generations.

The UK Department of Health took a slightly different line on mitochondrial transfer and the germline, noting that, ‘There is no universally agreed definition of ‘genetic modification’ in humans. . . . The working definition that we have adopted is that genetic modification involves the germline modification of nuclear DNA (in the chromosomes) that can be passed on to future generations’ (Department of Health 2014). In other words, this did not count as genetic modification because no nuclear DNA was changed.

It may be that the UK Department of Health wished to avoid describing these techniques as ‘germline genetic modification’ on the grounds that the Oviedo convention (to which the UK is not a signatory, but which carries some authority all the same) prohibits techniques whose aim is ‘to introduce any modification in the genome of any descendants’. Professor Sally Davies, the UK Chief Medical Officer, tried to justify the Department of Health’s reasoning, when she was questioned by the House of Commons Science and Technology Committee:

Germline is anything that is done to DNA that goes through the generations, and mitochondria go from woman to child through the generations. This is clearly a germline modification because it passes through, but we needed to make the distinction between nuclear DNA, which makes us who we are and how we are – our personalities, heights, weights and whether or not we get baldness – and the 37 genes in the mitochondria which are about energy for the cell, and which we describe as the power pack. That was why we adopted that working definition. (Sally Davies, quoted in Scott and Wilkinson 2017)
Davies’s comments are hard to interpret, but they cement the thought that in contexts like these, far from phrases like ‘germline genetic modification’ being wholly scientific terms whose ethical significance then needs to be assessed, these apparently technical terms are being tailored to suit ethical ends. Davies thinks of the ‘germline’ not as a series of germ cells, but rather she links it narrowly to inherited aspects of DNA: ‘germline is anything that is done to DNA that goes through the generations’. She therefore agrees—because mitochondria contain DNA, and mitochondria are inherited via the maternal line—that interventions to embryonic mitochondria are germline modifications. But she is unwilling to describe these interventions as ‘germline genetic modifications’, because she thinks there is good reason to reserve that term for changes to nuclear DNA.

The grounds she gives are that it is the nuclear DNA—not the genes in the mitochondria—that ‘make us who we are’. So Davies used ethical considerations to inform the definition of an apparently scientific term, namely ‘germline genetic modification’. Moreover, these ethical considerations are highly contestable (Scott and Wilkinson 2017). The very fact that faulty mitochondrial genes can lead to serious illness means that they have far-reaching effects on the lived experiences and self-conception of people who have those genes. In at least one very important sense, mitochondrial genes do contribute to making people who they are.

6. Why do we care about the germline?

So far this article has established two claims. First, international bioethical discussion regarding the rights and wrongs of medical interventions in the processes of inheritance tends to focus overwhelmingly on alterations to genes, in particular on alterations to nuclear genes. Indeed, these discussions have a tendency to simply equate the notion of potentially problematic ‘germline’ interventions with interventions to DNA sequence, especially nuclear DNA. Second, we have also seen an emergent body of work raising a significant probability that inheritance can be mediated through a variety of non-genetic mechanisms. Some, but not all of these, also pass via the germline. Some persist for a small number of generations, others appear to have more significant trans-generational stability. The question, then, is how, if at all, discussion within bioethics should be broadened in the
light of what we know about these forms of non-genetic inheritance, including non-genetic germline inheritance.

There are three options for how we might respond to this question:

1. Retain an ethical focus on germline inheritance, and retain the focus on interventions to nuclear DNA sequence, by arguing that forms of non-genetic inheritance do not raise the same ethical concerns as interventions to nuclear DNA.
2. Retain an ethical focus on germline inheritance, but expand this focus to include non-genetic inheritance that is mediated via the germline, by arguing that all effects inherited via the germline—as opposed to forms of inheritance that are not mediated via the germline—raise a special set of ethical concerns.
3. In the light of work suggesting a variety of different means—some which move via the germline, some which evade it—by which inheritance across generations can be effected, argue that while the notion of the germline may do important scientific work, it does not do useful ethical work.

Evidently we cannot decide between these three options unless we examine why ethical concern has been expressed about germline interventions in the first place. Only then can we understand whether in fact interventions to the nuclear genome are the only ones with the potential to trigger the intended form of ethical concern, or whether (in the case of option 2 above) epigenetic alterations inherited via the germline might trigger the same concerns, or (in the case of option 3 above) whether all forms of inheritance that persists across generations—whether mediated via the germline or not—should attract the same form of ethical concern, with the result that the germline ceases to matter for ethical purposes.

Ethical concern sometimes focuses on the germline simply because of the potential for changes to germ cells to persist over more than one generation. In other words, ethical focus is not on the germline per se, but rather on the prospect of changes that are passed to future generations regardless of whether it is achieved via germ cells. Consider again the UNESCO IBC’s ‘Update’ Statement: ‘In several countries somatic gene therapy has received
ethical and regulatory acceptance because the genetic changes induced are not passed on to the next generation’ (IBC 2015). If we think the problem with germ-line interventions is simply that they have effects that are ‘passed on to the next generation’, then we potentially open up the whole range of inherited epigenetic modifications—both inter-generational and trans-generational—to precisely the same set of worries.

Sometimes, however, a richer set of concerns is raised. The NIH’s statement of April 2015 on genome editing notes that:

...the strong arguments against engaging in this activity remain. These include the serious and unquantifiable safety issues, ethical issues presented by altering the germline in a way that affects the next generation without their consent, and a current lack of compelling medical applications justifying the use of CRISPR/Cas9 in embryos.

The NIH here suggests three reasons for opposition to germline interventions in the genome: there are safety issues that cannot be quantified, future persons born as a result of these technologies cannot consent to their use (since the technologies must, by necessity, be used before the people they give rise to exist), and there is no medically compelling case to use them.

Again, these reasons apply to interventions in all mechanisms by which later generations come to resemble earlier ones, regardless of whether those mechanisms work via nuclear genes, epigenetic germline inheritance, or mechanisms of inheritance that bypass the germline. All of these mechanisms for inheritance affect members of future generations who are unable to offer consent. The need to ask whether there are compelling clinical rationales to intervene on these forms of inheritance is evidently raised in all of these cases, too. If we are dealing with genome-editing, the specific questions we need to ask include whether various forms of screening might deliver similar outcomes in terms of disease transmission, but at lower risk. If we are dealing with mitochondrial ‘transfer’, we can ask whether the alternatives of PGD, or even the provision of donor eggs, yield similar levels of benefit at lower risk. Turning to epigenetics, Sales et al recently reviewed the question of
whether epigenetic mechanisms might explain the transmission across generations of metabolic diseases. They noted that ‘Evidence that the isolated germ cell can mediate offspring disease was recently described by Huypens and collaborators, who utilized in vitro fertilization to demonstrate that germ cells harvested from mice exposed to nutritional factors (low-fat diet, normal diet, and high-fat diet...) are able to transmit metabolic phenotypes to offspring’ (Sales et al 2017: 561). We might eventually determine with confidence that epigenetic inheritance is implicated in the transmission of metabolic disease in humans. But it is clearly essential to ask, in this kind of case, whether it would be clinically feasible and responsible to intervene in these epigenetic germline processes to reduce disease risk. If what makes it wrong to intervene in the nuclear germline is ‘a current lack of compelling medical applications’, then the very same concerns will bar many other forms of intervention in other forms of inheritance.

An even more detailed ethical discussion can be found in the AAAS’s document on what they call ‘Human Inheritable Genetic Modifications’. They restrict the scope of their discussion to genes:

IGM [Inheritable Genetic Modifications], as used in this report, refers to the technologies, techniques and interventions that are capable of modifying the set of genes that a subject has available to transmit to his or her offspring. (Frankel and Chapman 2000)

While narrowing the scope of discussion at this point in their document, they also give a very general account of the concerns raised by these technologies that seems to directly implicate many non-genetic ways of intervening in the processes of inheritance:

The kinds of interventions that fall within the scope of the definition of IGM are those that raise the following core issues:

• They are interventions that hold out the prospect of increasing our control over the specific hereditary traits of the next generation and beyond if they succeed;
• They are interventions that make inheritable changes in the genes of surviving offspring, rather than interventions that simply select among offspring on the basis of their naturally inherited genes;

• They are interventions associated with scientific research, i.e., biomedical interventions, rather than social practices;

• They pose the risk of creating iatrogenic and other genetic harms. (Frankel and Chapman 2000)

There is an instability in the AAAS’s treatment of these issues. Their list of the concern-raising characteristics of intervention that fall ‘within the scope’ of the definition of IGM implies that the term ‘IGM’ might reasonably stretch to cover a very wide range of interventions that introduce inheritable changes to offspring, whether mediated by genes or not. The very same worries triggered by genetic modification are also triggered by epigenetic germline alterations, and by non-germline inherited alterations. In all of these cases, we raise the prospect of controlling the inherited traits of subsequent generations; we may aim to introduce changes in identified individuals, as opposed to selecting among several different offspring; well-intentioned medical intervention may inadvertently damage the health prospects of future generations; all of this may end up being done in the name of biomedical science; and it may thus be an iatrogenic form of harm.

If we return to the NIH’s worries about genome editing, then we find that the ethical problems it raises for this technology apply not only to genome editing, but to nutritional advice designed to provoke epigenetic modifications to the germline, and even to urban planning, or changes to the organisation of schooling and specified educational curricula, that are meant to better the lot of future individuals. Here, too, we find interventions whose effects are uncertain, whose effects may also persist over several generations, and to which those affected by the changes—since they may not exist until after these structures have been put in place—cannot consent.

These remarks—which use germline epigenetic inheritance to draw parallels between nuclear genome editing, public health advice relating to nutrition, and urban planning—may seem flippant. They are not intended to be. We have good reason to ask to very same kinds
of questions, whether we affect future people via their germ cells, their social and technical environments, or combinations of both. Urban planning, like genome editing and the provision of broad nutritional advice, has effects that are uncertain, persistent and hard to reverse. In all cases we should ask whether the interests of future people have been properly taken into account in our discussions, whether our uncertainties are properly reflected in a precautionary approach to action, whether these risky interventions are justified by genuine need, and whether we are recklessly introducing potentially harmful changes with long-term effects in blinkered service of a notion of technical progress. There is no special sin of germline intervention. That is not to say that the questions raised by germline interventions are unimportant. But reflection on the cases of nutritional advice and urban planning reminds us simultaneously that any rigid stance against intervention in the processes of inheritance is absurd, since such interventions are inevitable and pervasive, and that the true significance of taking a humble attitude to our influence over the inheritance of future generations extends well beyond the germline as usually understood.

Before concluding this essay, we must consider two potential responses that might re-establish alterations to the nuclear genome as deserving of special ethical oversight, hence which might justify the tendency to focus only on changes to nuclear genetic structures when we consider the rights and wrongs of germline interventions. The first concerns the allegedly special link between nuclear genes and ‘identity’, the second the supposed ‘irreversibility’ of changes to the nuclear genome.

7. Epigenetics and Identity

First, we might follow Sally Davies and others in suggesting that changes to nuclear genes, unlike other interventions on the determinants of inheritance, have a special impact on traits that are relevant to an individual’s identity. I have already indicated scepticism about this move. There is a perfectly good sense in which the experience of disease can be a remarkably strong element of an individual’s self-conception, hence of ‘identity’ in that important respect. To the extent that non-nuclear DNA affects disease phenotypes, it follows that we should also credit elements of mitochondrial genomes, and also inherited epigenetic structures, with a role in determining identity.
A charitable reading of what Davies most likely intended by her remarks—a reading suggested by Scott and Wilkinson (2017)—is that when she and others talk about ‘identity’, they are expressing concern with the potential for changes in the nuclear genome to exert very fine-grained control over positively valued cognitive, emotional, behavioural or physiological traits, as opposed to making large-scale differences to the presence or absence of disease traits.

The first thing to note in response to this alleged asymmetry between nuclear genetic and epigenetic determinants of inheritance is that it is an open question whether, and how often, nuclear genes will truly enable such fine-grained control over valued phenotypes. For example, to the extent that phenomena of epistasis turn out to be pervasive (in particular, the contingency of effects of gene substitutions on genetic backgrounds that can vary from individual to individual), as well as forms of pleiotropy (i.e. a gene having multiple effects on different phenotypes), then intervention on nuclear genes may be unwise as a method of trying to control phenotypic outcomes, because of the unpredictable effects of gene alterations on traits other than the target phenotypes of interest (see NCOB 2016: 4.43). Further, to the extent that the phenotypic effects of nuclear genes are very small in magnitude, then if we are aiming to control valued cognitive, emotional or physiological traits, then we may simply find that direct interventions on other developmental processes—related to such mundane things as nutrition, training or education—will be far more efficient (Lewens 2002).

Second, if our specific ethical worry about inherited nuclear genetic interventions relates to the potential for the enhancement of valued traits, then some germline interventions to the nuclear genome ought to be tolerated after all, so long as those interventions are also of a kind that go no further than affecting the presence or absence of disease. It is instructive to note that there are disorders of mitochondrial function that arise not from disorders of the mitochondrial genome, but rather from disorders in nuclear genes that influence mitochondrial function (NCOB 2012: 1.11). The clinical conditions of people with nuclear, compared with mitochondrial, genetic abnormalities can be very similar. It is hard to see how one might come to the verdict that inheritable interventions on the mitochondrial
genome are unproblematic by virtue of having nothing to do with ‘identity’, unless we also agree that the necessary inheritable interventions to the nuclear genome, when they seek to remove precisely the same kinds of inherited clinical symptoms, are also unproblematic in this respect. Reflection on this confused notion of ‘identity’ indicates that it is not interventions to the germline per se that we should be worried about, but at best a certain kind of fine-grained control over the phenotypes of future generations, which even alterations to germline nuclear genes may rarely enable.

8. The ‘Reversibility’ of Epigenetic Interventions

The Nuffield Council on Bioethics noted that opposition to genome editing has been especially intense ‘where scope for unforeseen consequences is considered to be great or editing is regarded as irreversible’ (NCOB 2016). Meanwhile, Bohacek and Mansuy, in their discussion of the epigenetic inheritance of behavioural phenotypes, reported that while foetal exposure to alcohol in rats could increase their sensitivity to stress over two or three generations, ‘Some of the inherited effects can be corrected by environmental enrichment in adolescent rat offspring, suggesting reversibility of symptoms’ (2015: 642). I argue in this penultimate section that the notion of ‘irreversibility’ is unlikely to be able to ground a general ethical asymmetry between genomic, and epigenetic inheritance.

The most obvious thing to point out is that even when genes are inherited across numerous generations, their phenotypic effects will often be modifiable by altering the environment of development. Many effects of germ-line genetic alterations are, as a consequence, ‘reversible’ in just the same sense that the stress sensitivity of Bohacek and Mansuy’s rats is reversible. Moreover, if genome engineering becomes a very powerful tool for intervening on nuclear germline DNA sequence, then in theory the very same technology that allows genomic changes in one direction will allow the same changes to be undone in the opposite direction. Of course, this does not mean our efforts at reversal will be successful, and it is also unlikely that we will be able to track down and intervene on all later individuals affected by an inherited genomic alteration. But this does not establish a sharp difference between genomic and epigenetic interventions to inheritance systems. Once we try to intervene on non-genetic inheritance structures, we might also find it hard to control all of
the effects of our actions. This is especially obvious when we think of modifications to the social and cultural determinants of inheritance in humans: if we institute new ways for designing long-lasting social housing projects, or if we issue sweeping new forms of public health advice that turn out to affect germline-inherited epigenetic structures, then it is wishful thinking to assume the effects of our actions on future generations will be easy to undo if we realise we have made mistakes.

We have seen widespread agreement that many epigenetic states, such as methylation patterns, are ‘reset’ during the processes of reproduction, with the result that there is a significant difference between nuclear and epigenetic inheritance with respect to the number of generations for which modifications can be expected to be stable. We have also seen that this appears not to be the case for all epigenetic states, and that there are live research question about the extent to which some forms of epigenetic inheritance are transgenerationally stable in mammals. Hackett et al, for example, noted that in mice there are ‘rare regulatory elements that escape systematic DNA demethylation in PGCs [primordial germ cells], providing a potential mechanistic basis for transgenerational epigenetic inheritance’ (Hackett et al 2013).

There is an important scientific distinction to be drawn, as we have seen, between intergenerational and transgenerational effects. Moreover, the jury is still out on the transgenerational nature of epigenetic inheritance in humans. Even so, it is unclear how the intergenerational/transgenerational distinction could allow us to establish a bright ethical line between interventions to germline DNA sequence and interventions that might affect other inheritable germline structures. Suppose our ethical concerns with germline genomic interventions rest on the worry that over-zealous meddling will adversely affect multiple future generations. This would hardly show that, since most interventions affecting epigenetic inheritance will have effects that persist for only two or three generations, we should be unconcerned with them. We need to pay due ethical attention to a motley assortment of forms of inheritance mediated by everything from the built environment to histones and small RNAs, via social behaviour and the constituents of seminal fluid. Some of these do, and some do not, affect the germline, but all have impacts on the development of future generations.
9. Conclusion

In scientific contexts, the notion of a germline modification tends to receive a comparatively strict definition: it is not simply any intervention that is likely to be propagated over several generations. It is an intervention that is likely to be propagated over anything from two to an indefinite number of generations (depending on whether it is inter-generationally, or trans-generationally stable), and which acts via a modification to germ cells. We have seen that there are strong reasons to think that germline modifications, in this strict scientific sense of the term, do not merely concern interventions to nuclear DNA sequence. And yet, bioethical discussion continues to make this assumption. We have also seen that the very reasons that have triggered concern over modifications to germline nuclear DNA sequence—concerns about the potential for poorly-understood, irreversible, multi-generational change; concerns about consent; concerns about the strength of clinical rationale for the uptake of new forms of technical intervention—do not merely apply to germline modifications when expanded to include epigenetic germline modifications. There is increasing evidence for a class of inherited modifications that do not proceed via the germline at all, and which trigger precisely the same set of ethical concerns. The result is that scientific work is teaching us that the concept of a germline modification is one with no distinctive ethical job to play.

This very point has been made a long time ago, and it has been made in the context of a consideration of non-nuclear inheritance. John Robertson argued, in the context of a very early evaluation of the ethics of cytoplasmic transfer that, ‘The labels of...germ-line and somatic gene therapy are no longer adequate to frame ethical discussion’ (Robertson 1998: 217). What is perhaps more interesting is the failure of laypeople to discern the importance of the distinction between germline and somatic interventions. A recent opinion-gathering exercise on the ethics of genome editing led by the Genetic Alliance and the Progress Educational Trust noted what it called ‘The missing distinction’:
There is a distinction in genome editing that is thought to be of vital importance in science, ethics, law and policy, which we expected would arise spontaneously during the course of workshop discussions. This was not the case.

We refer to the distinction between somatic genome editing (which results in changes that are not heritable by the next generation) and germline genome editing (which results in changes that are heritable by the next generation)....

The distinction did not occur to participants, even when workshop moderators attempted to steer discussion in a direction that would bring it to light. One of the experts who spoke in our workshops was asked to focus on this distinction, but even after this, the distinction was mentioned only fleetingly by participants during workshop discussions. (Genetic Alliance/Progress Educational Trust 2017)

The Progress Trust moves on to advise that people need to be educated in the existence and importance of this distinction. Isasi et al (2016) have also urged, again in an ethical context, that what is needed is ‘scientific understanding and precision in legal definitions of what constitutes an embryo and/or its germ line’, and that these are ‘essential to developing coherent policies’. If the suggestions of this paper are correct, the laypeople involved in the Progress Trust’s workshop may, on the contrary, have been right not to see any special ethical significance attached to the germline.

References


