

Gender-specific medicine in the genomic era

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Abstract

This article is intended to illuminate several important changes in our concept of gender-specific medicine in the genomic era. It reviews the history of gender-specific medicine, pointing out the changes in our perception of the nature of biological sex and our expanding knowledge of how it affects the phenotype. The old debate about ‘nature versus nurture’ is now largely resolved; the two are inextricably intertwined as a result of epigenomic regulation of gene expression; many of the resulting phenotypic changes are inherited and affect future generations. More accurate, rapid and cheaper methods of editing genomic composition are implementing a more sophisticated understanding of how genes function and how individual components of the genome might be added or eliminated to maintain health and prevent disease. As Venter predicted, the new discipline of synthetic biology, based on the creation and use of novel ‘designer’ chromosomes is an inevitable expansion of our ability to decipher the naturally occurring genome and the factors that control its expression. As we move with unexpected and stunning rapidity into our exploration and manipulation of the genetic code, our investigations must acknowledge the solidly established fact that biological sex will have a profound impact on the interventions we have made and will make in the future. Unfortunately, in spite of the recent urging of the National Institutes of Health (NIH) that sex be included as an essential variable in all levels of scientific investigation, genuine issues remain to be resolved before all scientists accept not only the importance of doing this, but also how to implement it.

Key words: gender, genomics, sex, male, female, synthetic biology.

‘The great and unlooked for discoveries that have taken place of late years . . . have all concurred to lead many able men into the opinion that we were touching on a period big with the most important changes, changes that would in some measure be decisive of the future fate of mankind.’: Reverend Thomas Robert Malthus

INTRODUCTION

Malthus [1] wrote his famous essay in 1798, ecstatic about the invention of the printing press and the age of enlightenment, citing the ‘unshackled spirit of inquiry that prevails throughout the lettered and even unlettered world’. His comments are as fresh and relevant to our own era as they were to his. Exactly like our eighteenth century ancestors, we find ourselves in the midst of an explosion of undreamed of wonders: robots whose behaviour improves with experience, computer-designed chromosomes that function to seize control of cellular metabolism, stem cells that are coaxed into developing miniorgans in culture and pictures of planets 5 billion kilometres away at the edge of the solar system,

planets whose atmospheres we may one day control and make habitable for human colonization. And like Malthus (whose work was directly relevant to the concepts of Charles Darwin himself), we herald the cascade of our stunning accomplishments over the past quarter century with a mixture of astonished awe and ambivalence about whether we will use our exponentially expanding power for the improvement of life on earth (and indeed, throughout the universe) or as Schramski et al. [2] have warned, for its impending extinction. Even the most untutored, superficial glance at the achievements of the last 15 years affirms that we are in a period of unparalleled accomplishment, most of it unimagined and all of it providing us with astounding potential to enhance and perfect who we are until we will be forced to consider new answers to the question of what it means to be human. Kurzweil [3] commented on what we are doing and what is happening to us by reminding us that the rate of our accomplishments is expanding exponentially. It is an error to conceive of a future in which the rate of discovery is similar to the one that preceded it, he warns: ‘the future will be far more surprising than most people realize, because few observers have truly internalized the implications of the fact that the rate of change itself is accelerating’.

Abbreviations: CRISPR, clustered regularly interspaced palindromic repeat; DSD, disorders of sex development; NIH, National Institutes of Health.

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Our headlong plunge into ever more amazing technological triumphs is not without its critics; Wallach [4] comments in his new book:

‘Not only is it increasingly difficult to assess the risks of emerging technologies; even having realistic conversations can be a challenge. In emerging fields of research, endless speculation and hype foster the illusion of inevitable progress on both laudable and anxiety-provoking fronts. Yet, even commonly repeated possibilities such as personalized medicine, designer babies, brain simulations and smarter-than-human computers and robots will be extremely difficult to fully realize. Filtering out the hype from the reality is itself a complicated task.’

Gender-specific medicine: questions resolved and new questions to be answered

Like all science, this discipline began and continues to develop as a reflection of the society in which we exist. Each new discovery reshapes our interpretation of what we have and are observing in our scientific inquiry about the differences between male and female. *Pari passu*, we ask new questions at every turn, refining our ideas about how biological sex affects the phenotype and how the impact of the environment integrates into who we ultimately are and become.

To present any meaningful comments on the development and future of what we call gender-specific medicine without some description of the remarkable achievements of this century is impossible. I have chosen to discuss three important topics that affect a contemporary concept of gender-specific science. The first is an answer to the question that pre-occupied us as we formulated this discipline almost three decades ago; what about us is due to biological sex and what to our experience of the environment in which we live? Should we continue to laboriously define whether we are talking about biological sex or gender (a composite defined not only by sex chromosomes but also by experience) in our description of the phenotype? Or is there a single entity united by the impact of hormones, age and environment on the genome for which we should be coining a new name—perhaps ‘phenotypic medicine’? The second topic concerns a consideration of the implications of our ability to profoundly alter the genome of living things, including the sex chromosomes, pointing out that it is time to stop and consider what might be the consequences of that power. If it were ever important to consider the impact of the sex chromosomes on the phenotype when planning biological investigation, this is the most important time to do so. Miller and Reckelhoff [5] repeatedly affirm the unfortunate and persistent lack of attention to sex as a vitally important variable in biomedical investigation [5–7]. Their comments are particularly relevant to the work being done on genomic editing.

Finally, I would like to consider the nature and stability of biological sex in the light of twenty-first century science. Are sexual beings simply and unalterably either male or female or part of a continuum in which sexual identity must be actively maintained and continues to be modified by hormones and environment during the course of a lifetime? If we have learned anything at all in our pursuit of the differences between men and women, we now know that sex is a complex entity and probably more plastic

than we had originally understood. Ultimately, sexual identity and behaviour, like everything else in biology, are the products of a myriad of factors, both internal and external, which modify gene expression.

How has our concept of gender-specific medicine evolved?

Looking back over the past 30 years’ history of gender-specific medicine, it is almost unbelievable, even laughable, that until the end of the twentieth century, scientists had never tried to define the nature and extent of the differences between the sexes. In light of our historical and continuing insistence on what we term ‘evidence-based medicine’, it is ironic that we assumed without any direct evidence that women were essentially identical with men except for their reproductive biology. It was a bikini view of women that was in stark contrast with our concerted efforts to define the complete picture of male anatomy, function and experience of disease. As is inevitably the case, however, the interests and needs of society itself prompted the beginning of the new concentration on a more balanced and complete picture of the human phenotype. Activist laywomen, transformed by the two world wars that exposed them to unprecedented educational and vocational opportunities, demanded direct attention to the unique aspects of their biology. The last two decades of the twentieth century saw the birth and development of a growing awareness that the physiology and the experience of the same diseases were significantly different for men and women to a degree that had never even been imagined. It quickly became obvious that what was true of males could not be assumed to be true of females. In fact, the cornucopia of observations that resulted from comparing any aspect of the two sexes’ physiology irrefutably confirmed that biomedical investigation not only had to sample both sexes, but also a thoughtful consideration of the resulting data would generate questions that would never have been thought of had just one sex been studied [8–10].

The focus and extent of gender-specific medicine has changed profoundly since the 1990’s, when the majority of both lay and scientific communities considered it to be the product of militant feminism and not an intellectual imperative. Its acceptance as a legitimate science suffered because the new impetus to include females in biomedical research at all levels was disregarded as a politically fuelled effort without any rational basis. At its onset, gender-specific medicine was considered at best to be a euphemism for women’s health, an issue that still continues to obfuscate the definition of the science, which is, of course, a comparison of men and women’s normal physiology and their unique experience of disease. In fact, critics still articulate objections to the forced inclusion of two sexes in scientific protocols. For example, in an atmosphere of reduced resources/support for biomedical investigation, scientists argue, why should an expansion of the study subjects be required? It is not surprising that although the fuelling force of public opinion propelled Congress to pass the 1993 National Institutes of Health (NIH) Revitalization Act [11] mandating the inclusion of women as subjects in clinical research, the imprimatur of the NIH on the relevance of sex-specific data at other levels of investigation has lagged a quarter of a century

behind. The recent notice from the NIH ‘expecting that sex as a biological variable will be factored into research . . . *in vertebrate animal and human studies*’ is being implemented 22 years after the NIH Revitalization Act of 1993 [12]. (Italics mine) Of interest is that a ‘Comment’ in Nature co-authored by Dr Janine Clayton and Dr Francis Collins published a year ago mentioned the importance of extending the sex-specific mandate to experiments on cells as well as ‘animals’ (whether vertebrate or otherwise was not specified) [13]. They pointed out that NIH policies requiring a balance of male and female cells and animals in pre-clinical studies would be rolled out in phases. Presumably, studies at the cellular level will be included in future NIH directives. Even now, lively if sometimes unarticulated push back to including two sexes in research protocols still exists; investigators are asking for clearer directives on when and how to include sex as a significant variable in their research protocols (<https://www.sciencenews.org/blog/scicurious/nih-policy-best-way-sex-equality-studies>). Brookshire (<https://www.sciencenews.org/blog/scicurious/nih-policy-best-way-sex-equality-studies>) comments:

‘No matter how the policy is interpreted, the message is clear: it’s time to include females in all studies. If this means that every NIH-funded scientist is required to become an expert on both the sexes, it means huge increases in time for experiments and the personnel and materials to carry them out. There are many questions still unanswered, from how big the policy change will really be to how NIH is prepared to pay for it.’ [8].

The implication of the NIH’s broader directive reflects the fact that gender-specific medicine was a concept originally defined by clinicians concerned with the care of patients. The principle that sex was a defining factor in the phenotype extended only gradually to include the consideration of the physiology of simpler life forms. Now the concept must extend to the protocols of molecular biologists concerned with genomic science.

The impact of the genomic era on gender-specific medicine

The historic announcement in 2000 that an international consortium had produced the first description of the human genome transformed the nature and focus of biomedical research [14]. Recent advances in genomic science are exploring ways to alter the genes of living systems instead of simply describing them and probing their function. Virtually as soon as the announcement that the human genome had been decoded, Craig Venter pointed out that if we could describe a chromosome, we could design one. He and others have done just that and proven that when inserted into single cell organisms, a synthetic genome can successfully capture the metabolism of the cell [15,16]. One of the latest remarkable manipulations of the genome is the insertion of DNA containing two novel nucleotides into *Escherichia coli*; the new additions are not recognized as defects and are replicated without affecting growth of the bacterium [17]. As the new discipline of synthetic biology expands, as it surely will, to the point that synthetic chromosomes will be inserted into living tissue of multicellular, ever more complex organisms including humans, the impact of biological sex on the function of that chromosome must

be considered and factored into investigative protocols. In the landmark paper published in 2006, Yang et al. [18] demonstrated the ubiquity of the impact of sex on gene expression: ‘Differential gene expression between sexes has been observed for a significant portion of the genes in the transcriptomes of various organisms, despite nearly identical genome sequences between males and females’. The differences involved thousands of genes, many of which operated to produce tissue-specific biological functions and/or pathways relevant to common diseases and showed tissue-specific chromosomal enrichment. Ober et al. [19] pointed out that in addition to the impact of the sex chromosomes, the autosomal genome, which was thought to be essentially similar in both sexes, in fact had an important role as sex-specific governors of genetic expression. These autosomal sex-biased genes may be expressed solely in one sex or at a higher level in one sex (sex-enriched expression) [20]. Wijchers et al. [7] established another layer of complexity in the impact of the sex chromosomes on gene expression, showing that it is dependent in many instances on sex chromosome dosage, irrespective of physiological sex.

We are just at the beginning of our efforts to modify existing life forms and create new ones. In their witty and trenchant comments on the promise and pitfalls of synthetic biology, Arnold and Meyerowitz [21] put it perfectly:

‘Nature’s writing is intricate (some say convoluted and opaque), but is effective. We are just learning to hold the pencil. Rational design is hard when one cannot even predict the effects of a single mutation in a single enzyme, or the full impact of adding a single gene to the thousands already present in an organism.’

The latest achievement in designing genomes utilizes CRISPR (clustered regularly interspaced palindromic repeat)–Cas9 technology to accurately, cheaply and rapidly make site-specific lesions in the genome of living organisms, destroying genes and/or introducing a novel, cloned gene into the disrupted DNA [22]. The recipient cell can be a pluripotential embryo-derived stem cell or a variety of somatic stem cells [23,24]. In a cogent and eloquent review of the potential of genomic engineering, Doudna and Charpentier [23], who are two of the most accomplished investigators in this field, describe how profoundly CRISPR–Cas 9 technology is revolutionizing our understanding of human biology and how we might change it. Even in this superb paper, there is no mention of any need for assessing whether or not the sex of the organism will make a significant difference in data from manipulating the genome now and in the future. Fausto- Sterling’s [25] remarks are particularly relevant as we take the first steps at the level of single molecules of making profoundly important changes in the genetic code:

‘Molecular biologists rarely think about interacting organs within an individual body, and even less often about how a body bounded by skin interacts with the world on the other side of the skin. Their vision of what makes an organism tick is decidedly bottom up, small to large, inside to outside.’

It was inevitable that researchers should use the new CRISPR–Cas9 methodology to cut DNA in human embryos and repair it

by introducing new DNA [26]. Although they used polyspermic zygotes harvested and discarded in IVF clinics, which fail to develop normally, Liang et al. [26] demonstrated recently that they could effectively cleave an endogenous gene in these zygotes. The experiments were not efficient; the edited embryos were not only mosaic, but there was also extensive off-target cleavage. Church however remarked that had the investigators used the latest CRISPR–Cas9 methodology, many of the negative outcomes of the work might have been avoided [27]. The paper excited considerable controversy, of course, but whatever the drawbacks of this first experiment, it makes it apparent that more precise editing of the human embryonic genome is within reach. It is also apparent that manipulation of the sex chromosomes themselves will be possible. The impact of such a manipulation, given what we now know about the impact of the sex chromosomes on gene expression, cannot be anticipated and may well have unimagined consequences. It will be important for scientists manipulating genomic structure to test the impact of the intervention in both sexes to compare the impact on the phenotype.

What defines the phenotype? Nature or nurture?

From the initial burst of enthusiasm that followed the 2000–2003 achievement of decoding the human genome, which predicted a profound revolution in our ability to optimize health and prevent disease, we are finding that, in fact, the situation is far more complex than we had imagined. The phenotype is fashioned by a myriad of factors (collectively termed the epigenome) that modulate/regulate gene expression. Our increasing awareness of the mechanisms of epigenomics makes it possible to address in a more informed and hopefully more accurate way some of the most important questions in mammalian biology. One of the most important is the old issue of what produces the phenotype: hard-wired, sex-specific genes and their associated hormones or the environment? It is now clear that a principle Jean Lamarck [28] iterated two centuries ago was an accurate and profoundly important insight:

‘The environment affects the shape and organization of animals, that is to say that when the environment becomes very different, it produces in course of time corresponding modifications in the shape and organization of animals.’

From the very beginning of our efforts to establish the concept of gender-specific medicine, furious debates exploded (most notably among the members of the first advisory committees to Dr Vivian Pinn at the Office of Women’s Health at the NIH) about the difference between ‘sex-specific’ and ‘gender-specific’ (personal observation). Chromosomal sex was viewed as a distinct, hard-wired set of attributes that irreversibly affected the human phenotype. The idea that the environment also made a substantial contribution to form and function was eloquently advanced, its advocates maintaining that biomedical investigators were largely ignoring its affect.

The early efforts to separate out the elements of the phenotype that were the result of each of these two forces seemed to be virtually impossible to achieve. However, the advent of ge-

netic biology and our expanding understanding of the whole phenomenon of epigenetics explain that genes are regulated (in addition to hormones and developmental age) by elements in our environment in ways that modify and refashion individual physiology from conception to the end of life. The key between ‘nature’ and ‘nurture’ is the epigenome. The fact that over 99% of the human genome is identical in all members of the human family seemed to be contradicted by the enormous variation in the human phenotype. The explosion of investigation into the nature and extent of epigenetics helps to reconcile the two observations.

Kolb and Whishaw [29] summarized the evidence establishing the plasticity of the brain and its ability to change both structure and function in response to the environment, citing Dudai’s [30] exposition of the impact of environmental and behavioural events on gene expression in the adult brain. Meaney and co-workers [31] showed that some of the changes in the brain are reversible. They investigated the impact of positive intensive attention to newborn offspring by rat mothers, showing that pups who enjoyed an optimal experience and those who experienced significantly less attention from their birth mothers had a different epigenomic profile in their glucocorticoid receptor gene promoter in the hippocampus. The epigenomic mechanism was plastic during the first week of life: when deprived pups were fostered to attentive mothers, their epigenomic pattern was indistinguishable from those who had been born to attentive mothers. Interestingly, the epigenomic pattern could be reversed by central infusion of a histone deacetylase inhibitor. These same investigators described the epigenomic regulation of the hippocampal glucocorticoid receptor profile in humans with a history of abuse in childhood, resulting in an increased risk for suicide [32]. Regrettably, all subjects were males.

Many epigenetic changes in the genome are heritable and persist for generations: not only the habits and attributes of parents, but also of grandparents have now been demonstrated to have consequences for grandchildren [33]. These changes are often sex-specific in both ancestors and their probands; Kaati et al. [34] have observed a sex-specific effect of parents and grandparents on the risk of offspring for cardiovascular disease and diabetes. The final human product is thus the result of our own environmental experiences and in many instances, those of our ancestors, whose phenotypic modifications achieved by epigenomic activity, are heritable.

Male or female? Two distinct categories or a continuum?

A topic of pressing importance to gender-specific medicine concerns the nature and stability of biological sex. In a community of scholars specifically interested in the differences between male and female, disorders of sex development (DSD) should be a topic of central and openly discussed interest. A first step might be to consider using the term ‘variations in sex development (VSD)’ as a step in correcting the notion that these are pathologic phenomena. The consequences of DSDs for the individual can be at best problematic and at worst catastrophic and even the most scholarly papers on the molecular biology of DSDs include thoughtful and sensitive commentary on the issues of sexual identity and conflicts about sexual behaviour that arise in these individuals.

The term DSD refers *sensu strictu* to ‘a congenital condition in which development of chromosomal, gonadal or anatomical sex is atypical’ [35]. Interestingly, the mechanisms, both chromosomal and hormonal, that underlie intersex phenotypes first attracted widespread attention at the Olympic games of 1936, when the Olympic committee challenged the sex of two female sprinters, each of whom had a strikingly male habitus [36]. Their efforts to decide definitively whether athletes with ambiguous secondary sex characteristics were ‘male’ or ‘female’ are actually a kind of mirror of the evolution of changes in our own concepts of the nature of sexual identity. The Olympic Committee’s efforts to settle the issue started simplistically with inspection of external genitalia, but by 1968 more sophisticated methods prevailed [37]: first, the identification of the Barr body as an unequivocal authentication of being female and subsequently by chromosomal testing. All failed because of the complexities of the sex-determining process: women with androgen insensitivity have a complete feminine phenotype in spite of an XY chromosomal pair; even persons with a 46XX chromosomal complement have testes but no Sry gene, implying that other gene(s) as yet unidentified but collectively named the ‘Z’ gene, can induce testicular formation [38].

It is indisputable that biological sex is not cleanly divided into male and female; the data document transitional forms in the overlap between the two groups. The spectrum is not simply black and white; it is more accurately represented by including various shades of grey, shades that include discordance between chromosomal sex and genital anatomy (intersex), an individual’s pervasive and persistent conviction that he or she is not the sex that anatomy and even chromosomal identity indicate (transgender/transsexual) and variations in the choice of sexual partners that, by the standards much of the contemporary society has traditionally espoused, are aberrant (homosexuality). Although the first efforts to decide what proportion of live births did not fit neatly into our classic binary classification calculated the figure of 2% for intersex individuals [39], even that figure is very probably underestimated for a variety of reasons (aversion to reporting anatomical or behavioural anomalies or the serendipitous discovery of chromosomal abnormalities that would otherwise never have been reported [40], for example).

The precise nature of the complex processes that underlie biological sex determination is becoming clearer. Vilain and colleagues [41] has just produced a comprehensive summary of the mechanisms involved in sex determination (the formation of a testis or an ovary) and sex differentiation (the genetic and hormonal factors involved in defining not only the genitalia but also sexually-specific tissues such as the brain). Their paper also includes a cogent exposition of the details of psychosexual development, with precise definitions of the differences between gender identity, gender role and sexual orientation. It is beyond the scope of the present review to enumerate all of the latest data generated about the establishment of sexual identity, but some salient points are worth mentioning. Although it is still asserted in some contemporary scientific literature that the female sex develops by default, (in the absence of the masculinizing Sry gene), it is clear that early ovarian organization is a unique and active process, the result of several previously identified regulatory ele-

ments [42]. Furthermore, as outlined by Ottolenghi et al. [43] the determination of gonadal sex involves ‘the sum of antagonistic interactions among a number of testis and ovarian factors’ that act to precisely determine gonadal sex. DiNapoli and Capel [44] reinforce these data, showing that shaping the gonad is the consequence of both male-promoting and female-promoting signals: ‘Sox 9 and Fgf9 push gonads towards testis differentiation. These two genes are opposed by Wnt4 and possibly RSP01, which push gonads to ovary differentiation.’ There is a kind of developmental war between the sexes even in the earliest stages of life, raising the possibility of the battle’s going wrong in any one of several steps. Even more interesting challenges to our concept of human sexuality is the observation that, in fact, sexual identity must be actively maintained throughout life by specific genes (the Foxl2 gene in the ovary and the DMrt1 in the testis); when these defining genes are inactivated, the cells lose their original identity as ovarian or testicular cells and revert to the opposite form.

Whereas the mechanisms of how genetic and hormonal factors collaborate to form the sexed individual are well understood, those that create sexual preferences and practices are not as clear. Just as the lay public fuelled the initial interest of scientists in the differences between the sexes, it is driving us now to take a closer, more accurate look at the nature of sexual identity and behaviour. Social acceptance of lesbian and gay people has far outstripped the interest of biomedicine in unravelling the mechanisms that underlie departures from what society has considered ‘normal’ sexual behaviour for males and females. Professional advisory and policy-setting groups are responding [45,46].

Swaab and Garcia-Falgueras [47] advance a provocative hypothesis on the genesis of trans-sexuality based on an observation originally set forth in a 2006 consensus statement on the management of intersex disorders. They point out the sexualization of the genitalia takes place during the first 2 months of pregnancy but that the sexual differentiation of the brain begins in the second half of the pregnancy. They suggest that the two processes may be independently regulated and a mismatch can underlie the phenomenon of trans-sexuality [48].

How do all these data affect those of us involved in the gender-specific care of patients? It certainly calls for an acknowledgment of the fact that the world is not cleanly divided into one of the two categories and that the frequent variations on sexual identity and behaviour have a biological basis and must be addressed in our consulting rooms and laboratories. A recent Institute of Medicine (IOM) report sets out the many layers of complexity to the categorization of humans who are the subjects of clinical investigation into ‘male’ and ‘female’. The report will have significant affect. It is a logical sequence to the IOM’s earlier report emphasizing the importance of the need for inclusion of males and females in clinical investigation, a fundamentally important first step in authenticating the importance of sex in molding phenotype [6].

Our earliest ideas of the nature of gender-specific medicine seem clumsy and woefully inadequate in the light of the huge amount of recent information about the elements involved in the development and maintenance of sexual identity. As is often the case, we have been forced by important societal developments

to re-examine our perceptions of what it means to be human and, particularly, what it means to be a male or a female. Hopefully, the biological data will prompt us as clinicians to examine and, wherever necessary, restructure our own emotional responses to the phenomena of variations in human sexual identity and behaviour. Furthermore, it is obvious that as our understanding of basic molecular mechanisms that underlie biological sex and behaviour improves, some will undoubtedly find it irresistible to use our increasingly powerful ability to edit the genome to alter the sexual phenotype.

CONCLUSION

The power and promise of the genomic era are without any precedent in human history. Gender-specific medicine is a concept that was originally generated by clinicians. The latest iteration of our role as scholars concerned with the impact of sex on the phenotype is to closely monitor the increasing sophistication of interventions that modify the genome. As their attention extends to more and more complicated life forms, genomic scientists must be persuaded to include sex as a crucially important variable in their experimental protocols. It is becoming clear that our ideas about how to maintain health, prevent disease and treat illness will be increasingly affected by research that expands our understanding of the genome and the myriad of factors that modify its expression. In the coming era, our current treatment of disease in many instances will rapidly become obsolete and be replaced by interventions at the molecular level, which will inevitably improve the human condition. Venter's remarks on the imminence of our doing that effectively are worth quoting:

'Our knowledge is so primitive of the human genome that to start engineering it is just stupid. Hopefully, in 50 or 100 years our knowledge will be sufficient that we could do that intelligently. In the long run genetic manipulation of humans is not only inevitable (but) probably a very good idea.' [49].

If Kurzweil [3] is correct, we will be able accomplish that in a much shorter period of time.

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