Whole-Genome Sequencing in Healthy People

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CME Activity

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Learning Objectives: On completion of this article, you should be able to (1) identify the currently accepted clinical applications of genomic testing as well as the challenges to interpretation of the data, that is, variants of uncertain clinical significance; (2) describe how whole-genome sequencing conducted in individuals with no current diagnostic issues compares and contrasts—both scientifically and ethically—with diagnostic testing; and (3) list several arguments both for and against whole-genome sequencing in healthy individuals and recognize that there are insufficient data to yet decide whether there is net benefit.

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Abstract

Recent technological advances have radically changed genetic testing from an expensive and burdensome undertaking to a rapid and less costly option for many purposes. The utility of "next-generation" sequencing has been found to establish the diagnosis for hundreds of genetic disorders, to assess pharmacogenomic variants, and to identify treatable targets within malignant neoplasms. The ready availability of genomic information has led to the question of whether there would be clinical benefit of sequencing the genome of individuals who are not seeking a diagnosis, that is, genomic screening in generally healthy people, to provide anticipatory insights for their health care. Little research has been conducted in this area. We examine the considerable unresolved scientific and ethical issues encountered when considering whole-genome sequencing of healthy people.

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Precision medicine is a term recently made mainstream by President Barack Obama's 2015 State of the Union address,[1] in which he announced a major new initiative by that name that proposes moving from a "one-size-fits-all" approach to more individualized health care. The Precision Medicine Initiative will develop a voluntary national research cohort of 1 million or more volunteers to propel our understanding of health and disease. The long-term goal of this project will be to collect multiple types

of data, including comprehensive assessment of genotypes, to deepen current understanding of how our genetic differences result in observable differences in health and disease, so that health care providers can provide recommendations based on more precise estimates of personal risks. An initiative of this scale is now feasible because of recent major technological advances in genetic sequencing and bioinformatics.

Precision medicine is not synonymous with whole-genome sequencing (WGS), but rather seeks application of genomic technologies as a key strategy in tailoring care to maximize benefit and minimize harm to individuals. At some point, sufficient experience will be accrued to know how to "right-size" genomic testing to achieve this lofty goal for the population as a whole. Gaining experience from pan-genomic testing in the near term, in the form of WGS, may identify that subset of genomic analysis that is most rational and cost-effective for individualized care, but empirical data are needed to establish these boundaries. As evident from a description of the challenges to follow, WGS as a health-promoting strategy is a new journey in progress. This article will focus narrowly on WGS in healthy individuals as 1 potential branch of this journey.

THE HUMAN GENOME PROJECT

The Human Genome Project (HGP), the world's largest collaborative biological project, was launched in 1990 and was officially completed in 2003. The National Institutes of Health–funded sequencing of just a single human genome cost US$3 billion and involved 20 institutions in the United States, United Kingdom, Japan, France, Germany, and China.[2] Simultaneously, Celera Genomics Corporation, a private company lead by Dr Craig Venter, began human genome sequencing in 1998 and completed their project at the same time at one-tenth the cost of the public project.[3] In 2000, President Bill Clinton and UK Prime Minister Tony Blair jointly announced the completion of a rough draft of the human genome: "Today we are learning the language in which God created life" and expressed hopes that we "accept the responsibility to make these advances work for all our people in all our countries for the common good of all humankind."[4,5] Both Dr Francis Collins, Director of the HGP, and Dr Venter were on stage for the announcement of this monumental task, illustrating visually the public and private contributions that have pushed the frontiers on genomics.

The cost of sequencing those first human genomes using variations of the 1970s technology of Sanger sequencing (sometimes called first-generation sequencing) was not merely staggering but prohibitive for any large-scale use. Given the limitations and competition for health care dollars, introduction of new technologies must always consider costs vs value-added. Fortunately, an answer to the extraordinary costs of early genome sequencing was in the wings via a new technology—next-generation sequencing (also known as next-gen or massively parallel sequencing). *Next-gen sequencing* refers to several innovations involving DNA template preparation, sequencing of hundreds of millions to billions of individual DNA templates (hence, "massively parallel"), image capture, followed by sequence alignments and assembly and variant detection (Figure 1).[6] By 2013, the cost of WGS was estimated by the Human Research Genome Institute to be about US$5000 per genome.[7] Compared to the original HGP, the director of the Clinical Genome Service at Stanford University (USA) illustrated the plummeting of costs as the equivalent of a US$400,000 Ferrari now selling for 40 cents.[8] Today, WGS is available for as little as US$999 per genome (https://www.veritasgenetics.com/), which would be 8 cents on that US$400,000 car. Thus, issues of cost have been addressed in dramatic fashion but demonstrations of value-added for health care remain unproven.

Still in its infancy, the ability to sequence the entire genome has opened a new world in the quest to understand human disease and normal variability in the population. Although next-gen sequencing is becoming readily available and quite routine, we have barely scratched the surface of our understanding of the human genome. Most of the genome remains a mystery, and the ability to interpret the effect and importance of most genetic variants remains extremely limited.

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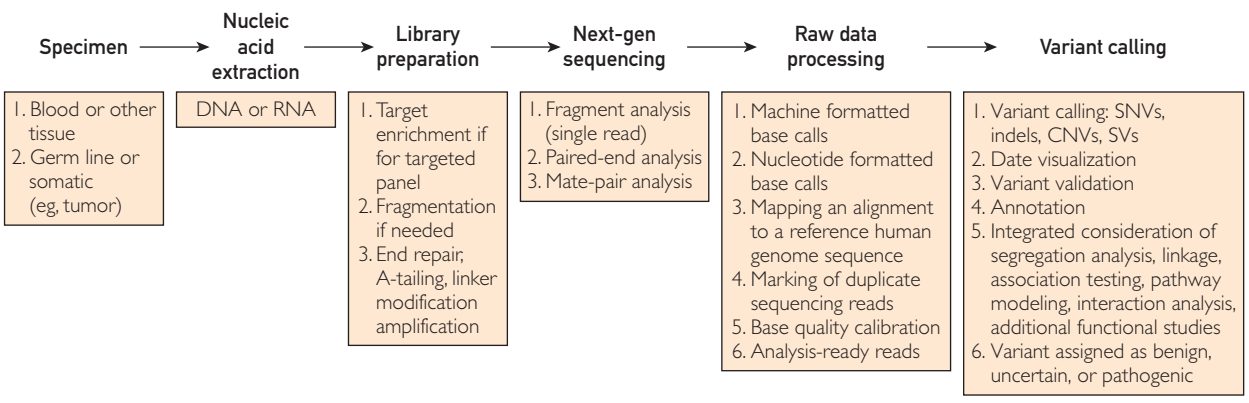

FIGURE 1. Overview of the next-generation DNA sequencing workflow. CNV = copy number variant; Indels = insertions and deletions; SNV = single nucleotide variant; SV = structural variant. Adapted from *Gut*,[6] with permission.

COMMON VARIANTS AS PREDICTORS OF COMMON DISEASES

The first widespread use of a comprehensive genetic assessment was via genome-wide association studies (GWAS) using platforms that looked at single nucleotides that were known to vary across the population (single-nucleotide polymorphisms [SNPs]). The hypothesis behind vigorous pursuit of GWAS was that most common diseases such as hypertension or type 2 diabetes were caused by the combined effects of multiple common SNPs across the genome. That is, alleles from certain SNPs would be found to occur at a higher or lower frequency in cases than in the matched controls. It was anticipated that by analyzing large numbers of carefully phenotyped ethnically matched cases and controls, one could identify SNPs that could predict disease risk. As of March 2016, the GWAS Catalog team (https://www.ebi.ac.uk/gwas/home) listed 2414 studies of 16,696 unique SNP-trait associations involving 1361 unique traits. Of these, results for 669 SNPs have been replicated. This area of research has been remarkable for the volume of replicated reports rapidly emerging and has provided new insights into biological pathways not previously suspected of being etiologic in many conditions. The magnitude of the effect of any 1 SNP, however, as expressed by odds ratios for most individual markers, is minor, typically in the range of 1.05 to 1.2 and rarely above 1.5. From the clinical perspective, therefore, the clinical utility of risk prediction based on these variants is unclear. Having a trivial or minor deviation of predicted risk for a common disease is unlikely to have a substantial effect on medical management. However, as more SNP associations are recognized, there is interest in exploring the potential value of an empirically derived cumulative risk score. For some disorders, more than 100 SNP risk loci have been reported, needing further study to understand how to integrate risks to achieve a per person risk score. Vast amounts of data from individuals enrolled in an unbiased manner and availability of thorough clinical phenotyping information will be required to derive meaningful risk scores for any specific condition. The President's Precision Medicine Initiative will give a boost to this science, but even genotyping a million people will not be adequate for all purposes. Given the potential number of combinations of SNPs any individual may have, it is impractical to have a molecularly matched cohort across multiple markers. Therefore, simpler empirical approaches using big/huge data will be necessary.

More important, the clinical utility of a genetic risk score depends on 2 use cases, neither of which is supported by the current data. The first is that genetic risk scores will either outperform or markedly improve other risk predictors. In fact, efforts to develop genetic risk scores for type 2 diabetes and coronary heart disease suggest that genetic risk contributes at best only modestly to risk prediction.[9-16] The likely explanation for this

poor performance is that these and most common disease risks are highly influenced by a range of social factors, including diet, level of physical activity, and level of social disadvantage. The potential value of this approach for identifying clinically meaningful differences in risk is therefore uncertain, although further assessment of genetic contributors to risk may yield important insights into disease pathogenesis. The ultimate test of the use of a genetic risk score for clinical prediction, currently lacking, would be a reported role of the risk information in improving health outcomes.[10,13] The second use case is the hypothesis that knowledge of increased genetic risk will lead to increased adherence to healthy lifestyle measures. Despite hopes that self-knowledge of potentially increased risks might serve as motivators for making constructive lifestyle changes, the evidence for this is lacking. A recent meta-analysis[17] of 18 studies concluded that "expectations that communicating DNA-based risk estimates changes behaviour is not supported by existing evidence." In contrast, the meta-analysis noted the absence of adverse effects such as depression and anxiety, which have also been of concern related to divulging personal risk predictions. It seems clear as well that some, perhaps many, people find their genetic results intriguing.

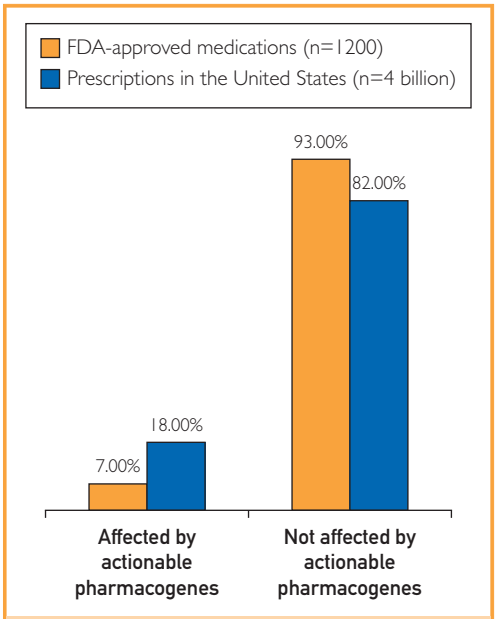

FIGURE 2. Clinically relevant pharmacogenomic variants are not uncommon in the general population. FDA = Food and Drug Administration. Data from *Nature*,[19] with permission.

PHARMACOGENOMICS

Individual differences in response to pharmacological agents have long been recognized, and studies found that some of the variability is due to genetic factors affecting pharmacokinetics (altered drug metabolism), pharmacodynamics (altered binding to receptors affecting therapeutic availability), idiosyncratic reactions (immune-mediated hypersensitivity reactions), and effect on disease response to therapies (having or lacking targets for specific therapies).[18] The enzymes performing these functions are highly polymorphic, often with marked differences across ethnic groups. These genetic differences that diminish or enhance the function of drugs can be assessed in a focused manner, but are also included in WGS. Xenobiotic metabolism, controlled by several hundred enzymes, evolved to handle substances foreign to the body, such as chemicals in different plants that were ingested. Differences in these enzymes are not considered as pathogenic or nonpathogenic, but rather are described by their effect on drug activity. The cytochrome P450 family of 58 human CYP genes makes lipophilic molecules more water soluble (phase I metabolism), and additional phase II enzyme reactions complete the processing to form excretable nontoxic substances. Clinically relevant genetic differences in non-CYP enzymes are also described in growing numbers and are common in the general population (Figure 2).[19] "When 12 pharmacogenes with at least one known, actionable, inherited variant are considered, over 97% of the US population has at least one high-risk diplotype."[20] Approximately 7% of 1200 Food and Drug Administration–approved medications are affected by actionable inherited pharmacogenes, and approximately 18% of US outpatient prescriptions (n=4 billion) are affected by actionable germ-line pharmacogenomics, which exhibits that several pharmacogenetically high-risk drugs are commonly prescribed.[19] These facts have been cited to argue for preemptive genotyping for pharmacogenomic variants, and there is growing availability of expert guidelines for modifying prescribing practices based on

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drug-gene pairs. However, implementation barriers remain substantial and unresolved, including absence of infrastructure in the fragmented health care system that exists in the United States to cope with genetic data in medical records and to provide clinical decision support at point of care; clinician resistance to change and lack of pharmacogenomic education; and issues of costs and reimbursement, which are compounded by the absence of empirical evidence that this sort of genetic testing is, in the long run, cost-effective.

RARE HIGH-PENETRANCE VARIANTS

Moving to the other end of the risk spectrum, if genomics is not yet capable of clinical effect on health via common SNPs for common disorders, might it be a more salient strategy to find those individuals in the population who carry rare but highly actionable genetic variants for which knowledge of this could more likely lead to improved health interventions? Testing for high effect but rare disorders would be analogous to universal newborn screening, in which infants are screened by all 50 states for approximately 30 individually rare disorders because the technology costs were low enough and the benefits to the individual and to society were sufficiently great to justify this undertaking.

Clinically, next-gen sequencing for rare variants was rapidly harnessed for 2 distinct purposes. The first was for making molecular diagnoses, whereas the second was for defining the underlying genetic changes in cancer. The initial introduction of clinical next-gen sequencing was targeted to panel testing for specific disease entities (Figure 3). This quantum leap in diagnostic capacity permitted the development of disease-focused multigene panels that allowed an individual to be tested for all well-established genes related to their phenotype simultaneously for less cost than just 1 gene might have cost using "first-generation" sequencing methods. The targeted panels often included genes merely suspected of being related to the phenotype. Through this process, many people received expected diagnoses, unanticipated diagnoses, and news of having novel DNA variants that cannot presently be interpreted. As the next-gen sequencing technology matured and the cost continued to decrease,

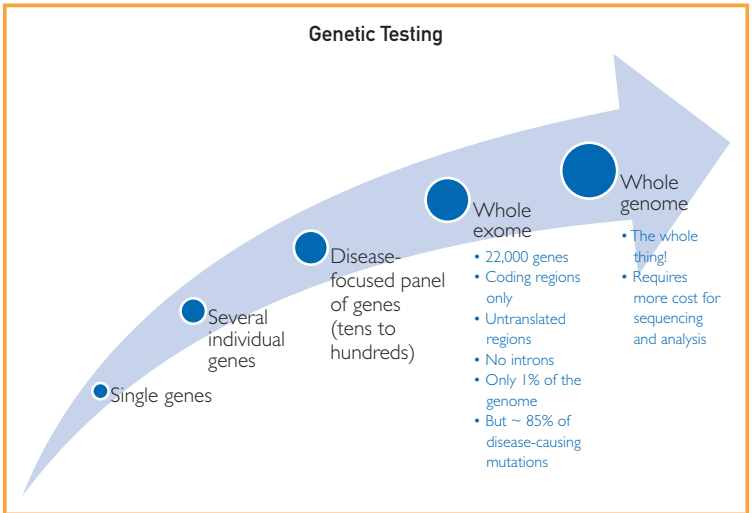

FIGURE 3. Genomic sequencing trajectory from the 1990s to the present.

whole-exome sequencing (WES; sequencing limited to the gene-coding portion of the genome, the exome) emerged as a viable option. For individuals or families with suspected genetic disorders not explained by panel testing, WES entered clinical practice. The diagnostic yield of recent WES has been reviewed and varies with the amount of prior testing and type of phenotype being studied.[21] In patients with suspected rare genetic disorders, an astounding ~20% to 40% of previous diagnostic dilemma cases received a molecular diagnosis even though the new technology was known to have gaps in coverage of certain areas of the genome and for certain types of molecular lesions. Dozens of new clinical syndromes with molecular diagnoses have been described since clinical WES began. Whole-genome sequencing may increase this diagnostic yield even further.[22-25]

One of the surprising lessons emerging from both research and diagnostic next-gen sequencing has been the high prevalence of de novo sequence variations in all of us. It has been estimated that 50 to 100 de novo mutations occur in each healthy individual. These are mutations not carried by a parent but those that occurred around the time of conception of this new individual. Fortunately, most do not affect coding regions or critical genes in the exome.[26]

Although not covered further in this article, another fruitful application of next-gen

sequencing involved sequencing of genomes of cancers in search of therapeutic targets. This area of somatic genetics (as opposed to germline genetics) continues to rock clinical practice, and new drugs are being rapidly developed to align with the newly discovered targets in cancers.[27-30]

WHOLE-GENOME TESTING IN HEALTHY PEOPLE

With the cost of WGS now approaching that of WES, this technology seems to be overtaking the latter, and researchers and industry have been considering the application of WGS for predictive medicine in the healthy population in general. It is notable that the exome (coding portion of the genome) comprises only 1% of the genome but contains essentially all genes. However, driven by increasingly powerful bioinformatics and sequencing technologies, laboratories have become ever more capable of detecting human genetic diversity over the entire genome. This includes the ability to detect all the common variants (those studied by GWAS, most of which are not in the exome), rare single-nucleotide variants, insertions and deletions, copy number variants, and some structural variants. Whole-genome sequencing has been called the "ultimate genetic test" because of its comprehensiveness and its timeless nature; that is, your genome at conception is the same throughout your life.[31] Whole-genome sequencing, a genetic selfie, is available to consumers who are willing to pay for this, and paraphrasing a business report it was suggested that consumers want their genomes sequenced and they have a right to have that—price is the only question and when that comes down far enough, it will just happen: everyone will get their genomic data—we will be living in a kind of "genetic utopia."[32] Research on attitudes reports a high level of interest and openness in the populations studied in knowing about one's genetic makeup and a feeling of an emerging fundamental right to know what is found in one's genome, even when the interpretation is unclear. Are those being surveyed representative of the general healthy population or do they represent early adopters or those with a specific genetic concern prompting such a search? Is the science ready for this next step in a general population? Is the population ready for this next step? Should WGS be available outside a health care setting? Will WGS in healthy people in fact lead to better health care? What knowledge must we extract from the National Institutes of Health Precision Medicine Initiative and other efforts to determine whether value is added with WGS as a predictive tool for the general population? Further consideration of the state of science and the ethical issues are provided.

SCIENTIFIC CONSIDERATIONS OF WGS

What is likely to be found today in conducting WGS in a "typical" individual? Although humans are said to be 99.9% genetically identical, the enormity of the human genome means that differences still comprise a long list. The human genome has 6 billion base pairs: 3 billion from a person's mother and 3 billion from a person's father, arranged on 23 pairs of chromosomes. When working with a data set this large, accuracy matters greatly. If there is only 1 error in base calling every 10,000 bases, this will result in 600,000 erroneous calls for each person being analyzed. Publications suggest that a per person list of DNA differences from a reference human genome sequence will include 3 to 4 million SNPs (~20,000 per exome) and 400,000 to 500,000 insertions and deletions per genome.[33] In 2010, around 20% of the SNPs and approximately one-third of the insertions and deletions were reported to be novel, that is, not appearing in public databases or otherwise known to exist previously. Most of these variants occur in areas of the genome not composed of known genes or occurred in genes of unknown function. Of the approximately 20,000 genes thought to reside in the human genome, only approximately 4000 have a known clinical phenotype associated with that gene. The same authors filtered one of the studied genomes for known variants and reported that only 14 could be interpreted as having possible known medical implications. Of these 14 variants, 10 were monoallelic variants for recessive disorders (individuals were carriers, not at personal high risk of disease). In a similar early study[34] of WGS in 12 healthy scientists, a mean of 5 (range, 2-6) personal disease-risk findings per person was reported.

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Thus, most of what is found in WGS is either in a noncoding region (99% of the genome as we know it today), a known neutral variant in a gene, a variant of unknown significance (VUS) in a gene of known significance, or a variant in a *gene of unknown significance*. Presently, the group of variants classified as VUS is the most problematic for reporting laboratories. Their sheer number is daunting, but no longer surprising, in light of the number of variations reported from any reference genome. There have been recommendations by the American College of Medical Genetics and Genomics (ACMGG) to use specific standard terminology—"pathogenic," "likely pathogenic," "uncertain significance," "likely benign," and "benign"—to describe variants identified in genes that cause Mendelian-type disorders.[35]

These ACMGG categories for Mendelian- or single gene–type disorders are not applicable to the low penetrance GWAS-identified SNPs, for which odds ratios are usually cited to describe associations with specific phenotypes. There remain significant computational and empirical barriers to clinical use of the GWAS markers. Translation into clinical practice has been thwarted by lack of empirical information on how to combine multiple SNPs to provide a legitimate posterior genetic risk score. Vastly more research is needed to know what to make of an individual's collection of SNPs associated with a particular disease.

The VUS Dilemma

Contrary to popular belief, bioinformatic machine processing of variants is incapable of adequately curating and classifying a large number of single-nucleotide variants for clinical use using all available computational and data management techniques. Manual curation is still necessary and time-consuming. Granular information is required to classify any given variant to determine whether that variant is pathogenic or not, for example, family-based studies, functional studies, and disease association studies. Unfortunately, there is little to no useful data for extremely rare or novel variants. In the absence of biological information on a variant, various computational methods to predict pathogenicity have been developed, but these are less than ideal for clinical purposes. There remains substantial discordance across laboratories using the same data on the same variant. An element of subjectivity persists, and there is no biological gold standard to determine which laboratory is right. A recent study[34] reported that "curation of 90 to 127 genetic variants in each participant required a median of 54 minutes (range, 5-223 minutes) per genetic variant, resulted in moderate classification agreement between professionals (Gross κ, 0.52; 95% confidence interval, 0.40-0.64), and reclassified 69% of genetic variants cataloged as disease causing in mutation databases to variants of uncertain or lesser significance." To understand the potential biological significance of any 1 variant requires amassing of sufficient evidence of multiple types.[36] This level of evidence is simply not available for most genes. The effect of this interpretation bottleneck leads to a memorably titled commentary that has been echoed multiple times by those wrestling with this subject: "The $1,000 genome, the $100,000 analysis."[37]

Aggressive efforts by researchers and funding agencies to promote data sharing across laboratories and internationally will, over time, result in "big data" that will eventually enhance the ability to reclassify some of the more common VUSs, though it may take many years to collect sufficient data to have a large effect. A huge number of VUSs are so extremely rare that there is no realistic hope of amassing sufficient epidemiological data to reclassify in the foreseeable future. Using population-based approaches, Shirts et al[38] calculated that it would take hundreds to millions of cases to classify rare variants for breast cancer (Table 1), with larger samples required for rarer and less penetrant variants. This table does support the concept behind the million-person Precision Medicine Initiative, as it will provide necessary controls for the many disease-focused efforts underway around the world. For the rarest variants, Shirts et al also reported the importance of pursuing family-based strategies that require much smaller numbers and may be a preferred and more realistic method for resolving the significance for some variants. Functional assays also hold much promise in being able to distinguish harmful from harmless variants and have been developed for some genes,

TABLE 1. Subjects Necessary to Characterize a Breast Cancer Variant as Pathogenic[a,b,c,d]

Relative risk	Tumor type	MAF=0.001	MAF=0.0001	MAF=0.00001
12	Breast	663	6544	65,358
6	Breast	1652	16,392	163,792
3	Breast	5491	54,650	546,238
1.5	Breast	49,162	490,135	4,899,864

[a]MAF = minor allele frequency.
[b]Data indicate that especially for variants associated with low risk, the number of individuals required to assess pathogenicity is high and increases as the frequency of the variant diminishes.
[c]Subjects are composed of 50% cancer cases and 50% cancer-free controls.
[d]Pathogenicity required 80% power and tolerated a 10% false-positive rate.
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but for most, this is not yet an option. Note that many genes have multiple biological functions and individual variants may affect only a subset of these activities. Thus, functional assays also come with many caveats. Thus, even if one could devote an unlimited amount of resources to an analysis, no amount of money can purchase validated interpretation of most genetic variants today.

The facts as understood today are that very few genes are fully deterministic of one's chance of disease (exceptions exist such as hemoglobinopathies). Emerging data indicate that the genetic contribution to most traits and common diseases is based on an aggregate effect of variants from many genes (often dozens or more), with most genetic variants having a small effect and the net contribution of genetic risk is often modest compared to the effect of strong environmental modifiers. For example, the risks of lung cancer or coronary artery disease do have measurable genetic associations, but the effects of most genetic variants are mostly dwarfed by risk factors such as smoking and aging.

Rare, Important Variants

Rather than focusing on what cannot be interpreted, an alternate approach pursued by some groups is to generate a set of well-defined and carefully vetted high effect genes and view the efficacy of genomic testing through a lens of the rate of discovery of defined pathogenic variants in well-defined high-effect genes. The term *actionable* has been used to describe genes for which some medically accepted treatment or prevention can be offered, though it must be noted that there is no agreed-upon standard of which genes belong on that list (more below on this subject). Examples of such genes include the hereditary breast-ovarian cancer gene, *BRCA1*, and a gene that causes Marfan syndrome, *FBN1*. In a University of Washington study,[39] variants in 112 "actionable" genes that might be undiagnosed in adulthood (ie, people may not already know they carried some specific genetic predisposition) were studied in 4300 European and 2203 African ancestry participants. Two percent of European and 1.1% of African ancestry individuals have pathogenic or likely pathogenic variants in highly actionable genes. Likewise, using a smaller partially overlapping list of 56 genes defined as actionable by the ACMGG, it is estimated that 2% of the population will have a pathogenic variant in one of these important genes. Are these numbers compelling enough to embrace WGS at the population level and in medical practice? For the individuals found to be carriers of variants in these high-effect genes, one can argue that these findings could be important, like the newborn found to have phenylketonuria. For those found to be carriers for autosomal recessive disorders, knowledge provides the opportunity to determine whether their partner is also a carrier, thus understanding risks to future children.

Penetrance: Beware

Part of considering the value proposition of WGS is understanding the risks and benefits derived from discovery of a reported pathogenic gene mutation. The level of risk determines the clinical management of those carrying a particular variant. What is known about the magnitude of genetic risk associated with variants defined as pathogenic? For nearly all genes causing Mendelian-type disorders (single-gene disorders), the penetrance for disease (the percentage of people with a pathogenic variant in that gene who get the disease) has been estimated on the basis of individuals with the disease phenotype, often based on families in which the disease had already been manifest. The ascertainment bias inherent in studying families that surface because of manifesting a phenotype leads to marked overestimation of the risk associated with that gene. Empirical corrections of risk

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estimations have not been possible because of the rarity of most genetic disorders and the barriers to sequencing huge numbers of randomly ascertained members of the general population. An illustrative cautionary tale was recently published.[40] In a prospective cohort study of more than 2000 individuals from 7 US academic medical centers recruited for non—antiarrhythmic drug exposure, variants in 2 arrhythmia disease genes (*SCN5A* and *KCNH2*: causes of Brugada syndrome and long QT syndrome, both designated as actionable genes by the ACMGG) were assessed and scored for pathogenicity by 3 laboratories with ion channel expertise and compared to the ClinVar database (http://www.ncbi.nlm.nih.gov/clinvar/). Phenotypes were evaluated by electronic medical record (EMR) review. Forty-two variants in 63 participants designated as potentially pathogenic were called by at least 1 laboratory or ClinVar. An *International Classification of Diseases, Ninth Revision* code for arrhythmia was found in 17% of carriers and also in 13% of those without variants (P=.35 for difference). Corrected QT intervals were not different between variant carriers and those without. After manual review, 35% of those with pathogenic DNA variants had any electrocardiographic or arrhythmia phenotype and 2 people had corrected QT intervals longer than 500 milliseconds. These are the types of variants that lead to interventions such as pacemakers and defibrillators, yet the conclusion to this study states, "In an unselected population, the putatively pathogenic genetic variants were not associated with an abnormal phenotype. These findings raise questions about the implications of notifying patients of incidental genetic findings."

Others have noted the same phenomenon in the cancer world, in which the penetrance of a putatively pathogenic DNA variant is lower in the absence of a family history for that phenotype. This may be counterintuitive; however, it implies that the family phenotype is not driven solely or primarily by the gene in question but risk is polygenic or multifactorial in nature. The risk estimates generated by studying multicase families reflect the underlying polygenic and multifactorial risk associated with the disease multiplied by the risk related to the gene variant being studied. Genetic risk estimates obtained by studying affected families cannot be extrapolated to the general population. They can only be extrapolated to populations similar to those from which they were determined.[41] This is directly relevant to the question of readiness to interpret genomes in a healthy person.

Nongenetic Factors

Not all (or even most) diseases will be predictable on the basis of the inherited genetic code, no matter how much knowledge is amassed. We know that infectious agents, epigenetics, microbiomes, and random events of nature can all highjack cellular activities. Environmental exposures and aging are crucial in understanding disease risk, especially for the most common diseases. Societal determinants such as education, occupation, and access to health care also affect health and disease outcomes in a most nongenetic manner. At present, more empirical data are needed to understand how disease penetrance of each "actionable gene" is modified by the context (eg, family history and other variables) in which the reported gene penetrance was determined.

ETHICAL AND LEGAL CONSIDERATIONS OF WGS

The ethical and legal considerations raised by WGS in healthy people are not unique, but the scale of the potential screening process raises important unresolved questions. Specific to this discussion, the use of WGS to screen for disease in the absence of any signs or symptoms raises questions about what should and should not be made accessible. Just because the technology exists to sequence the genome, should this be done? How is this similar to or different from consideration of whole-body magnetic resonance imaging scans? There is unresolved tension between making genomic testing available directly to those who are curious and desire information—no matter how limited or flawed the interpretation is—vs being able to justify genomic testing within a traditional health care system that weighs risks, benefits, and costs against other options for health care spending.

Policy development, expert guidance, and regulatory standards for WGS are lagging. Broad consensus has not been achieved across experts and industry and the general

population about defining 2 key end points in genomic analysis done within a health care system or in some other setting: (1) What genes should be included in genetic testing reports? and (2) What threshold should be set for being considered "returnable"? These decisions will hinge on conclusions about the intent to disclose only well-validated information vs the alternative option that is to provide as much as possible (well-validated plus a range of poorly understood results). There is much work still needed to develop practical, ethical, useful policies on all these matters and to establish how testing within a health care setting might differ from testing conducted in other settings, noting that these distinctions may be blurred by individuals tested outside the health care system who then present their results to health care providers for advice or follow-up. Even the decisions about who should be at the table and who has the authority to determine these issues are unresolved. What skill sets are needed and who can speak for stake holders? With next-gen sequencing pulled into clinical testing so quickly, research quantifying risks and benefits of varied approaches had not kept pace and is only recently starting to be backfilled with some empirical data. For example, members of the Clinical Sequencing Exploratory Research Consortium, a group of 9 studies investigating clinical genomic sequencing, have made varied decisions about which genes to interrogate for actionable findings when sequencing is offered. The results of those decisions are being assessed. Much more work of this type is needed to define best practices.

Clinical informed consent for genetic testing of single genes was developed and widely used since the 1990s. Those principles are still applicable, but a new challenge presented by WGS in healthy people is attempting to provide anticipatory counseling in the face of the untargeted nature of the potential findings. In WGS pursued for disease diagnosis, genes related to the phenotype are said to be on target whereas those unrelated are said to be incidental. Every WGS finding could be said to be incidental as testing was not driven by seeking a diagnosis for a manifesting phenotype. Alternatively, one could say nothing is incidental as one knows the entire universe of possibilities could theoretically be reported. (Presently, most laboratories offering genomic testing limit their return of results at least to genes about which something clinical is known, though some research projects have provided participants all raw data). There is an intrinsic high level of uncertainty about the test outcomes in WGS, unlike a traditional disease-focused testing, in which a person might be counseled for the possibility of a pathogenic variant, a VUS, or no findings at all, while focused solely on a specific gene or small group of genes. In WGS, most people will have genetic variants found, but the types of conditions involved are not known during the pretest counseling. In addition, the volume of information one might provide before testing, if trying to be anywhere close to comprehensive, would overload most individuals. How can one counsel meaningfully for all possible contingencies before such a test? Even among the most health literate, the ability to comprehend the scope of the test, and its limitations, is questionable. How does one judge what sort of informed consent is sufficiently informed?[42]

There have been vigorous conversations about the importance of individual choice in receiving genomic results, both clinically and in the research setting. Although most experts agree that facilitating personal autonomy is desirable, the ability to categorize genomic conditions and results into standardized granular categories for purposes of exercising this autonomy is elusive. Different methods for binning may involve considering actionability, severity, age at onset, organ and disease type, or inheritance pattern (dominant vs recessive) of the condition. Experts do not agree with experts and consumers do not agree with consumers on which conditions fit best into which bin or even how those bins should be defined. The amount of data available on views and preferences is still limited.[43-46] A recently proposed scheme involved scoring each disorder for severity, likelihood of disease, efficacy of intervention, burden of intervention, and knowledge base and then using an aggregate score to bin conditions for these elements of actionability.[47] Much additional research is needed on whether this approach is reproducible, offers individuals the right choices and an acceptable level of autonomy, and is ultimately clinically beneficial.

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TABLE 2. Considerations of Whole-Genome Sequencing in the General Population

Pros	Cons
Exercise of personal autonomy to explore one's genome—not justified for health care	Lack of evidence of net benefit across the population
2% will have a variant detected in an actionable high-effect gene that could lead to effective prevention or treatment of personal health risks Reassurance of those 98% with no findings	98% will not have a variant in the actionable high-effect gene found (costly) Failure to explain many health events in self and family
VUS may be found related to a known familial phenotype, leading to a new diagnosis in the family	Many VUSs will be found, which may incur unnecessary medical follow-up and expenses
Learn of carrier status for recessive disorders that may be used to address risks of having affected children	Difficulties in interpretation of DNA variants can complicate efforts to use for reproductive decisions Information may not be desired
Might offer personal satisfaction of facilitating knowledge on genomics if data are contributed to public databases	Potential discovery of data by forensics (eg, relative searches to find the rapist using DNA at the crime scene) or loss of privacy to hackers
Knowing of risk alleles could promote healthy behavior changes for some individuals	Current evidence suggests lack of behavior changes on the basis of knowledge of risk alleles across multiple studies
Studies to date indicate adequate ability to resolve psychosocial issues with pretest and posttest among people who choose to be tested	Inability to obtain true informed consent because of complexity and health literacy limits
Broader testing can contribute to eventual big data that will provide more precise information to future generations	Lack of high concordance on interpretation of variants in known genes even by using standardized rules for interpretation
Opportunity to investigate and correlate empirical family history with variants discovered, leading to more precise penetrance estimates	Discovery of predicted damaging variants that would never have been penetrant for disease outside context of classical family history
Multiple family members may benefit by learning about predispositions to specific health issues	Lack of consensus on how to offer choices on which parts of the genome people are interested in learning May generate intrafamily stress
Most studies indicate lack of significant psychological adverse effects related to genetic testing	Long-term psychosocial effects of predictive genomics are unknown
Most test results are highly reproducible	Lack of standards for quality control
Promotes development of clinical decision support in EMR (eg, EmERGE project)	Obligation for record keeping of important information—not yet well integrated in most EMRs and with clinical decision support
Early adopters/self-payers are underwriting some of the costs for later integration into the traditional health care system	Raises issues of distributive justice for those with less economic means if conducted outside standard medical practice for those who can pay
GINA protects against health care insurance discrimination for most circumstances[51]	Potential for insurance discrimination laws do not protect re life, disability, long-term care, and some exclusions for health care in the 2009 GINA
Several groups have developed short lists of genes they have been carefully vetted and considered actionable	No agreed-upon standard list of genes that are considered actionable or how to think about those genes in individual context (eg, Is *BRCA1* actionable in a 92-year-old man?)
Tests are inexpensive enough that long-term data storage may not be indicated, as complete reanalysis is more effective in the longer term	Storage of data is expensive and security risks not yet worked out
No reason to let perfect get in the way of good enough	But have we reached "good enough"? Unresolved issues of reinterpretation over the years: who does it, who pays, who is responsible?

EMR = electronic medical record; VUS = variant of unknown significance.

In the context of elective WGS, perhaps the people stepping forward for this new test have already resolved that they want to "know it all." This simple approach, while appealing on the surface, is not realistic. Individuals need to be advised of the limits of current testing interpretation (we cannot know it all now and not all conditions are purely due to genomics). It is worth noting that if health care dollars are paying for the test (ie, shared resources in a financially constrained system), providing information just because the patient wants it is not medically justifiable. Some aspects of WGS that are of interest to people have nothing to do with health (eg, ancestry and minor traits such as eye color). It would still seem optimal to be capable of offering individual choices, though

in a thoughtful manner on the nature of conditions that people are interested in knowing, and efforts are underway to continue to develop these options.

Unresolved ethical and legal issues surrounding genomic testing in general will apply to WGS in healthy people. These include addressing the potential for new gene discovery or interpretation: today's WGS report is outdated tomorrow. Who bears the cost of updated reporting and is there an implied obligation for periodic updating of genomic reports? How long must a laboratory store the data? Given the enormous size of the genome results, data storage is a major logistical challenge. Yet, to realize the full vision of WGS results as informing a lifetime of medical care, this issue must be addressed. If data are stored, this creates the possibility of identification risks and privacy breaches whose ramifications remain unclear. Presently, most EMRs are incapable of absorbing WGS data, so genomic data is held outside the EMR. Because of the logistics involved with vast amounts of data storage, resequencing periodically has been suggested as a better and cheaper alternative to long-term storage and reanalysis.

Many of these issues involve public policy. Will WGS widen the gap in access to health care by being accessible only to those with financial means? A review and comparison of policies and laws related to genomic testing enacted internationally shows that most countries have no policy guidelines or laws yet formulated and those that do have notable gaps in keeping up with the multiple issues related to genomic testing.[48]

Trying to assess the balance of potential benefit vs risks in the conduct of WGS in healthy people is an inherently unquantifiable task. Benefits may include satisfaction in knowing more about one's genome, for example, discovering an important, actionable condition that was not previously known, understanding more about recessive reproductive risks, and assurance of being generally normal and typical. Risks associated with WGS in healthy people may include disappointment in how little is actually interpretable,[34,49] decisional regret after receiving undesired information that cannot be reeled back,[50] loss of privacy, false expectations, perhaps of the utility of genomics in health management, misinterpretation of VUS or false reassurance from absent findings, and overscreening related to findings of minimal risk. Table 2 lists some of the pros and cons of WGS in healthy people.

CONCLUSION

Dramatic reductions in costs due to multiple new technologies have permitted WGS to be conducted for interested individuals in the general population. Arguments can be made for offering WGS to healthy people, but there are important reasons for caution about using this approach in health care at present. There are insufficient data and such rapidly changing data that no systematic and comprehensive analyses can currently give a definitive analysis of risks and benefits of WGS in healthy individuals. Likely, over time, the circumstances under which WGS becomes a useful undertaking will come to focus. Until then, data must be collected and careful attention must be paid to lessons learned as the cohort of early adopters grab a hold of this promising, powerful technology.

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Abbreviations and Acronyms: **ACMGG** = American College of Medical Genetics and Genomics; **EMR** = electronic medical record; **GWAS** = genome-wide association study; **HGP** = Human Genome Project; **SNP** = single-nucleotide polymorphism; **WES** = whole-exome sequencing; **WGS** = whole-genome sequencing; **VUS** = variant of unknown significance

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The Symposium on Precision Medicine will continue in an upcoming issue.

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