Practical and Ethical Issues with Genetic Screening

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Clinical hematologists are faced with a growing list of new genetic-based tools for identifying a patient's risk of disease. While many of the disease-specific tests are readily available, validation studies are required. Furthermore, genetic-based tests are being pushed to their technical limits, such as testing a single cell prior to embryo selection and transfer for couples at risk of genetic disease. As a result, misdiagnosis or misinterpretation of the data may result. As new genetic testing opportunities proliferate, the hematologist needs to be aware of the medical and legal issues surrounding their use. Furthermore, the hematologist needs to consider the psychological, ethical and social implications of this new field of genomic-based medicine.

Over the past decade, genetic tests have become available for numerous heritable lympho-hematopoietic disorders. As a consequence, the hematologist can now order a test that may predict with a defined degree of certainty whether an individual will develop a specific disease or related complications. For example, longitudinal studies in patients with Fanconi anemia (FA) have previously shown that patients with specific mutation(s) in FA genes are at higher or lower risk of myelodysplasia (MDS) and leukemia.[1,2] Such genotypic-phenotype correlations allow the hematologist to assess risk of disease progression and early death[3,4] relative to the population as a whole. Importantly, results of genetic tests may also be useful beyond the individual affected by the genetic disorder. Depending upon the disorder, knowledge of carrier status may be important. Further, genetic information in couples known to carry a recessive or dominant single gene defect or sex-linked condition permits the hematologist to counsel the couple on their reproductive options, such as prenatal diagnosis and possible pregnancy termination in the case of an affected fetus, and preimplantation genetic diagnosis (PGD). In vitro fertilization (IVF) in combination with PGD aims to provide such high-risk couples a pregnancy with no chance of genetic disorder transmission.

Patients and their families have the right to expect genetic information and appropriate genetic counseling, and health care providers are obligated to provide such information. While it is clear that the hematologist needs to be aware of the genetic tests relevant to hematology that are now available (**Table 1**), the ability to predict disease poses a number of practical and ethical challenges. Not only must hematologists be aware of the availability of such genetic tests but they must also understand the limitations of the tests and how to interpret the results. Furthermore, the hematologist must consider the ethical, emotional, social and economic consequences of genetic testing on the patient and on immediate and extended family members.

In order to illustrate some of the ethical and practical challenges associated with genetic testing in a hematologist's practice, two representative cases are presented.

Case Histories

Case 1: A 2-month-old male was tested for biallelic BRCA2 mutations because of a history of proven biallelic BRCA2 mutations in an older male sibling. The older sibling had had refractory acute myelocytic leukemia. At age 11 months, the older sibling was found to have elevated chromosomal breakage induced by diepoxybutane (DEB) testing of PHA-stimulated peripheral blood lymphocytes (14.0 breaks/cell) as reported elsewhere.[4] History was remarkable for intrauterine growth retardation and failure to thrive. Physical findings consisted of café au lait spots and

Table 1. Candidate lympho-hematopoietic diseases for genetic testing.

- Fanconi anemia
- Sickle cell anemia
- Thalassemia
- Hemochromatosis
- Diamond-Blackfan anemia
- Kostmann neutropenia
- Glanzmann thrombasthenia
- Wiskott-Aldrich syndrome
- Severe combined immune deficiency
- Adrenoleukodystrophy
- Gaucher disease
- BRCA2
- Hemophilia A and B

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microcephaly. Family medical history was remarkable for brain tumor in the maternal great grandmother diagnosed at age 50 years; no cases of breast, ovarian or other BRCA2-associated cancers were known. Germline mutations were proven by analysis of the coding regions and intron/exon junctions of breast cancer susceptibility gene *BRCA2* was performed on genomic DNA, utilizing direct DNA sequencing (Myriad Genetic Laboratories, Inc.).

At age 5 months, a HLA-matched unrelated donor was identified. Due to the high risk of acute leukemia in patients with biallelic mutations in BRCA2,[4] the patient's family was counseled on the potential risks and benefits of 'prophylactic' hematopoietic stem cell transplantation (HSCT). During the evaluation, an occult Wilms tumor, which has also been associated with BRCA2,[5] was identified. Four weeks after nephrectomy alone, the patient underwent HSCT.

This case illustrates at least one major ethical challenge: the development of a treatment plan based on preliminary clinical genotype-phenotype observations (e.g., association between BRCA2 and acute leukemia with resultant plan for 'prophylactic HSCT').

Case 2: A 3-year-old female previously diagnosed with FA was referred for evaluation of bone marrow failure and genetic counseling. Past medical history was remarkable for multiple congenital anomalies, including bilateral radial ray defects, bilateral congenital hip dislocations and left ear deafness. The diagnosis was confirmed by excess chromosomal breakage upon DEB exposure. Mutation analysis revealed that the patient was homozygous for the FANCC IVS4 A>T mutation.[6] At age 2 years, pancytopenia was first observed and treatment with oxymethalone was initiated. On evaluation, the patient had moderate pancytopenia, hypoplastic marrow without MDS or leukemia and normal karyotype. As patients with FANCC IVS4 A>T mutations are at high risk of early myelodysplastic syndrome and acute myelocytic leukemia (median age of onset 8 years) and early death,[3] counseling included heightened marrow surveillance as well as reproductive risks and choices, including potential for embryo selection.

The parents elected to undergo IVF with PGD to have a healthy child who was HLA-identical with the sibling with FA. Over the succeeding 4-year period, there were 5 IVF cycles with 51 embryos tested and 7 embryos transferred. During this period of time, the affected child's disease progressed. Histopathology and cytogenetics revealed multilineage dysplasia and 46 XX, der(1)t(1;3)(p36.1;q21), der(17)t(1;17)(q23;p11.2), respectively. The final embryo transfer successfully implanted and the child was delivered on August 29, 2000. The umbilical cord blood was collected. HLA typing and DEB testing confirmed HLA identity and the carrier (FA negative) status. The proband underwent HSCT from her HLA-matched brother. The child is alive with normal hematopoietic function 5 years later.

This case illustrates at least 7 major ethical challenges: 1) increased risk of leukemia resulting from a delay in HSCT, while pursing a fifth cycle of IVF and PGD, 2) use of PGD to select genetic traits with no inherent benefit to a child, 3) assessment of appropriate parental motivation (i.e., creation of child who will be a loved and cherished member of the family versus creation of a life-saving HSC donor?), 4) absence of an unbiased advocate for the child-to-be, 5) conflict of interest potentially related to the publicity surrounding this case, 6) absence of a centralized review of all aspects of the clinical investigation (rather IRB approval was granted for each of the individual technologies of IVF, PGD and HSCT), and 7) excess number of embryos due to high exclusion rate.

Issues Relevant to the Case Histories

BRCA2 genes and the hematologist

180,000 new cases of breast cancer develop in the US annually with ~5%-10% considered to be hereditary. In the early 1990s, causal associations between BRCA mutations and breast and ovarian cancer susceptibility were identified.[8] Recently, it was determined that BRCA proteins play a critical role in enabling cells to repair DNA damage. Disruption in DNA repair results in increased cancer risk.[9]

Biallelic mutations in BRCA2 have recently been shown to result in an exceptionally high risk of acute leukemia as well as brain and renal cancers with age of onset prior to 6 years.[4,5] These children have an FA-like phenotype including short stature and failure to thrive, café au lait spots and hypersensitivity to clastogeneic agents, such as DEB. As both parents are obligate carriers of a BRCA mutation, increased incidence of breast and ovarian cancer is often (but not always) observed. As a result of these observations, several recommendations have been proposed:

1) Children with an FA phenotype presenting with acute leukemia, brain tumor (particularly medulloblastoma) or Wilms tumor prior to age 6 years should be evaluated for BRCA2 mutations.
2) Children with known biallelic mutations in BRCA2 should have intensive surveillance for leukemia and solid tumors of the brain and kidney with possible consideration of 'prophylactic' HSCT prior to the development of acute leukemia. After reaching adulthood, patients should be monitored for BRCA2-related cancers and some should be offered prophylactic surgery and chemoprevention.
3) Families of children with biallelic mutations in BRCA2 should have genetic counseling regarding the risks of solid tumors in *BRCA2* carriers and the potential use of preimplantation genetic diagnosis to eliminate the risk of FA in future generations within the family.

However, these observations have yet to be validated with larger numbers.

As for the patient discussed in Case 1, upon recovery after diagnosis of occult Wilms tumor and nephrectomy,

the patient underwent HSCT from an unrelated donor. The child had multiple severe regimen-related toxicities and died 3 months after HSCT. Having had 2 affected children, the parents are now considering IVF and PGD, and embryo selection as a means to have a healthy (non-carrier) child. Uncertainties regarding cancer risk and the specific mutations within this family have led to close surveillance in the couple and genetic testing in some extended family members.

However, other ethical issues are now surfacing. As a maternal and paternal grandparent are likely BRCA2 carriers and other extended family members are likely to have inherited a mutant BRCA2 allele, it is unclear how to inform them of their cancer risk. In contrast to adults requesting genetic screening (e.g., BRCA testing) because of strong family cancer history, Case 1 illustrates an instance where there is no strong BRCA2-associated family cancer history. Further, even for those BRCA2 mutations that have been associated with high cancer risk, gene penetrance varies.[10-12]

Preimplantation genetic diagnosis, embryo selection and the hematologist

FA is an autosomal recessive genetic disease that is characterized by multiple congenital physical abnormalities, progressive bone marrow failure and marked predisposition for acute myelocytic leukemia and epithelioid cancers, particularly of the head and neck.[1] The only proven means to potentially cure the hematological complications of FA is by HSCT from a healthy allogeneic donor. When available, HLA genotypic identical donors are used, as survival rates of 65%-100% have been reported in this setting.[13] However, in approximately 80% of patients, a healthy, unaffected, HLA-identical sibling donor is not available. In the setting of partially HLA-matched related or unrelated donor HSCT, morbidities and mortality rates are considerably greater with overall survival rates of 18%-65% reported in the literature.[1]

Therefore, faced with a high probability of requiring HSCT, many couples with an affected child and no existing HLA-identical sibling donor have been highly motivated to have another child—one that is both healthy and HLA matched. In fact, in the late 1980s and early 1990s, without IVF and PGD, couples had multiple pregnancies in attempt to have a HSC donor.[6] Although desperate to have a healthy child, the process was both inefficient (18.75% chance of desired outcome) and high risk (25% chance of having an affected fetus). Therefore, with the realization that new technologies could potentially alter the risk and potentially increase the efficiency, many couples pursued the option of IVF and PGD.

As for the patient discussed in Case 2, the proband underwent HSCT from her HLA-matched brother and remains alive with normal hematopoietic function 5 years later (September 2000). The sibling donor remains healthy and is loved as a cherished family member. Other than blood sampling to verify health status prior to HSCT, the donor has never had any invasive procedure (only banked umbilical cord blood was utilized). Subsequently, the couple attempted to have additional healthy child using remaining cryopreserved healthy embryos but unsuccessfully.

Discussion

Genetic Testing and Counseling

Using genetics to predict disease poses multiple risks and ethical issues, as outlined in **Table 2**. Patients and their physicians must be able to understand the potential implications of the results. Further, the decision to test children for risk of genetic disease is complicated by the fact that parents must make decisions on their behalf until they reach 18 years or are deemed emancipated minors. Testing for genetic conditions, such as sickle cell anemia, is relatively uncontroversial as there are significant benefits to be derived from early diagnosis and medical treatment. However, genetic diagnosis becomes more controversial when benefit is questionable. For example, in Cases 1 and 2, what was gained by the knowledge of the mutations in BRCA2 and FANCC IVS4 A>T, respectively? While genotype-phenotype correlations suggest high risk of leukemia and early death, these outcomes are not universal and existing data are limited.

However, we should go a step further in the case of BRCA2. While the parents are obligate carriers of BRCA2 and should be counseled as to the specific meaning of their mutations, should extended family members be tested?[12,14] And if so, should children in the family be tested, as BRCA2-related cancers rarely occur prior to the age of 18 years? In addition to the health concerns of the individual him- or herself, those with a positive family history of BRCA2 might request testing prior to pregnancy. In an attempt to prevent transmission of the mutant BRCA2, couples at risk could request prenatal testing and subsequent abortion if affected or alternatively IVF and PGD, as in Case 1.

With regard to counseling patients about genetic testing, great emphasis has been placed on a non-directive approach. Specifically, the counselor provides information about genetic risk and explains choices regarding genetic testing and further management. However, the counselor typically does not provide any specific recommendation as

Table 2. Risks of genetic testing

- Discrimination by insurers, employers, schools, others
- Stigmatization
- Psychological distress and harm
- Use of unproven medical therapies
- Use of proven but unnecessary medical therapies
- Misinterpretation and misuse of the genetic test
- Testing errors

to best course of action, thus acknowledging that an individual's decisions are dictated by personal preferences.[15]

Whether or not counseling should be non-directive is a subject of debate. Burke et al,[15] however, argue that genetic tests can be categorized by a joint consideration of clinical validity and availability of effective treatment for persons who test positive. For genetic tests that have high clinical validity and effective treatment (e.g., sickle cell disease), the testing is justified based on the importance of insuring appropriate access to care. For tests that have limited clinical validity but effective treatment (e.g., HFE mutation testing for hereditary hemochromatosis), there may be a net benefit as the treatment (intermittent phlebotomy) is both safe and effective. For tests where clinical validity and treatment efficacy are uncertain (e.g., BRCA2), the value of testing may vary according to different testing contexts (e.g., higher value if the patient's pedigree meets criteria for an autosomal dominant inheritance of breast/ovarian cancer before age 60 years).

Preimplantation genetic diagnosis

PGD was introduced as a viable alternative to prenatal diagnosis for couples at known high risk for conceiving a child with a genetic disease. However, PGD is only possible because of new technologies in reproductive medicine that allow dissection and rapid testing of a single cell from an 8-cell embryo.[16] The process starts with ovarian hyperstimulation, oocyte retrieval and IVF. After 48-72 hours, the embryo typically consists of 6-10 cells called blastomeres. PGD requires a blastomere biopsy by aspirating a blastomere through an opening in the zona pellucida. DNA is extracted from the blastomere, amplified by polymerase chain reaction, and analyzed. Technically, PGD is challenging in that a single cell from each embryo must be accurately analyzed with a 24-hour period to ensure survival of the embryo prior to uterine transfer.

Case 2 illustrates the successful use of the combined technologies of IVF, PGD and HSCT with a good outcome for both the proband and HSC donor.[6] However, risks remain both for Case 2 and others in general. For example, it is possible that the proband could have developed kidney failure as part of FA or transplant procedure.[17] Further, the proband has had a mild but undefined hepatitis that preexisted HSCT. Could kidney or hepatic lobe harvesting from the HSC donor be an option?

While is commonplace for discussion on the ethics of PGD to lead to eugenics and 'designer babies,' the real ethical issues lie with 1) its use outside the realm of research, 2) inequitable access, 3) well-intentioned but misguided uses by IVF clinics, and 4) parental motivation. Based on current experiences and potential risks, a practical series of considerations and recommendations have been proposed regarding the use of IVF and PGD for the purpose of 'creating' an HSC donor (**Table 3**).

Table 3. Practical considerations and recommendations on use of in vitro fertilization (IVF) and preimplantation genetic diagnosis (PGD) for 'creation' of a HSC donor.

- Proband's condition should be life-threatening
- Insure that the risk of donor-child is not increased to benefit the recipient.
- Risk or burden to the donor-child should be limited until the child can decide for self.
- Consider independent psychological evaluation prior to IVF and PGD to determine the couple's ability to rear the child lovingly and without exploitation.
- Prior to any invasive harvesting (marrow or organ harvest), donor child should have an independent physician advocate, and there should be an ethics review. Further, consider psychological evaluation of the parents and donor child to any invasive tissue harvesting.
- Research protections should apply to the 'stacked technologies' of IVF, PGD and HSCT.

Conclusions

Growing access to genetic tests requires greater availability of practical guidelines on their appropriate use and interpretation.[12,14,15,17-19] While such testing is targeted to the healthy, presymptomatic individual with a strong family medical history for the disease, these tests typically only reveal the 'probability' of developing the disease or complication from a disease. Accordingly, some individuals carrying the mutant gene will never develop the disease or complication. Furthermore, laboratory errors remain a risk. The health care provider is legally obligated to provide genetic testing when appropriate and access to high quality genetic counseling. Medical malpractice cases have held health care providers liable for not informing patients of their genetic risk status. Therefore, the practicing hematologist must be more aware of the both the technical aspects of relevant genetic tests as well as the ethical, social and psychological implications.

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