I. Introduction
   A. Clinical optometry as currently practiced is undergoing a fundamental, revolutionary transformation. This transformation will change the way optometrists understand, classify, diagnose, treat and manage disease.
   B. The Human Genome Project and other revolutionary advances have started to increase and broaden the importance of genetics/genomics in all health care. This revolution will alter eye care forever, and it involves a tremendous change from the “old genetics” to the “new genetics.” Genetics is no longer an esoteric academic specialty that involves rare diseases.
   C. The genetic and molecular basis of the estimated 7,000 suspected Mendelian conditions was understood in less than 2% of those conditions in 1990, whereas in 2011 it was known in about 40% of the conditions. Immediate benefits include access to accurate diagnostic testing, and future benefits will include better treatments.
   D. In tomorrow’s eye exam a variety of genetic, lifestyle, and environmental factors will be measured. These will include: serum proteomics for early detection, gene expression to herald clinical disease, and molecular profiles of tumors to guide therapy.

II. Basic concepts
   A. A new vocabulary comes with the revolution.
      1. The genome is the set of all genetic information (e.g., nuclear genes/chromosomes, and mitochondrial genes) in an organism. In other words, it consists of all the DNA in each human cell.
         a. DNA is made up of a variable sequence of 4 nucleotide bases (A, T, C, & G) – the “letters”
         b. A sequence of 3 bases (the “word”) codes for an amino acid.
         c. A gene (the “sentence”) codes for a protein.
         d. Chromosomes (the chapters of the “book of life”) contain the genes.
         e. The genome is the “book of life.”
      2. The transcriptome is the set of all messenger RNA (mRNA) molecules or transcripts in a cell or organism.
      3. The proteome is the complete set of proteins expressed by a genome, cell, tissue or organism. It is the set of expressed proteins at any given time point.
      4. The metabolome is the set of all metabolites (e.g., metabolic intermediates, hormones) in an organism.
      5. Systems biology is the study of an organism, viewed as an integrated and interacting network of genes, proteins and biochemical reactions which give rise to life. Instead of analyzing individual components or aspects of the organism, such as sugar metabolism or a cell nucleus, systems biologists focus on all the components and the interactions among them, all as part of one system.
   B. The general flow of genetic information is from DNA to RNA to protein. A set of three nucleotide bases in the DNA sequence specify an amino acid in the protein sequence. Thus the “blueprint for life” consists of a 4-letter alphabet and 3-letter words.

III. Human Genome Project (HGP)
   A. This was a 15-year worldwide research effort (1990-2005).
   B. It was considered so important that the National Human Genome Research Institute was formed at NIH.
   C. The HGP involved the sequencing of human DNA (3 billion base pairs).
   D. The HGP was completed ahead of schedule and under-budget in April of 2003.
   E. It sequenced human DNA and identified the 20,000 to 25,000 genes in human DNA.

IV. Paradigm shift
   A. The “old genetics”.
      1. The old genetics dealt with conditions caused by a mutation in a single gene or by an extra or missing chromosome or part of a chromosome.
2. These conditions are **relatively rare**. Most patients are not directly affected by them. Thus, these conditions play a relatively small role in health care.
3. The conditions are rare enough that genetics care could be provided by medical geneticists and genetic counselors, with occasional involvement of primary care providers.

**B. The “new genetics”**.
1. The new genetics emerged due to knowledge derived from the HGP.
2. **Essentially all medical conditions have a genetic component and can be viewed through a “genetic lens.”**
3. The new genetics **deals with multifactorial conditions that are partly caused by mutation(s) in gene(s).**
   a. Multifactorial systemic conditions include diabetes, atherosclerosis, hypertension, Alzheimer disease, colon cancer, breast cancer, etc.
   b. Multifactorial ocular conditions include the glaucomas, age-related macular degeneration (AMD), cataracts, myopia, diabetic retinopathy, etc.
   c. **While many genes are involved, most genes have small effects.**
4. These conditions are **common** enough that genetics care will be supplied mainly by primary care providers from many health disciplines with occasional involvement of medical geneticists and genetic counselors.

**5. Multifactorial diseases**
   a. Many genes involved along with environmental influences
   b. Most genes involved have small effects

   a. GEI was launched in 2006 to support efforts to identify major genetic susceptibility factors for diseases of substantial public health impact and to develop technologies for reliable and reproducible measurement of potentially causative environmental exposures.
   b. This research will help us understand genetic contributions and gene-environment interactions in common diseases.

**7. Essentially all disease is a result of genes interacting with the environment.**
   a. **Ex.: Smoking and lung cancer**
      1) Smoking is the cause of almost all small-cell lung cancer.
      2) Ex.: 51-year-old man with lung cancer
         (a) > 50,000 mutations were found in his lung cancer cells vs. his normal somatic cells
         (b) Caused by the > 60 carcinogens in smoke
         (c) Smoked 25 cigarettes/day for 15 years
         (d) 1 mutation for every 3 cigarettes smoked
   b. **Ex.: UV and cancers/growths**
      1) **UV can trigger DNA mutations that cause skin melanomas, basal cell carcinomas, squamous cell carcinomas, pterygia, & pinguecula.**
      2) Ex. 45-year-old man with malignant melanoma
         (a) 33,000 mutations found in the cancer cells
         (b) Caused by UV exposure from sunlight as a child

**8. There will be a shift from an emphasis on disease treatment to health maintenance.**

**9. The rate of discovery of genes has accelerated due to:**
   a. The **International HapMap Project**.
   b. A **profound decrease in the cost of sequencing genes**.
   c. The **rise of Genome-Wide Association Studies (GWAS).**
      1) GWAS have identified coding variants in genes and noncoding variants associated with common diseases and traits.
      2) This method **searches the genome for small variations, called single nucleotide polymorphisms or SNPs (pronounced “snips”), that occur more frequently in people with a particular disease than in people without the disease.** Each study can look at hundreds of thousands of SNPs at the same time. The data from this type of study are used to pinpoint genes that may contribute to a person’s risk of developing a certain disease.
      3) Such studies are particularly **useful in finding genetic variations that contribute to common, complex diseases, such as age-related macular degeneration, asthma, cancer, diabetes, heart disease and mental illnesses.**
4) As of February 2014, the database “A Catalog of Published Genome-Wide Association Studies,” hosted by the National Human Genome Research Institute (http://www.genome.gov/gwastudies), listed more than 12,462 associations between single nucleotide polymorphisms (SNPs) and complex traits with \( P < 10^{-5} \) coming from more than 1,800 GWAS publications.

V. DNA Polymorphisms.

A. A Single Nucleotide Polymorphism (SNP)

1. A SNP is a DNA sequence variation occurring when a single nucleotide (A, T, C, or G) in the genome differs between individuals or between paired chromosomes in an individual.
2. It is a site in the genome where the DNA base varies in at least 1% of the population.
3. A SNP is the most common type of genetic variation, and may occur every 100 to 300 bases.
4. Single nucleotide polymorphisms may fall within coding sequences of genes, non-coding regions of genes, or in the intergenic regions between genes.
   a. SNPs within a coding sequence will not necessarily change the amino acid sequence of the protein that is produced, due to degeneracy of the genetic code.
   b. A SNP in which both forms lead to the same polypeptide sequence is termed synonymous (sometimes called a silent mutation) - if a different polypeptide sequence is produced they are non-synonymous.
   c. SNPs that are not in protein-coding regions may still have consequences for gene splicing, transcription factor binding, or the sequence of non-coding RNA.
5. Variations in the DNA sequences of humans can affect how humans develop diseases and respond to pathogens, chemicals, drugs, vaccines, and other agents.
6. A key aspect of research in genetics is the association of sequence variation with heritable phenotypes. It is expected that SNPs will accelerate the identification of disease genes by allowing researchers to look for associations between a disease and specific sequence differences (SNPs) in a population of individuals.

B. The DNA of any two people is about 99% identical.


1. Million's of SNPs exist in the human population. Of these, more than 8 million have been identified via this project.
2. Sets of SNPs close together tend to be inherited as a block.
3. A “haplotype” is this pattern of SNPs in a block (region of a chromosome).
4. The goal of the project was to develop a haplotype map of the human genome which would describe the common patterns of human DNA sequence variation.
5. 4 populations were sampled: Nigerian, Japanese, Chinese, and U.S. (Northern/Western Europe ancestry). A total of 270 people were tested.
6. The project revealed that a smaller set of 300,000 to 500,000 variants can serve as a proxy for the roughly 10 million common genetic variants in the human genome.
7. This smaller set can now be used in GWAS studies to find the genetic bases of common diseases. This lead to the first gene found for AMD (CFH). The HapMap has been used to find more than 100 regions of the genome that are associated with common human diseases such as coronary artery disease and diabetes.

D. The 1000 Genome Project (started January 2008) (http://www.1000genomes.org/)

1. An International Consortium was formed to sequence the genomes of at least 1000 people from around the world. They now plan on sequencing about 2,500 samples from about 25 populations around the world.
2. The goal of the project is to find most genetic variants that have frequencies of at least 1% in the populations studied. This information can then be used in genome-wide association studies (GWAS) relating genetic variation to disease.

E. The VA's Million Veteran Program (started in 2011) (http://www.research.va.gov/MVP/default.cfm)

1. This is a longitudinal genomics research study in which the VA is aiming to enroll one million veterans by 2017. More than 240,000 have already signed up. This number already exceeds the enrollment numbers of any single VA study or research program in the past.
2. The aim of the program is to use the VA's medical and other personnel records with genetic information to identify gene-health connections that could be used to screen for, diagnose, and prognose diseases, as well as develop personalized therapies.
3. With informed consent, VA researchers are collecting biological samples for genetic analysis and are giving volunteers questionnaires to gather data on their lifestyle, health, and military exposure.
4. The VA has contracted with a number of genetic testing firms to genotype veterans, as well as sequence the exomes or the whole-genome of certain participants.

5. “Data from MVP is being stored in a different database than the EMR system containing veterans’ healthcare information. MVP findings are for research use only and the genetic testing is not being performed under clinical laboratory conditions that would enable using the information to inform patient care.”

6. All samples will be stored in a secure VA biorepository. All samples and health information will be labeled with a code so that no one can directly identify participants. Researchers who are approved access to analyze samples and data will not receive name, date of birth, address, or SSN of participating veterans.

   1. The TCGA is a joint project of the National Human Genome Research Institute (NHGRI) and the National Cancer Institute (NCI) to comprehensively apply genome analysis technology to the study of the biomolecular basis of cancer.
   2. This project has analyzed tumor and normal samples from over 6000 patients, which resulted in the collection and public availability of clinical and genomic data from 33 cancers. These data types include gene expression, single-nucleotide polymorphism, miRNA, copy number, DNA methylation and somatic mutations, along with tissue slide images and clinical outcomes.
   3. This will improve our ability to diagnose, treat, and prevent cancer.

G. The ENCyclopedia Of DNA Elements (ENCODE) project (launched by the National Human Genome Research Institute (NHGRI) in 2003) (http://www.genome.gov/Encode/)
   1. HGP found that < 2% of the DNA codes for proteins.
   2. ENCODE sought to figure out the role of the rest of the DNA sequence (previously considered to be “junk”). At least 80% of the genome had biochemical function & was capable of being transcribed.
   3. The main goal was to identify functional elements including protein-coding and non-coding RNAs, gene regulators (e.g., promoters, enhancers, silencers), and elements linked to epigenetic mechanisms.
   4. Epigenetics is somatically heritable variation in gene expression that does not involve changes in DNA base sequence. It includes such processes as DNA methylation, histone modification, and nucleosome remodeling. These are biochemical features of the genome that superimpose (epi, “upon” or “above”) the DNA sequence, but still act in modulating gene expression.

VI. Molecular optometry.
   A. Clinical optometry is undergoing a fundamental, revolutionary transformation.
   B. This will change the way optometrists:
      1. Classify diseases
      2. Understand diseases
      3. Diagnose diseases
      4. Treat & manage diseases.
   C. Look at the patient through a “genetic lens” (i.e., incorporate genetic thinking & principles).
      1. Using this lens effectively means rejecting the notion of normality & embracing the idea of variation. The changing paradigm of medicine is framed around individual variability rather than the distinct separation between “normal” and “abnormal.”
      2. Every patient is an individual & there is infinite variation on the spectrum of health & disease.
      3. The clinician should take into account the complex interrelationships among genes, along with each patient’s protein profile, environmental experiences & exposures.
      4. We should no longer regard the human body & the visual system as a biological machine in which the OD acts as a mechanic when the parts break down. The clinician’s challenge is to understand the variability of the genetic & environmental factors that lead to disease, develop a prevention plan, and, if necessary, a treatment plan based on the patient’s unique variability.

VII. Improved classification of diseases.
   A. The present system is based on clinical description (phenotype).
   B. Knowledge from the HGP & other advances will allow a more rational classification based on genetic causes (genotype) and influences.
   C. Ex. Retinitis pigmentosa.
      1. This is a group of retinal dystrophies.
2. It includes autosomal dominant (AD), autosomal recessive (AR), X-linked recessive (XR), and mitochondrial forms of inheritance.
3. It results from mutations on at least 10 chromosomes (48 loci; 37 genes).
4. Knowing the particular mutation in your patient will eventually determine the appropriate therapy.

D. Ex. Corneal dystrophies - (At least 14 genes have been identified on 12 chromosomes)

VIII. Improved understanding of pathogenesis/pathophysiology.

A. We are moving toward a molecular level of understanding of diseases & conditions.

B. Twin studies yield heritability estimates for a number of ocular traits
1. Cataracts: nuclear = 48%; cortical = 55% (In other words, genetics accounts for about 48% of nuclear cataract variability & about 55% of cortical cataract variability)
2. Myopia: ≈ 71% (meta-analysis result)
3. Central corneal thickness: ≈ 85% (meta-analysis result)
4. Intraocular pressure: ≈ 55% (meta-analysis result)
5. Optic nerve head disc area = 72% and cup area = 75% (meta-analysis results)
6. Anterior chamber depth: ≈ 70% (meta-analysis result)

C. Familial correlation and heritability analysis of data from the Beaver Dam Eye Study yield heritability estimates for traits underlying refraction:
1. Spherical equivalent: 58%
2. Corneal curvature: 95%
3. Axial length: 67%
4. Anterior chamber depth: 78%

D. Estimates of heritability for various disorders from twin studies.
1. Obesity 0.80
2. Type 2 diabetes 0.75
3. Schizophrenia 0.65
4. Hypertension 0.55
5. Alcoholism 0.55
6. Cirrhosis 0.50
7. AMD 0.45
8. Atherosclerosis 0.40
9. Breast cancer 0.35
10. Type 1 diabetes 0.25
11. Open-angle glaucoma 0.13

E. Ex. The Glaucomas.
1. Glaucoma is the 2nd leading cause of blindness in the U.S. (after macular degeneration).
2. The prevalence of open-angle glaucoma (OAG) in the U.S. in people over 40 years old is ~2%.
   a. Prevalence increases with age (e.g., 0.9% for people 43-54 years old; 4.7% for people over 75 years old).
   b. Black subjects have about 3-5 times the age-adjusted prevalence of OAG compared to whites.
   c. It is noteworthy that glaucoma is undiagnosed in nine of ten people worldwide and is undiagnosed in ~50% of those in developed countries.
3. There is about a 10-fold increase in risk for first-degree relatives (22% lifetime risk) of glaucoma patients. This is associated with genetics and means that you should screen relatives of your glaucoma patients (i.e., cascade genetic screening).
4. The glaucomas can be inherited as:
   a. autosomal dominant (e.g., Juvenile-onset Open-Angle Glaucoma (JOAG), pigment dispersion),
   b. autosomal recessive (e.g., primary congenital glaucoma (PCG)), &
   c. multifactorial forms (e.g., most adult-onset types of glaucoma – e.g., primary open-angle glaucoma (POAG), Normal-Tension Glaucoma (NTG), Exfoliation Glaucoma (XFG)).
5. Identification of genes for the glaucomas (e.g., MYOC, OPTN, WDR36, ASB10, CYP1B1, & LOXL1)
6. Exfoliation glaucoma (XFG; also known as pseudoexfoliation glaucoma).
   a. XFG is the commonest cause of secondary glaucoma. It affects up to 12% of all glaucoma patients in the U.S. & its prevalence varies by ethnic group. The prevalence of exfoliation syndrome (XFS) in the U.S. is about 1.6 to 1.8%.
   b. SNPs in the gene LOXL1 account for most of the XFG cases. The population attributable risk (PAR) in various studies was 99% (Iceland/Sweden), 88% (Iowa), & 71% (Utah).
c. The risk of pts carrying 2 copies of the high-risk SNPs is about 700 times the risk of pts carrying 2 copies of the low-risk SNPs and about 2.47 times the population average risk.

d. The product of the gene modifies elastin fibers that are a major part of exfoliation syndrome changes.

e. Testing for LOXL1 variants is available.

F. Ex. Age-related macular degeneration.

1. Introduction.
   a. AMD is the most common cause of blindness in the elderly in the Western world.
   b. Its prevalence is 0.05% before the age of 50 and 11.8% after the age of 80.
   c. 1st-degree relatives of patients with AMD have a 4.2 times increased lifetime risk of late AMD.

2. CFH gene.
   a. A polymorphism in the complement factor H gene (CFH) on chromosome 1q31 accounts for 20-50% of the overall risk of developing AMD.
   b. Individuals who carry a single copy of this variant have a 2- to 4-fold increased risk of AMD. Individuals who carry two copies of the variant have a 5- to 7-fold increased risk.
   c. Atrophic tissue damage and neovascularization are potential results of chronic inflammation mediated by the complement system. Factor H is a negative regulator of the complement system, is made in the macula, & is present in drusen.

3. LOC387715/ARMS2 (Age-Related Maculopathy Susceptibility 2) & a secreted heat-shock serine protease HTRA1 (High Temperature Required Factor A-1) gene.
   a. The LOC387715/ARMS2 & HTRA1 genes are extremely close to each other on chromosome 10q26, & polymorphisms in them account for as high as 57% of the overall risk of developing AMD.
   b. Individuals who carry a single copy of this variant have a 3-fold increased risk of AMD.
   c. If a patient were homozygous for risk alleles at both CFH and LOC387715HTRA1, the disease odds ratio (OR) would be about 58.

4. Complement Factor B (CFB) & Complement Component 2 (C2) genes
   a. Variants of these genes can confer a significantly reduced risk of AMD & other variants can increase the risk. CFB is a complement activating factor.
   b. Analysis of the CFH, CFB, and C2 variants can predict the clinical outcome in 74% of AMD pts & 56% of controls.

5. Other complement-related genes.
   a. SNPs in the C3 gene, on chromosome 19q13, increase the risk of AMD 1.8-fold for 1 risk allele or 2.4-fold for two risk alleles.
   b. A SNP just 3′ of complement factor I (CFI) demonstrated an odds ratio of 1.31.
   c. One study implicated a C7 SNP (rs2876849) as being protective for AMD.
   d. A SNP variant (rs2511989) in the SERPING1 gene encoding the complement component 1 (C1) inhibitor may also be involved.

6. Two companies currently are selling genetic tests to determine a patient’s risk of AMD progression. Should you be ordering one of these tests for some of your patients? (see below). What added value does this genetic testing bring to surveillance and management, if any?

7. Innate immunity and inflammation in general and complement overactivation in particular appear to play a central role in AMD pathogenesis.

8. In addition to genes in the complement pathway, gene variants involved in oxidative stress, extracellular matrix remodeling, and lipid metabolism have been associated with AMD.

9. The new genetic data suggest new targets for early intervention & predictive DNA testing as a future option. It may also lead to the development of new biomarkers for the disease. The challenge is to develop risk models incorporating gene-gene interactions, gene copy-number variations, epigenetics, environmental interactions, Tx effects, & clinical covariates.

IX. Improved diagnostic testing.
   A. Thinking will shift to probabilities (statistical risks) instead of definitive answers.
   B. Microarrays (DNA chips, RNA chips, Protein chips), “Lab-on-a-chip” microfluidics, point-of-care testing?
   C. Single-cell DNA and mRNA sequencing are now possible.
   D. Types of genetic tests.
1. **Molecular genetic test** – tests for a genetic condition by testing for nucleic acid (DNA or RNA)
   a. **Single-gene testing** (testing for variants in a single gene)
   b. **Whole-exome sequencing** (sequencing all DNA elements that encode proteins, representing a little over 1% of the genome)
   c. **Whole-genome sequencing**.
2. **Cytogenetic test** – tests for anomalies of number or structure of chromosomes.
3. **Biochemical genetic test** – test to study enzymes, proteins or metabolites that may be altered by a genetic condition.

**E. Goal of $1000 whole-genome sequencing.** The cost of sequencing the first genome (HGP) was about $3,000,000,000.
1. In January of 2014, Illumina was the first to claim that the cost of sequencing a human genome with their new technology is less than $1,000. The company says their instrument has the ability to sequence five whole human genomes in a single day.
2. Also in 2014, Life Technologies claims that its Ion Proton™ System can sequence a whole human genome for about $1,000 in a few hours.
3. Human exome sequencing can be obtained for $899 (BGI Americas).

**F. Reasons for genetic testing.**
1. **Family health history assessment** – Use of information about the health status of relatives to estimate risks for an individual based on shared genes and environment
2. **Predictive testing**: offered to asymptomatic individuals with a family history of a genetic disorder
   a. **Presymptomatic testing** for predicting the future onset of genetic disorders (e.g., Huntington disease)
   b. **Susceptibility (predispositional) testing** for variants that increase the risk for a disease, but where many people who test positive will not actually develop the condition (e.g., AMD, breast cancer, Alzheimer disease).
3. **Diagnostic testing of a symptomatic individual**: Testing to determine whether symptoms are caused by a genetic disorder and potentially to guide treatment
4. **Carrier testing**:
   a. This kind of testing identifies individuals who are themselves usually unaffected but are carriers of a recessive-disease-causing mutation. They could pass a mutated gene on to children who could show effects.
   b. These are tests on blood or other tissue samples (e.g., buccal scrapings) used to determine if an individual has a single copy of a mutant gene that is inherited in an autosomal recessive or an X-linked recessive manner.
   c. This kind of testing is offered to patients who have family members with a genetic condition, family members of an identified carrier, or patients in ethnic or racial groups known to have a high carrier rate for a particular disorder.

**Table 1. Examples of ethnicity-based carrier screening**

<table>
<thead>
<tr>
<th>Disease</th>
<th>Ethnic group</th>
<th>Carrier frequency</th>
</tr>
</thead>
<tbody>
<tr>
<td>Hereditary hemochromatosis</td>
<td>Northern European</td>
<td>1/8</td>
</tr>
<tr>
<td>Cystic fibrosis</td>
<td>European</td>
<td>1/25</td>
</tr>
<tr>
<td></td>
<td>Hispanic</td>
<td>1/46</td>
</tr>
<tr>
<td></td>
<td>African</td>
<td>1/60-65</td>
</tr>
<tr>
<td></td>
<td>Asian</td>
<td>1/150</td>
</tr>
<tr>
<td>Sickle cell anemia</td>
<td>Black</td>
<td>1/14</td>
</tr>
<tr>
<td>Gaucher disease</td>
<td>Ashkenazi Jewish</td>
<td>1/12</td>
</tr>
<tr>
<td>Tay-Sachs disease</td>
<td>Ashkenazi Jewish</td>
<td>1/26</td>
</tr>
<tr>
<td>Thalassemia</td>
<td>African/Asian/Mediterranean/Middle Eastern</td>
<td>Varies with population</td>
</tr>
</tbody>
</table>

5. **Prenatal diagnostic testing (aka antenatal testing):**
   a. This involves testing conducted on an embryo or fetus prior to birth to assist parental reproductive decision making.
   b. Collection procedures include **amniocentesis** (between 15 and 20 weeks’ gestation), and **chorionic villus sampling** (at 10 to 12 weeks’ gestation). More specialized procedures include **periumbilical blood sampling** (PUBS; after 18 weeks of gestation), placental
biopsy, and fetoscopy with fetal skin biopsy. There is also a new noninvasive test that involves the isolation of cell-free fetal DNA from the maternal circulation (10-22 weeks' gestation).

6. Newborn screening: Screening conducted as part of a public health program that tests most newborns for rare genetic conditions that have treatment options

7. Pharmacogenomic testing. Testing for variants known to influence how drugs are processed, leading to personalized use or dosage recommendations

8. Forensic testing

9. Genealogical testing (ancestry testing)

   a. After *in vitro* fertilization, a 6-8 cell embryo can be tested for a genetic condition prior to transfer of the embryo to the mother's uterus.
   b. This could allow a woman to avoid birth of an affected offspring without undergoing prenatal diagnosis and pregnancy termination.
   c. It is generally offered to couples who have a high probability of having a child with a serious disorder.
   d. This technique has already been used in the case of monogenic diseases (e.g., retinoblastoma, cystic fibrosis, sickle cell disease, Huntington disease) and chromosomal aberrations (e.g., Down syndrome, aneuploidy screening).
   e. The cost is usually very high, and it is not often covered by medical insurance.
   f. Theoretically, PGD is possible for all diseases that can be diagnosed by direct analysis of DNA.
   g. Acceptability of PGD is not universal.
      1) Jewish and Muslim laws consider the embryo a protected life form only after it has been implanted. This perspective thus allows IVF and PGD.
      2) Catholicism considers the unimplanted embryo as human life. Therefore, this perspective does not allow an abnormal embryo to be discarded.
   h. Mitochondrial replacement. The U.K. is moving toward approving a new type of in vitro fertilization that could enable patients with mitochondrial diseases to avoid passing the condition on to their children. It involves transferring the nuclear DNA from the sperm & the egg of the potential parents into a second egg from a donor.

G. Population screening.
   1. Population screening involves the explicit and systematic application of a diagnostic genetic test across a whole population of asymptomatic people, or a subset of a population like newborn infants or pregnant women.

H. Cascade genetic screening.
   1. This involves screening for genetic variants that predispose individuals or their offspring to disease by targeting relatives of previously identified carriers.
   2. Carrier risk of close relatives of known carriers is generally higher than the population risk.
   3. This type of screening is more efficient than population screening, in the sense that fewer individuals have to be genotyped per detected carrier.
   4. Example: glaucoma.
      a. 10-fold increase in risk for 1st–degree relatives of glaucoma patients.
      b. Therefore screen relatives of your glaucoma patients.

I. Examples of currently available tests/laboratories for ocular diseases:
   1. National Ophthalmic Disease Genotyping Network (eyeGENE; NEI). – see below
   2. The Carver Nonprofit Genetic Testing Laboratory (Univ. of Iowa) (www.carverlab.org) offers relatively cheap testing with fairly fast turn-around times for 26+ ocular disease panels (e.g., cone-rod dystrophy panel, juvenile open angle glaucoma, primary open angle glaucoma, retinitis pigmentosa panel, Stargardt disease panel, Usher syndrome panel)
   3. The Casey Eye Institute Molecular Diagnostics Laboratory (www.ohsucasey.com/diagnostics) offers testing of about 30 ocular disease panels (e.g., anterior segment dysgenesis panel, cone-rod dystrophy panel, Leber Congenital Amaurosis panel, retinal dystrophy panel).
   4. Quest Diagnostics Nichols Institute San Juan Capistrano, CA (http://www.questdiagnostics.com/) offers SNP detection for ARMS2 and CFH gene variants for age-related macular degeneration.
   5. Fulgent Clinical Diagnostics Lab (http://fulgent-therapeutics.com/) offers a macular degeneration panel with next generation sequencing.
   6. Macular Risk® PGx (Arctic Medical Laboratories)
7. **RetnaGene™ AMD test** (Sequenom Laboratories)
8. **Avellino Gene Detection System** (AGDS™; Avellino Lab USA)

J. **Diagnostic testing caveats.**
1. Most SNPs associated with common diseases explain a small proportion of the observed contribution of heredity to the risk of disease – in many cases less than 5 to 10%. This substantially limits the use of these individual markers to predict risk.
2. The future challenge is to develop risk models incorporating gene-gene interactions, gene copy-number variations, epigenetics, environmental interactions, treatment effects, and clinical covariates.

K. **Evaluation of genetic tests.** One important approach is the ACCE framework developed by the Office of Genomics and Disease Prevention at the Centers for Disease Control and Prevention (CDC). An ACCE evaluation begins with a description of the relevant disorder and the setting in which the test will be used. It then moves on to an assessment of the Analytical validity, Clinical validity, Clinical utility, and the Ethical, legal, and social implications of the test.
1. **Analytic validity.** This is the ability of a genetic test to accurately & reliably measure the genotype of interest.
2. **Clinical validity.** This is the ability of a genetic test to accurately & reliably predict the presence or absence of a clinically defined disorder or phenotype of interest.
3. **Clinical utility.** This refers to the likelihood that the test will lead to an improved health outcome. In other words, it is the improved measurable outcomes shown by a genetic test, and the usefulness and added value to patient management resulting from a test compared to current patient management without genetic testing.
4. **Ethical, legal, and social implications.**

X. **Improved management and treatment.**

A. **Identification of patients with susceptibility genes** may help determine:
1. Risk level
2. Types of testing/treatment
3. Timing of surveillance visits
4. Targeting of education and counseling about known modifiable factors.

B. **Gene therapy.**
1. China approved the world’s first commercially-licensed gene therapy (October, 2003). It is an injectable medication (Gendicine) using an adenoviral vector to deliver a p53 tumor suppressor gene for head-and-neck squamous cell carcinoma. The U.S. has not approved any gene therapies yet.
2. The European Medicines Agency granted regulatory approval for Glybera (alipogene tiparvovec) in November of 2012, making it the first gene therapy to receive regulatory approval in the Western world. In patients with lipoprotein lipase deficiency due to mutations in the LPL gene, an injection of normal LPL genes in an adeno-associated virus (AAV1) vector resulted in increased levels of lipoprotein lipase and hence a dramatic reduction in triglycerides, incidence of pancreatitis, and hospitalizations of the patients.
3. **Gene therapy for RPE65 gene mutations causing Leber Congenital Amaurosis (LCA).**
   a. Eight clinical trials on more than 200 humans are underway in the U.S., England, Israel, and France (Clinical phase1 through phase 3).
   b. So far, the clinical trials have indicated that the gene therapy is safe and led to increased vision in the subjects, with the youngest subjects showing the most improvement.
   c. This treatment is the most clinically advanced gene therapy for any retinal degenerative disease.
   d. Spark Therapeutics is a spin-off of the Children’s Hospital of Philadelphia (CHOP). It will commercialize gene therapy for LCA and other inherited retinal diseases. The company plans to seek marketing approval for the RPE65 treatment at the conclusion of its phase 3 study, which is expected in 2015.
4. **Gene therapy for wet age-related macular degeneration (AMD).** A number of clinical trials are underway in the U.S., Australia, and China (Phase 1 to phase 1/2).
5. **Gene therapy for choroideremia.** Currently recruiting for a phase 1/2 trial in England. Phase I trials are expected in the U.S. and Canada in 2014.
a. Choroideremia is an X-linked recessive disorder caused by mutations in the CHM gene, which encodes Rab escort protein-1 (REP1).

b. The disorder affects the retina, retinal pigment epithelium, and choroid, exposing the underlying sclera in patches.

c. The treatment involves a subfoveal injection of adeno-associated viral (AAV) vector carrying normal CHM genes.

6. **Gene therapy for Stargardt disease.** Phase 1/2a trials are underway in the U.S. and France.

7. **Gene therapy for Retinitis Pigmentosa (RP).**
   a. **RP.** A clinical phase 1 trial is underway in Japan using a Simian Immunodeficiency Virus (SIVagm) Vector Expressing Human Pigment Epithelium-Derived Factor (hPEDF) Gene.
   b. **MERTK-related autosomal recessive RP.** A clinical phase 1 trial is underway in Saudi Arabia.
   c. **RP associated with Usher syndrome Type 1B.** A clinical phase 1/2a is recruiting in the US and France to examine the safety of an experimental gene transfer agent, UshStat.

8. **Gene therapy for Leber hereditary optic neuropathy (LHON).** A phase 1 trial has started in China.

9. **Ciliary neurotrophic factor (CNTF) for human retinal degeneration.**
   a. There are 10 clinical trials that have started or have been completed. These included trials for macular telangiectasia, glaucoma, ischemic optic neuropathy, CNGB3 Achromatopsia, RP, & dry AMD. The trials for RP did not show a therapeutic benefit.
   b. CNTF protects rod and cone photoreceptors from apoptosis during retinal degeneration.
   c. CNTF is also a neuroprotective factor and an axogenesis factor for retinal ganglion cells.

C. **Human stem cell therapies for macular degeneration and dystrophies.**
   1. At least twelve phase 1/2 clinical trials are underway for wet and dry AMD, Stargardt macular dystrophy, and retinitis pigmentosa.
   2. Many use autologous bone-marrow derived stem cells or human embryonic stem cells.

D. **Induced pluripotent stem cell (iPSC) therapy for wet AMD.**
   1. A new pilot clinical study in Japan involves the establishment of autologous iPSCs from each of the research participants, which will then be differentiated into RPE using a novel technology that allows these epithelial cells to be transplanted in monolayer cell sheets without the use of synthetic scaffolds or matrices. The cell sheets will be shaped into 1.3 × 3 mm grafts and transplanted into the affected site of a single eye, following excision of the damaged RPE and neovascular tissues.
   2. This pilot study follows on extensive preclinical safety and feasibility testing in rodent and non-human primate models.

E. **Antisense drugs.**
   1. These drugs are complementary strands of portions of messenger RNA (mRNA).
   2. The drugs bind to mRNA and inhibit transcription of the protein.
   3. Vitravene (fomivirsen) was the first antisense drug marketed (1998). It was used for treating cytomegalovirus (CMV) retinitis. It was discontinued in 2004 because it was unprofitable.
   4. Kynamro (mipomersen) was approved by the FDA in January 2013 for treatment of homozygous familial hypercholesterolemia. This is a second-generation drug that targets the mRNA for apolipoprotein B–100 (ApoB-100) and aids in reducing apolipoprotein B, low-density lipoprotein-cholesterol (LDL-C), total cholesterol, and non-high density lipoprotein-cholesterol (non HDL-C).
   5. A 2nd-generation antisense inhibitor, iCo-007 (iCo Therapeutics Inc.), targets C-raf kinase mRNA & has completed phase I clinical trials for treating diabetic macular edema and retinal neovascular diseases like diabetic retinopathy and wet AMD. This drug may prevent the growth of new blood vessels and inhibit increased vascular permeability by decreasing the production of C-raf kinase through which multiple growth factors (e.g., VEGF) signal. Phase I indicated that the drug was safe and showed an encouraging trend to decrease retinal thickness (towards the normal value). Phase II testing is ongoing.

F. **RNA interference (RNAi).**
   1. Small double-stranded interfering RNAs (siRNAs) can silence messenger RNAs carrying a complementary sequence. siRNAs occur naturally in mammalian cells and can also be generated artificially and used in therapeutic clinical trials.
   2. RNAi is used to selectively inhibit gene expression in various diseases, including infectious diseases, cancer, inflammation/immune dysfunction, CNS disorders, and cardiology.
3. RNAi drugs for AMD & other ocular disorders.

G. Complement-targeted therapeutics for AMD.

XI. Pharmacogenetics / Pharmacogenomics.
   A. View interactive tutorial on Pharmacogenomics (http://www.phgfoundation.org/tutorials/home/)
   B. Each year, 100,000 people die from adverse reactions to drugs & over 2 million have serious reactions.
   C. Some drugs work well in some patients & not as well in other patients.
   D. DNA variations (SNPs, insertion/deletions, & copy-number variations) in genes that are involved in drug metabolism, drug transportation, drug targets, & intracellular signaling pathways can account for much of the ability of some drugs to cause adverse reactions and/or to be ineffective.
   E. The Amplichip CYP450 Microarray (Roche/Affymetrix) is now commercially available for testing two of the genes involved in drug metabolism (CYP2D6 & CYP2C19). This test accounts for ~99% of known poor and ultra-rapid metabolizer genetic variation in these genes.
   F. ODs will prescribe medications and determine drug doses based on the patient’s genotype and SNP pattern:
      1. Improving efficacy & safety of drugs
      2. Decreasing morbidity & mortality
      3. Decreasing overall cost of health care.
   G. “Personalized medicine” will evolve for many drugs and diseases by the year 2020.
   H. Example: Aminoglycoside-induced ototoxicity.
      1. A1555G mutation in the mitochondrial gene MTRNR1 can cause hearing loss in patients exposed to aminoglycoside antibiotics.
      2. Clinical testing is offered in 22 labs.
      3. Prevention of hearing loss in maternal relatives is achievable.
      4. Every patient with NSHL should be screened – unless maternal inheritance can be excluded.
   I. Pharmacogenomic biomarkers that help predict drug-related toxic effects. Pharmacogenomics has entered day-to-day clinical practice. Today, about 10% of labels for FDA-approved drugs contain pharmacogenomic information.

XII. Ethical, legal, social implications (ELSI).
   A. Informed consent
   B. Confidentiality/privacy
   C. Discrimination (e.g., employers, insurers)
   D. Psychological impact
   E. Fairness in access to genetic services
   F. Conceptual/philosophical/religious implications
   G. Clinical issues including education of clinicians, patients, & the general public

XIII. Why the genomics revolution is important for optometric practice.
   A. Virtually all diseases have a genetic component. This means optometric physicians will need to:
      1. raise genetic hypotheses with every patient (i.e., look at each patient through a “genetic lens”)
      2. realize when genetic factors play a role in a patient. Thus, we will have to be aware of genetic contributions to the common diseases seen in practice
      3. improve family history taking skills and, in selective cases, draw a three-generation pedigree
      4. be able to identify patients who need genetic services and/or referral
      5. know how, when, and where to obtain advice and refer patients
         a. Find a genetic counselor in your area by using the Website of the National Society of Genetic Counselors (http://www.nsgc.org, click on “Find a Genetic Counselor”).
         b. Find a medical geneticist in your area by using the Website of the American College of Medical Genetics (ACMG) (http://www.acmg.net, click on “Find a member”).
         c. Find genetic services in your area by using the ACMG Website (http://www.acmg.net, click on “Find genetic services”)
         d. Identify patient support groups for specific conditions by using the Website of the Genetic Alliance (http://www.geneticalliance.org; http://www.diseaseinfosearch.org/Glaucoma/3065).
      6. know how to access new knowledge on medical genetics and use it in patient care
   B. Everyone has 5-50 significant genetic flaws.
   C. There is an exponential rise in genetic knowledge.
   D. New diagnostic/prognostic/treatment options are increasing. This means optometric physicians will need to:
1. Improve family history taking skills, and in selected cases draw a 3-generation pedigree.
2. Identify individuals who may benefit from genetic services. These individuals include those with a genetic disorder, as well as those at increased risk for having or transmitting a genetic disorder.
3. Recognize the historical and physical features of common genetic conditions
4. Learn the indications for genetic testing and the availability of genetic tests for specific ocular conditions
5. Monitor the health of individuals with a genetic disorder. Optometrists in collaboration with appropriate subspecialists work to monitor the health of patients with a genetic disorder or those with an increased risk for having a genetic disorder.
6. Provide basic genetics information to patients and families. By providing this information, optometrists are able to help their patient’s understanding and informed decision making.
7. Possess knowledge of how to access the full range of genetics services from which patients might benefit
8. Appropriately refer patients. Optometrists are in the position of knowing which patients require referral and are able to refer appropriately.
9. Use genetics to individualize patient care and preserve health
10. Be able to read the increasing number of ophthalmic journal articles on genetic/genomic issues, and thus to keep current
11. Know how to access new knowledge on medical genetics and use it in patient care
12. Be able to identify, understand, and address the ethical, legal, social, and financial issues associated with genetic conditions as they arise in primary and specialty (e.g., pediatric optometry and low vision) practice.

E. There are not enough genetic specialists to handle the increase in demand.
F. There is a growing demand for genetic information and services by patients. Thus, optometrists will need to be able to:
   1. address the increasing number of patient’s questions and concerns about new genetic technologies and information related to eye care
   2. explain basic genetic concepts

G. Direct marketing of genetic tests to health care professionals & potential customers is occurring.
1. 23andme.com – For $99 they will take a saliva sample and genotype ~1,000,000 of your SNPs. Their report will give you your risk for 240+ health conditions & traits (including AMD) as well as trace your ancestry. This has been discontinued because of a FDA warning letter. They are offering only ancestry information now.
2. Counsyl – For $599 (or ≤ $99 with insurance) your PCP will take a blood sample, send it to this lab and it will check for 458 causal genetic variants for 105 monogenic recessive disorders.
3. cyGene Direct’s Glaucoma & Macular Degeneration DNA Analysis ($99.95).
4. Nutrigenetic testing. Some companies say they can use genetic information to develop an individualized diet plan. They often recommend costly dietary supplements from their Website.
   a. Such tests may be misleading or even harmful. They tend to make claims that can’t be proven scientifically.
   b. After the “nutrigenetic testing,” some of the companies have provided the following recommendation for their diet: “Eat more fruits and vegetables. Reduce intake of sugar, starch, red meat, and trans and saturated fats.” A genotype analysis for that diet recommendation is not necessary.
   c. Furthermore, almost no clinical studies have been done to ascertain the efficacy of diet recommendations based on genetic variants.
   d. There is ongoing debate about the regulation and use of such tests.
5. Direct-to-consumer (DTC) genetic testing has raised concerns about:
   a. The potential for inadequate pretest decision-making
   b. Misunderstanding of test results
   c. Access to tests of questionable clinical value
   d. Lack of necessary genetic counseling & follow-up
   e. Unexpected additional responsibilities for primary care providers.
   f. Different risk assessments for samples from the same individual sent to different companies
   g. Testing that is unregulated & unvalidated
   h. Testing often done without a physician’s intercession.
6. The **FDA is planning to regulate DTC testing**. In June 2010, the FDA sent letters to 5 of the major personal genomics companies saying that their products are considered diagnostic medical devices that have not been approved yet. In July 2010, the FDA sent similar letters to 14 other DTC companies. The message was that the companies’ genetic services met the classification of a device that required FDA approval as medical devices. Thus, the DTC business is in a state of flux.
   a. One of the DTC labs, 23andme, has applied for FDA clearance.
   b. DeCODEme DTC testing has been discontinued since the company was acquired by Amgen.
   c. Navigenics has been acquired by Life Technologies and is no longer accepting orders.

H. **National Ophthalmic Disease Genotyping Network (eyeGENE)**.
1. Created by the National Eye Institute (NEI) in partnership with 12 CLIA-approved laboratories
3. Submit the patient sample (blood draw & shipping are the only costs).
4. Currently, over 100 genes for 34 diseases are assessed.
5. You receive the report.
6. The referring OD must ensure that the patient receives genetic counseling before & after.
7. **eyeGENE goals**:
   a. To facilitate translational eye research by:
      • Providing accurate molecular diagnostic genotyping for persons with heritable eye disorders
      • Maintaining a repository of DNA coupled to accurate phenotypic data for future research and discovery
      • Establishing standardized clinical phenotypic descriptors, especially for complex ocular disorders
      • Developing a shared, open source database of genotype/phenotype information for disease research and future trials of therapeutic interventions
   b. To identify and engage patients in clinical trials designed to diagnose, manage, treat, and prevent genetic eye diseases.
   c. To promote collaborations between clinicians and researchers for the benefit of the American public.
   d. To enhance professional and public awareness and understanding of the genetic basis of ophthalmic disorders and inform of the availability and the value of diagnostic genetics for ophthalmic care.

I. **Enhanced genetic literacy & competency of ODs is necessary.**

J. **What you can do now.**
1. **Start learning about genetics & eye diseases** (see references below).
   a. These competencies have been endorsed by the American Academy of Optometry, the American Optometric Association, & the Association of Schools & Colleges of Optometry.
3. **Become familiar with: Online Mendelian Inheritance in Man (OMIM).**
   a. Catalogues all known diseases with a genetic component. It has come to be known as the “bible” for genetics.
   b. Assigns a 6-digit number to every gene & disease (the MIM code)
   c. The 1st number indicates the type of inheritance
   d. Check out the tutorial and training materials by Open Helix at: [http://www.openhelix.com/cgi/tutorialInfo.cgi?id=125](http://www.openhelix.com/cgi/tutorialInfo.cgi?id=125) This includes a short (~27 min) video tutorial at [http://www.openhelix.com/cgi/showMovie.cgi?id=125](http://www.openhelix.com/cgi/showMovie.cgi?id=125)
4. **Identify patients that could benefit from gene testing.**
   a. Many disease genes are available for testing now.
   b. An **excellent resource** is **GTR: Genetic Testing Registry**. ([http://www.ncbi.nlm.nih.gov/gtr/](http://www.ncbi.nlm.nih.gov/gtr/)). This is a free online resource that provides centralized access to comprehensive genetic test information that is voluntarily submitted by test providers. The GTR covers tests for heritable mutations, including pharmacogenetic tests and tests using complex arrays and multiplex panels. The GTR provides a wide range of information such as the purpose of the test and its limitations; whether it is a clinical or research test; the testing method and what the test measures; analytical validity information, and evidence of clinical validity and clinical utility; as well as the name, location, and credentials of laboratories providing the test. The web site links to context-specific information
about conditions, genes, test standards, practice guidelines, and consumer support sites. The primary audience of the initial phase of GTR is the health care community. On this website, you can:

1) Find tests specific to a condition or drug response
2) Find a laboratory
3) Find GeneReviews: these are expert-authored, peer-reviewed disease descriptions that apply genetic testing to the diagnosis, management, and genetic counseling of patients and families with specific inherited conditions.
4) You can go to Youtube for a short (3 min 11 s) tutorial about the “GTR: Home page and Basic Search Functions” (http://www.youtube.com/watch?v=VmDo1DjOyBY&list=PL1C4A2AFF811F6F0B&index=1)
5) You can also go to Youtube to see how to “Locate a Test in Under Three Minutes” (http://www.youtube.com/watch?v=HeS4Jvdy194&list=PL1C4A2AFF811F6F0B&index=2)

5. Identify patients for studies & clinical trials.
   a. eyeGENE – become familiar with this resource, register for it, and start sending blood samples of patients who have genetic diseases that it addresses
   b. ClinicalTrials.gov (http://clinicaltrials.gov/) is a registry of federally and privately supported clinical trials conducted in the US and around the world. It gives you information about a trial's purpose, who may participate, locations, and phone numbers for more details.
   c. PubMed comprises more than 22 million citations for biomedical literature from MEDLINE, life science journals, and online books. Citations may include links to full-text content from PubMed Central and publisher web sites. Articles pertinent to your patient will give information about where the research was done and who was involved.

K. Final caveat.
   1. The coming deluge.
      a. The $1000 genome is here. The cost will continue to go down further.
      b. We will be awash with new highly complex genetic information.
      c. It will often be touted as ready to revolutionize healthcare.
      d. It does hold great promise, especially for drug development and laying bare the molecular foundation of disease.
      e. But the rules of medicine haven't changed.
         1) There’s nothing magical about genetic information.
         2) Take a good family history!
         3) Insist on actual outcome data before embracing attractive ideas.
   2. The need for clinical outcome data.
      a. The history of medicine is riddled with the corpses of good ideas that didn’t pan out:
         1) Extracranial – Intracranial (EC-IC) bypass to prevent stroke
         2) Hormone Replacement Therapy (HRT) to prevent every possible bad outcome of female aging
         3) Prostate-specific antigen (PSA) testing?
      b. Good ideas are not enough to guide medical care.
         1) We have the power to harm.
         2) Even through "non-invasive" testing
         3) Such information has the potential to put our patients on a trajectory that leads to dangerous and harmful interventions.
      c. We need to insist on data to prove that our good ideas actually result in improved outcomes.
      d. We have to be leery of shortcuts and attractive theories.

XIV. Selected References.

A. Books.


B. Journal articles.


C. Selected Websites

1. Disease specific information for health care professionals.
   a. OMIM (Online Mendelian Inheritance in Man):
   c. GeneReviews (http://www.ncbi.nlm.nih.gov/books/NBK1116/)
   e. RetNet (Retinal Information Network): http://www.sph.uth.tmc.edu/Retnet/

2. Genetic tests.
   a. GeneTests ( http://www.genetests.org/)
   c. American College of Medical Genetics (select "Find Genetic Services”):
      http://www.acmg.net//AM/Template.cfm?Section=Home3
   e. The John & Marcia Carver Nonprofit Genetic Testing Lab: https://www.carverlab.org/

3. Family history tools.
   b. Centers for Disease Control & Prevention:
      http://www.cdc.gov/genomics/famhistory/famhist.htm

4. Directories of clinical genetics specialists.
   a. National Society of Genetic Counselors (select “Find a Genetic Counselor):
      http://www.nsgc.org/
   b. American College of Medical Genetics (select " Find a Member”):
      http://www.acmg.net//AM/Template.cfm?Section=Home3

5. General genetics resources.
   a. National Human Genome Research Institute: http://genome.gov/Education/
   b. Genetic Science Learning Center: http://learn.genetics.utah.edu/
   c. Genetics Education Center: http://www.kumc.edu/gec/
   d. March of Dimes – Genetics & Your Practice:
      http://www.marchofdimes.com/gyponline/index.bm2
   e. National Coalition for Health Professional Education in Genetics (NCHPEG):
      http://www.nchpeg.org