Genetics of Posterior Segment Disease

Sherry J. Bass, OD, Jerome Sherman, OD, Jerry Rapp, PhD

Genotype-Phenotype Correlations in Ocular Disease: Genomic Implications for Patient Care

I. Basic considerations
   A. Ocular vs. systemic disease
      1. single nucleotide polymorphism (SNP)
   B. Genetic counseling
   C. Pharmacogenomics
   D. Gene therapy
   E. eyeGENE

II. Review of basic protein biochemistry (proteomics) and molecular genetics (genomics)
   A. 20 naturally occurring amino acids
      1. nonpolar
      2. polar
      3. aromatic
      4. acidic (negatively charged)
      5. basic (positively charged)
   B. DNA codon
      1. Four different bases – adenine (A), thymine (T), guanine (G), cytosine (C)
   C. Mutations
      1. missense
         a) substitution – conservative vs. non-conservative
      2. deletion
      3. insertion
   D. Protein structure
      1. primary
      2. secondary
      3. tertiary
         a) covalent disulfide bond
         b) electrostatic interactions
         c) hydrogen bonds

III. Role of Rhodopsin (R) in Visual Excitation
   A. Microanatomical relationship to rod cell
   B. Structure
      1. Chromophore: 11-cis retinal (vitamin A)
      2. Molecular genetic sequence of opsin
      3. Chromophore binding site
      4. Heptahelical, transmembrane G-protein coupled receptor (GPCR)
      5. Sodium (Na⁺) dark current
   C. Effects of illumination
1. 11-cis → all-trans retinal
2. Denaturation of opsin

D. Molecular mechanism of visual neural impulse generation
   1. Formation of photoactivated R (R*)
   2. Interaction with transducin (T)
   3. Activation of phosphodiesterase (PDE)
   4. Breakdown of cyclic GMP (cGMP)
   5. Hyperpolarization of rod outer segment (ROS) membrane

IV. Rhodopsin Mutations in Autosomal Dominant Retinitis Pigmentosa (adRP)
   A. 3 major types of RP
      1. autosomal dominant
      2. autosomal recessive
      3. X-linked recessive
   B. Types of mutations
      1. Missense, deletion and insertion
      2. Intradiscal, transmembrane and cytoplasmic
      3. Specific residues involved
         a) LYS 296 and GLU (glutamate) 113
         b) CYS (cysteine) 187 and CYS 110
         c) PRO (proline)

V. Clinical expression of disease
   A. Genotype vs. phenotype
      1. correlation of clinical expression with location of mutation in R
         a) cytoplasmic vs. intradiscal
      2. correlation of clinical expression with type of mutation in R
         a) case history
            i) ASP 190 ASN
      3. Genetic counseling

VI. Age-related macular degeneration (AMD)
   A. Multifactorial disease – ocular, demographic, environmental, behavioral and
      genetic factors.
   B. Genetics
      1. Complement pathway
   C. Nutritional intervention
      1. Possible mechanisms
      2. AREDS Report No. 8
      3. Possible adverse effects
      4. AREDS 2
   D. Prediction models
      1. Genetic, demographic and environmental variables
   E. Nutrigenomics - genetic polymorphisms re: nutritional intervention
VII. The Dawn of Pharmaco-Genetics in AMD

a. Rationale for Pharmacogenetics
   i. AMD is the most common etiology of vision loss in the US. The AREDS 1 and 2 studies confirm that anti-oxidants and zinc can reduce the risk of progressing from Category 3 AMD to more serious vision loss by about 25%.
   ii. Based upon a landmark study published in Aug 2013 in the journal Ophthalmology, the presence or absence of high risk alleles in CFH and ARMS2 genes determines the most effective nutraceutical for each patient.

b. Introduction
   i. What is AMD and how common is AMD?
   ii. Age related? Race related?

c. Genetic and environmental considerations

d. Diagnostic Modalities
   i. Standard Fundus Exam
   ii. SD OCT
   iii. Fluorescein angiography (FA)
   iv. Fundus Autofluorescence (FAF)
   v. Multispectral Imaging
   vi. Microperimetry
   vii. Pattern ERGs (pERGS) and VEPs
   viii. Preferential hyperacuity perimetry (PHP) and Home PHP

e. Genetics of AMD
   i. 12 known genes define nearly all of AMD genetic risk
   ii. Genetics can now predict the 2,5, and 10 year risk of marked vision loss
   iii. Genetic risk can be stratified into one of five categories
      1. Determines type and frequency of follow-up
      2. Low risk patients (e.g.category 1)
         a. Have only about 3% chance of vision loss
         i. Can be followed annually
      3. High risk patients (e.g.category 5)
         a. Have greater than 70% chance of significant vision loss
i. Should be followed more aggressively-every 3-4 months

4. Best estimate: Genetic and epigenetic considerations
   a. Smoking history plays a role
   b. Systemic diseases play a role
      i. Obesity
      ii. High cholesterol
      iii. Alzheimer’s and AMD?

5. Significance of peripheral drusen

f. Nutritional considerations
   i. Carotenoids
      1. Lutein
      2. Zeaxanthin
      3. Meso-zeaxanthin
   ii. Omega 3’s

g. AREDS 1 Results and Implications

h. AREDS 2 Results and Implications
   i. Continue to use 80 mg of zinc but beta carotene should be replaced by lutein and zeaxanthin
   ii. Omega 3’s not of help in the overall group but did show an effect in the lowest quartile

i. What are the appropriate nutraceuticals for each patient?
   i. Landmark Study: Ophthalmology September 2013-based on 12 year follow-up of 995 subjects from AREDS 1 with intermediate AMD in one eye
      1. Patients with high risk complement factor H (CFH) alleles should not be taking zinc
         a. Zinc may have a deleterious effect on such patients and may increase risk of progressing to advanced AMD
      2. Patients with high risk age-related macular degeneration (ARMS2) alleles should not be on beta carotene
      3. Lutein, zeaxanthin and meso-zeaxanthin were not included in the original AREDS study hence no 12 year follow-up

   ii. Implications
      1. Genetics determines what nutraceutical is best for each intermediate AMD patient.
      2. One formulation is not good for all
B. The Evolving Standard of Care in AMD
   a. ODs are recently getting sued for poor detection of wet and dry AMD
   b. Consider risk factors to provide better care as we have been doing in glaucoma for years
C. Treatment of Stage 3 Intermediate AMD
   a. Cessation of smoking
   b. Weight loss
   c. Improved diet
   d. Appropriate neutraceutical based upon genetic identification of high risk alleles
   e. Less blue and UV light
   f. Home Amsler grid or home PHP for detection of early wet AMD
      i. If wet AMD develops, referral for treatment ASAP
      ii. Careful follow-up
      iii. Consider changing anti-VEGF
         1. E.g. if Lucentis or Avastin are not working, Eyelea should be tried.
      iv. Genetics may predict which patients will respond better to specific anti-VEGF treatments
D. Future Considerations

VIII. Genetics of Orphan Retinal Diseases
   a. Retinitis Pigmentosa
      i. Prevalence
      ii. Clinical characteristics
      iii. Symptoms
      iv. Types
      v. Diagnostic testing
         1. SD-OCT
         2. RNFL assessment: RNFL is above normal thickness except in later stages of the disease
         3. ERG
         4. Visual fields
      vi. Fundus Autofluorescence
      vii. Visual Fields
      viii. Hereditary patterns of RP
      ix. Differential from cone-rod dystrophies
      x. Genetics
         1. Dominant RP Genes
         2. Recessive
         3. X-linked RP
            a. RPGR gene (75%)
            b. RP2 (25%)
xi. Treatment
1. Vitamin A and lutein
   i. Not evidence-based
2. Clinical trials
   i. Valproic acid for adRP
   ii. Gene therapy for arRP caused by MERTK mutations
   iii. Brimonidine
   iv. Enacpsulated Cell Technology delivering Neurotrophic Growth factor in early RP
   v. Alga (9-cis rich powder) for adRP
   vi. Bone marrow transplantation for RP and cone rod dystrophy
2. No FDA approved medical treatment for the disease itself
   a. Treatment is available and effective for CME secondary to RP in some patients
      i. Oral methazolamide
      ii. Topical dorzolamide not as effective
3. Retinal prosthesis” Second sight Argus II
b. Leber Congenital Amaurosis
   i. Prevalence
   ii. Clinical characteristics
      1. Reduced VA, nystagmus from birth
c. Diagnostic tests
   i. SD-OCT: absence of PIL and RPE thinning throughout scan
   ii. Fundus hypoautofluorescence
   iii. Flat ERG-scotopic and photopic
d. Genetics
   i. Value of molecular genetic testing in the diagnosis of LCA
   ii. Treatment: Clinical Trials
   iii. Gene therapy for RPE65
      1. QLT091001 (oral retinoid) for RPE 65 and LRAT mutations
II. Stargardt Disease/ Fundus Flavimaculatus
i. Prevalence
ii. Clinical characteristics (may include one or more)
iii. Symptoms
iv. Diagnostic testing
   1. ERG
      a. Three types of responses
   2. SD-OCT
   3. Visual Fields
   4. Fundus autofluorescence imaging
      a. Central zone of hypoautofluorescence
b. Hyperautofluorescence of fundus flavimaculatus flecks if present

v. Hereditary patterns
   1. AR
   2. AD: Stargardt-Like Dystrophy (ELOVL mutations)

vi. Differential Diagnosis from other White Dot Diseases

vii. Genetics
   1. Known \textit{ABCA-4} mutations on short arm of chromosome 1 account for 50% of cases

viii. Treatment
   1. ALK-001
   2. Stem cell therapy

b. Other \textit{ABCA-4} Dystrophies
   i. Cone and Cone-rod dystrophy

III. Other Hereditary Retinal Dystrophies

a. X-Linked Juvenile Retinoschisis
   i. Clinical Characteristics
      1. Foveal schisis of inner retinal layers
      2. Reduced VA at birth
      3. Inferotemporal schisis (50%)
   ii. Hereditary pattern
      1. Sex-linked
      2. Males affected
   iii. Diagnostic testing
      1. SD-OCT: demonstrates macular schisis and thickened fovea
      2. ERG: Electronegative (normal “a” wave, but abnormal or no “b” wave), because the photoreceptors are intact “a” wave
   iv. Treatment
      a. Dorzolamide 2% TID for 3-5 months may reduce foveal thickness and can improve VA
   v. Genetics
      1. RS1 gene mutations-affect the retinoschisin protein that provides the retinal “glue” to keep the inner retinal layers together

b. Best’s Vitelliform Dystrophy
   i. Clinical characteristics
      1. Lipofuscin disorder of the RPE
      2. Egg-yolk macular lesion early on with good VA
      3. Scrambled-egg lesion with reduced VA
         a. CNV development
         b. Monitor and treat the CNV
ii. Diagnostic testing
   1. SD-OCT: hyperreflective lipofuscin in outer retina elevates the macula
   2. EOG: Abnormal Arden ratio: greatest light peak response over the lowest dark trough response (ratio=less than 1.6)

iii. Hereditary pattern- mostly AD
iv. Differential from other lipofuscin vitelliform maculopathies
   1. Adult-onset vitelliform dystrophy
v. Genetics
   1. Bestrophin gene mutations

c. Importance of Genetic Testing in Diagnosing Hereditary Retinal Disease
   i. Pedigree construction
      1. Determine pattern of inheritance in single gene diseases
      2. Ask about consanguinity or close geographic relation
   ii. Genetic testing
      1. Can be costly
      2. Ethical considerations
         *Ophthalmology* 2012;119: 2408-2410
   4. Available sites:
      a. **Commercial**: Patient or insurance pays but results are faster-usually 3 months
         i. [www.carverlab.org](http://www.carverlab.org)
      b. **Research**: No fee for testing but results can take a year or more
         i. EyeGene: [www.nih.nei.gov/Eyegene](http://www.nih.nei.gov/Eyegene)
   5. Reasons for Ordering Genetic Testing
      a. Confirmation of a diagnosis when the clinical picture is not clear
      b. Identification of specific mutations to determine eligibility for future clinical trials
      c. Targeting of specific mutations by future therapies
   d. Finding a Clinical Trial to Investigate On-Going Research
      i. [www.clinicaltrials.gov](http://www.clinicaltrials.gov)
         1. Enter the ocular disease in the search box to find NIH trials that are recruiting patients
      ii. [www.Foundationfightingblindness.org](http://www.Foundationfightingblindness.org)
   e. Obtaining information about an Orphan disease and updates in clinical trials and available treatments, contact Orphanet
      i. [www.orpha.net](http://www.orpha.net)
II. The Role of the Optometrist as Genetic Counselor
   a. Knowledge
      i. In addition to hereditary retinal and optic nerve disease, approximately 2300 genetic diseases have ocular involvement
   b. Information
      i. Reporting results to the family
   c. Risk Assessment
      i. Disease development in future progeny
      ii. Risk of progression based on the polymorphism
   d. Support
      i. Assistance in patient education about the disease
      ii. Determining eligibility for interested patients in clinical trials for future treatment