

MIT OpenCourseWare
<http://ocw.mit.edu>

HST.161 Molecular Biology and Genetics in Modern Medicine
Fall 2007

For information about citing these materials or our Terms of Use, visit: <http://ocw.mit.edu/terms>.

Question 1

Based on what you learned in our clinic on Rett syndrome, address the following questions.

a. What enzymatic function is missing in patient J. Explain clearly what the substrate and product of the enzymatic reaction catalyzed by this enzyme are.

Loss of function mutations in the enzyme MeCP2 are responsible for Rett syndrome. Methyl CpG binding protein 2 (MeCP2) binds selectively to methylated CpG sites and recruits the transcriptional silencer Sin3A and histone deacetylase.

b. What data supports the view that if a method for correcting the enzyme deficiency were to be developed, it is likely that patient J will undergo considerable clinical improvement.

A study showing that in a mouse model for Rett syndrome, reactivation of expression of MeCP2 in mice already expressing significant pathology restored the mice to a near normal condition.

Question 2.

Based on what you learned in our clinic on phenylketonuria, address the following questions.

a. Explain how patient M's status affected status for PKU was originally identified. Be explicit in your answer describing an initial screening test and a confirmation test that was carried out and describe the rationale for each test used in identifying her as affected with PKU.

M's PKU status was originally identified by mass screening of a dried blood sample taken on a piece of filter paper from a heel prick just after she was born. Her PKU status was confirmed by blood sampling and direct testing of blood phenylalanine levels.

b. Explain how patient M is currently being treated to address the difficulties associated with her PKU status and why this treatment is effective.

M has been placed on a diet which carefully controls her phenylalanine uptake. She receives just enough phenylalanine to allow proper growth but her levels of phenylalanine are limited so no toxicity occurs from excess phenylalanine.

c. When patient M reaches adulthood and chooses to have children of her own, what major concerns will she have related to her PKU status. How should these concerns be addressed?

When having a child of her own M will face two problems. First and foremost if her phenylalanine level is not very carefully controlled during pregnancy her fetus will be

subject to neurotoxicity even if the fetus has a normal copy of the PKU gene inherited from her father. M also faces an increased risk that her child may also have pku. She has a 100% chance of transmitting one PKU allele to her fetus and the child's father (depending on his ethnic background) may have a very significant chance of being a PKU carrier. If he is a carrier they have a 1 in 2 chance of having a child with PKU.

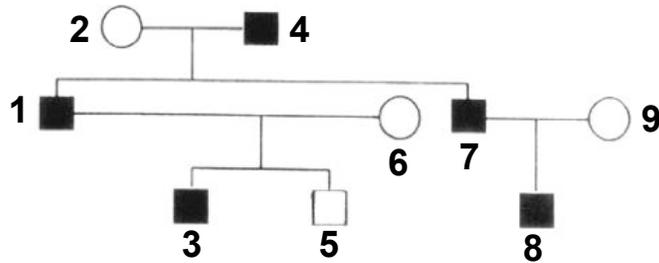
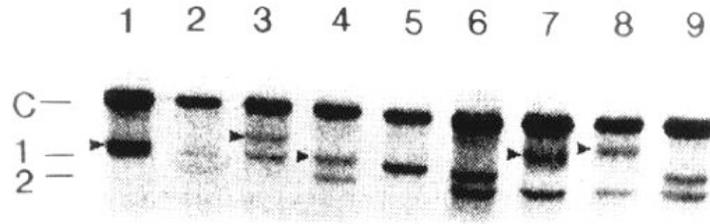
Question 3. The myotonic dystrophy gene was mapped to chromosome 19 through linkage to the secretor locus. A series of RFLP markers were then tested to determine the position of the gene on chromosome 19 with greater precision.

a. (2 points) Approximately how many informative meioses could allow localization of the gene to within a 1 centimorgan interval? Explain your answer.

Approximately 100 informative meioses would allow localization of the gene to a 1 cM interval.

b. (2 points) Explain how linkage disequilibrium could be helpful in localizing the gene more precisely. Include in your explanation a clear statement of the assumptions which must be correct in order for linkage disequilibrium to be useful in more precisely localizing the gene.

If most if not all of the myotonic dystrophy families under study have received their mutant allele of the myotonic dystrophy gene from a common ancestor, then an approach based on linkage disequilibrium would be helpful.



c. (2 points) A series of RFLPs were tested for their association with the myotonic dystrophy gene. The Southern blot shown above was carried out on a family with myotonic dystrophy. The numbers 1 and 2 indicate the positions of the two alternative alleles of an RFLP on chromosome 19 which were under examination for linkage to the myotonic dystrophy gene. What property of this RFLP is unusual in this Southern blot?

The upper band labeled 1 is changing position relative to the other bands in this Southern blot.

d. (2 points) What is the explanation at the level of DNA sequence for the unusual behavior of this RFLP?

This is because the CTG repeat sequence in the DMPK 3' UTR is expanding in length causing the band to be larger and larger.

e. (2 points) Explain how the data shown in this figure provides a physical explanation for the phenomenon of genetic anticipation. (Include in your answer a brief explanation of the term “genetic anticipation”.)

Genetic anticipation—each generation having a significant chance of receiving a gene which causes a more severe phenotype is the consequence of the expansion in length of the triplet repeat sequence as it passed from one generation to the next.

Question 4. (3 points) Anne Smith and Richard Jones were both born deaf, attended the same school for the deaf, married and had five children three boys and two girls. All of their children were born deaf. Neither Anne’s parents nor Richard’s parents were deaf. Based on these facts alone what modes of Mendelian inheritance would be compatible with Anne and Richard’s family? For each mode of inheritance comment on whether a new mutation could be relevant to explaining the pattern of inheritance in the family.

It is possible that Anne and Richard are each homozygous for the same recessive deafness gene. No new mutation required.

It is possible that either Anne or Richard are new mutations for a dominant deafness gene with 100% penetrance. It is possible that either Anne or Richard carrying existing mutations for a dominant deafness gene with less than 100% penetrance.

X linked recessive inheritance for a deafness gene is possible if both Richard and Anne were homozygous for this gene only if one of Anne’s copies of the gene (the one inherited from her father) is a new mutation (if this copy were not a new mutation her father would have been deaf.)

b. (3 points) Anne's sister Rachel, who was not deaf, met Richard's brother, Mark who also was not deaf while visiting at the school. Rachel and Mark married and had four children. Two of the children were born deaf, one boy and one girl. What is the ***most likely*** mode of inheritance for deafness in the family based on these new facts? Explain your answer.

Autosomal recessive. Rachel and Mark are both carriers of the same autosomal recessive deafness gene and have a 1 in 4 chance of having a child who is homozygous for this recessive deafness allele.

Question 5. (30 points) A screening test for colon cancer involves testing by PCR for the presence of cells which carry certain mutations in the Kras gene in stool. Explain the rationale for this test.

a. What types of mutations in the Kras gene is the test designed to detect? (3 points)

Gain of function mutations which lead to a constitutively active state for KRas

b. How could these mutations contribute to the etiology of a colon tumor? (3 points)

Activated oncogenes such as KRas1 allow cells to proliferate in the absence of normal signals to stimulate cell proliferation.

Three patients have positive results in this test and undergo colonoscopy.

Patient 1, 47 years of age, has hundreds of colonic polyps and a malignant tumor which is surgically removed.

c. What gene would you screen for mutation in the tumor and in the DNA of the patient? (3 points)

APC

d. Explain to the patient how mutations in this gene can contribute to her polyps and to the development of her cancer. (4 points)

You have inherited an inactivated copy of the APC gene from one of your parents. A second event in some of the cells of your colon leads to loss of function of the functional copy of the APC gene you have inherited and the initiation of the formation of a polyp. The dividing cells within each polyp are now potentially able to progress in a step by step manner with further genetic changes to form an adenoma, a carcinoma and eventually a metastatic colon cancer.

e. Explain to the patient the possible increased risk for colon cancer to other members of her family who carry the same mutation as she does. (3 points)

The risk that other family members who inherit this mutation in the APC gene will have colon cancer is extremely high approaching 100% if a person lives to age 70 or so. Often individuals who carry so much mutations have colon cancer at a quite early age in the 30s or 40s.

Patient 2, 52 years of age, has no polyps but does have a malignant tumor, When the tumor is removed it is shown to have microsatellite instability. The patient an only child also has three first cousins ages 47,51 and 53 who have recently been diagnosed with colon cancer with microsatellite instability.

f. Which class of genes would you test for mutation in the germline DNA of the patient in this case? (3 points)

Mismatch repair genes.

g. Explain to the patient how mutation in one of these genes could contribute to the development of his cancer. (3 points)

The mismatch repair genes are part of the system of the body for dealing with potentially mutagenic damage to DNA. In the absence of function of these genes cellular mutation rates increase which in turn leads to colon cancer.

h. If DNA samples can be obtained from the three first cousins before the mutation screening is carried out, explain how these samples can be used to narrow the search for the mutation in the gene likely to be contributing to the high frequency of occurrence of colon cancer in this family. (4 points)

A linkage study can be performed to determine which haplotype is shared among the family members inherited from a common ancestor for the DNA regions which include the mismatch repair genes. The chromosomal region shared among the family members is the one most likely to harbor the mutation.

Question 6. (6 points) Prader-Willi syndrome, which includes mental retardation and an inability to be satiated, is caused by the absence of expression of genes in a critical region of chromosome 15 which is subject to genomic imprinting. The maternal copies of these genes are not expressed. In a normal individual expression of the copies of these genes from the paternal chromosome 15 is necessary and sufficient to support normal development. Two men and two women each have Prader-Willi syndrome. The genotypes of the affected individuals, as well as their parents, have been determined at 7 different polymorphic sites on chromosome 15.

	♀	♂	♀	♂	♀	♂	♀	♂
Marker 1	1,3	1,1	1,1	2,4	1,2	2,2	2,2	1,3
Marker 2	1,2	2,4	1,2	1,3	3,4	1,3	1,4	1,4
Marker 3	1,3	2,4	2,3	1,3	2,3	2,3	3,5	2,3
Marker 4	2,4	1,3	2,4	5,7	5,5	1,4	1,2	4,6
Marker 5	5,5	4,5	1,3	2,4	2,3	1,3	6,7	1,2
Marker 6	3,4	1,2	1,6	3,5	1,1	2,2	2,8	2,5
Marker 7	1,4	1,3	2,4	1,6	2,3	4,5	4,5	1,3
	↓		↓		↓		↓	
	Man 1		Man 2		Woman 1		Woman 2	
Marker 1	1,3		1,4		2,2		2,2	
Marker 2	1,2		1,3		1,3		4,4	
Marker 3	1,3		1,3		3,3		3,3	
Marker 4	2,4		2,5		4,5		1,1	
Marker 5	5,5		1		2		7,7	
Marker 6	3,4		5,6		1		2,2	
Marker 7	1,4		1,2		3,4		4,4	

For each of these four individuals, (Man 1, Man2, Woman 1 and Woman 2) give an explanation of the chromosomal event which gave rise to their Prader-Willi syndrome. Please indicate, when possible, whether the relevant events occurred in meiosis I or meiosis II.

Man 1- Uniparental disomy resulting from non-disjunction in MI of mom.

Man 2- Deletion in paternal germline for region encompassing at least marker 5.

Woman 1- Deletion in paternal germline for region encompassing at least markers 5 and 6.

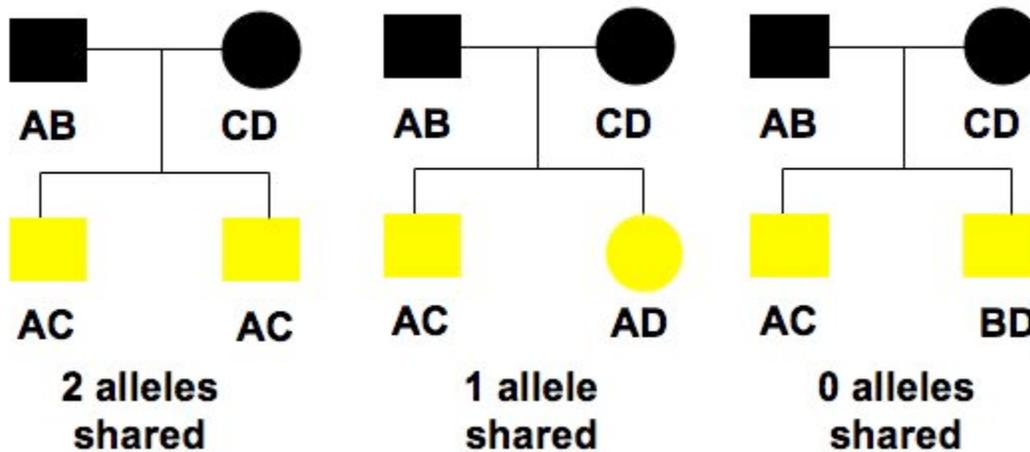
Woman 2- Uniparental disomy resulting from non-disjunction in MII of mom. paternal copy at the disease locus.

Question 7

The population frequency of Crohn's disease is approximately 0.3%. The likelihood of an individual having Crohn's disease is 20 fold higher for siblings than for the general population. Two strategies for a genome wide search to identify the chromosomal sites of genes contributing to Crohn's disease are under consideration. A and B described below. Two possible models for the genetic basis of susceptibility to Crohn's disease C and D are given below. Match a genome wide search strategy to a model for Crohn's disease susceptibility based on the likelihood that the genome wide search strategy is most likely to be effective given the model of the genetics of the disorder. Explain why the strategy you match to each model is most likely to yield meaningful results for that model.

Strategy A

Five hundred pairs of siblings affected both affected with Crohn's disease and their parents are collected. Highly polymorphic DNA markers spaced 10 cM apart spanning the genome are genotyped for each family grouping. Sibpairs are scored as to whether they match for two, one or zero alleles inherited from their parents as shown below. Deviations from a 25:50:25 ratio for the three classes expected by chance will be considered to be evidence of linkage between a susceptibility allele and the DNA marker.



Strategy B

2,500 individuals with Crohn's disease are identified and 2,500 individuals age and sex matched without Crohn's disease are collected as well. SNP polymorphisms approximately 100 kbp apart throughout the genome selected to be haplotype tagging SNPs are genotyped in both groups. A deviation in allele frequency between the cases and controls is taken as evidence that a gene predisposing to Crohn's disease is located in the vicinity of the marker.

Model A

Crohn's disease susceptibility is contributed to by alleles at five loci in the population. For each of these loci, a single major allele with an allele frequency of 20% in the population is responsible for susceptibility to Crohn's disease.

Strategy B would work well here and could give you very good resolution for the location of the Crohn's disease genes. Strategy A would also work to some degree but the chromosomal regions identified will be quite large.

Model B

Crohn's disease susceptibility is contributed to by alleles at five loci in the population. For each of these loci, there are one hundred alleles each with an allele frequency of 0.2% in the population which are responsible for susceptibility to Crohn's disease.

Only strategy A would work here.