

## Problem Set

### I. CFTR

You are an investigator at a crime scene and you are collecting evidence. One thing you have collected is a DNA sample left by the suspect who conveniently chewed their nails while waiting for an opportune moment. Another bit of evidence is a partially used inhaler for the drug Dornase Alfa. You begin to wonder what the drug is used for and if it is possible that the drug and the DNA sample are connected?

1. What disease is treated with the drug Dornase Alfa?
2. Here is a partial sequence of the mRNA from the nail clippings. The DNA has then been translated into mRNA to eliminate excess code. Do a BLASTn search on this sequence and see if it reflects a specific mutation which is common in a genetically inherited disease. Narrow the BLASTn search to RefSeq records and to homo sapiens.

```
attatgcctg gcaccattaa agaaaatatac attgggtg tttcctatga tgaatataga  
tacagaagcg tcatcaaagc atgccaacta gaagaggaca tctccaagtt tgcagagaaa  
gacaatatag ttcttggaga aggtggaatc aactgagtg gaggtcaacg agcaagaatt  
tctttagcaa gagcagtata caaagatgct gatttgtatt tattagactc tccttttggg
```

3. Describe the alignment between the search sequence and the sequence retrieved in the first hit by defining what nucleotides are missing? Does it seem that the search sequence would produce a complete Amino Acid sequence if it was translated?
4. What is the record identification number which reflects the best alignment with this sequence?
5. Go to the nucleotide record. Review the record. Note it contains multiple links to many journal articles. After the references is a Comment field which discusses the protein produced by the mRNA. What is the function of this protein?
6. What two diseases are described in the Comment field which are caused by mutations in this mRNA?
7. Does it seem possible that there is a link between the drug Dornase Alfa and a mutation in this mRNA sample?
8. How many nucleotides are found in the nonmutated CFTR mRNA?
9. Use the Links function to go to the OMIM database. Look for the record about the cystic fibrosis disease. Note that this is the first record retrieved. What chromosome is the CFTR gene on?
10. List a few research animals which might be used to study the disease?

### II. Tay Sachs

You are a genetic counselor working with a couple who are identified carriers of several genetic diseases. They have asked for invitro fertilization services to insure that their child would be free of these diseases.

Four eggs are harvested and fertilized. The resultant DNA of the 4 embryos are tested for the presence of the following diseases Canavan disease and Tay Sachs disease.

1. Here is a partial DNA sequence from one of the embryos. Do a BLASTn search on this sequence. Narrow the search to just mRNA curated RefSeq records and human.

```
ggcgagagga tattccagtg aactatatga aggagctgga actggtcacc aaggccggct  
tccgggccct tctctctgtatccc ccttggtacc tgaaccgtat atcctatggc cctgactgga  
aggatttcta cgtagtggaa ccctggcat ttgaaggtac cctgagcag aaggctctgg
```

2. Review the first retrieval of the BLASTn search. Look at the alignment between the sample and the first answer retrieved. How does the sample in the search sequence differ from the first answer retrieved sequence?
3. Click on the nucleotide database link for this record [\[gi|13128865|ref|NM\\_000520.2\]](#) What is the name of the gene which this mRNA sequence was coded from?
4. What mutation does the first article in the list of references describe? Click on the link to the MEDLINE abstract. Is this article available electronically in full text?

### **III. Hereditary Hemochromatosis**

You are a physician with a bustling clinical practice. An adult male in his 40's has come into your office with a concern over an increasing number of dark spots on his skin. The patient also has complained of fatigue, impotence, and stiffness in joints. Blood and urine tests indicated a high amount of iron. The doctor does a skin biopsy and confirms a case of hereditary hemochromatosis. This patient is also the father of several young children.

1. What is hereditary hemochromatosis?
2. Is this disease best described as autosomal dominant or autosomal recessive?
3. The physician has decided to test the children for hereditary hemochromatosis. Here is a sequence from the DNA of one of the children.

```
atccaagga gttcgaacct aaagacgtat tgccaatgg ggatgggacc taccagggt  
ggataacctt ggetgtaccc cctggggaag agcagagata tacgtaccag gtggagcacc  
caggcctgga tcagcccctc attgtgatct gggagccctc accgtctggc acctagtea
```

4. Do a BLASTn search on the sequence. Narrow the search down to mRNA RefSeq records and sequences from homo sapiens. How many retrievals have the same high score?
5. Click on the Nucleotide link retrieved for the first answer retrieved. ([\[gi|21040354|ref|NM\\_139010.1\]](#)) Look at the Definition field of the record. What variant number does this sequence represent? How many base pairs are in this mRNA sequence?
6. Use the Links option to move to the Gene record. Look in the Summary to find out what mineral this protein regulates.

7. Look through the PubMed links. Describe a mutation which is identified in the GeneRifs.

#### **IV. BRCA2**

A woman in her early thirties is being treated for breast cancer. She is concerned that her relatively young age and the fact that she has several close relatives with early onset breast cancer would indicate that she might have a genetic trait for this disease. Her physician has referred her to you, a genetic counselor. Since she has several daughters, and she wants to be sure she is not genetically predisposed to this disease.

The genetic counselor takes a DNA sample and examines sequence at a commonly known locus of the BRCA2 gene for mutations.

```
atatacctt gtgatgtag ttggaaact tcagatatat gtaaatgtag tataggggaag
cttcataagt cagtctcatc tgcaaataact tgtgggattt ttgacacagc aagggaanaa
tctgtccagg tatcagatgc ttattacaa aacgcaagac aagtgttttc tgaaatagaa
gatagacca agcaagtctt ttccaaagta ttgttaaaa gtaacgaaca ttcagaccag
```

Take this sequence and use the BLASTn search system. Narrow the search to human and RefSeq records.

1. Look at the alignment of the between your search sequence and the highest scoring retrieval. Is there evidence of missing or altered nucleotides?
2. Click on the nucleotide record of the highest scoring retrieval. What is the Gene name and the Definition for this mRNA record? How many base pairs are in this mRNA sequence?
3. Use the Links button to move to the OMIM. Remember to click on the BRCA2 record. ( Hint – the Record number is → +600185 - BREAST CANCER 2 GENE; BRCA2) Look at the OMIM record for this disease. What is the frequency of this mutation in Ashkenazi Jewish women?
4. Why do they describe the BRCA2 gene as a caretaker gene?

#### **V. Sickle Cell Disease**

The parents of a child with severe sickle cell anemia which died at an early age are interested in having another child. They also want to be sure their next child does not have sickle cell anemia. They have come to you for invitro fertilization services. You have obtained several eggs and fertilized them. The DNA from one of the embryos is below. You are examining this sequence for genetic mutations causing sickle cell anemia.

```
1 acatttgctt ctgacacaac tgtgttcact agcaacctca aacagacacc atgggtgcatc
61 tgactcctga ggagaagtct gccgttactg ccctgtgggg caaggtgaac gtggatgaag
121 ttgggtggtga ggccttgggc aggctgctgg tggcttacc ttggaccagc aggttctttg
181 agtccttttg ggatctgtcc actcctgatg ctgttatggg caaccctaag gtgaaggctc
241 atggcaagaa agtgctcggt gccttttagtg atggcctggc tcacctggac aacctcaagg
301 gcacctttgc cacactgagt gagctgcact gtgacaagct gcacgtggat cctgagaact
```

```
361 tcaggctcct gggcaacgtg ctggtctgtg tgctggccca tcactttggc aaagaattca
421 ccccaccagt gcaggctgcc taccagaaag tgggtggctgg tgtggctaata gacctggccc
481 acaagtatca ctaagctcgc tttcttgctg tccaatttct attaaaggtt cctttgttcc
541 ctaagtccaa ctactaaact ggggatatt atgaagggcc ttgagcatct ggattctgcc
601 taataaaaaa cttttttttt cattgc
```

1. Do a BLASTn search of this sample (you do not need to delete the line numbers) and limit it to RefSeq records and human. Look at the alignment between the search sequence and the first answer. Is there evidence of loss of nucleotides or alteration of nucleotides in the search sequence? Based on this evidence, does it seem that the DNA sample reflects a mutation for sickle cell anemia?
  2. Click on the best retrieval and go to the nucleotide record. What is the name of this gene and the definition?
  3. What is the Coding sequence for this gene?
  4. Use the Link feature to connect to the Entrez Gene. What chromosome is the HBB gene on?
  5. What is the description of the protein provided in Entrez Gene?
  6. Describe several of the phenotypes produced by mutations in the HBB protein.  
OMIM & BOOKS
  7. Click on the “LinkOut” in the Links option. Click on Genetic Home Reference. How does this resource differ from Entrez Gene? When might you use this resource?
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