INSTRUCTOR
SCHOOL
COURSE
TERM

Name_____

Course Section/Period_____

Exploring Genetic Characteristics of Diabetes Type 1

Objective/s:

Learn about Diabetes Type 1 (insulin-dependant) at the genetic level and practice accessing bioinformatics on the Internet. At the conclusion of this exercise, you should be able to do the following:

- Search for and locate genetic information about a specific human disease
- Use bioinformatics tools to discover detailed information about the structure and function of particular genes
- Use the *BLAST* database to find information about nucleotide sequence homology
- Explore genetic characteristics of Diabetes Type I

Background:

Diabetes results in an increase in glucose (sugar) in the blood. There are multiple genetic factors contributing to the progression of diabetes in an individual. Sugar provides the body with energy and is essential for humans to survive. Sugar is processed by a hormone (insulin) produced by the pancreas. In order for the body to function effectively, the body must have an appropriate balance between sugar and insulin; diabetes results when there is an imbalance between these two molecules in the bloodstream.

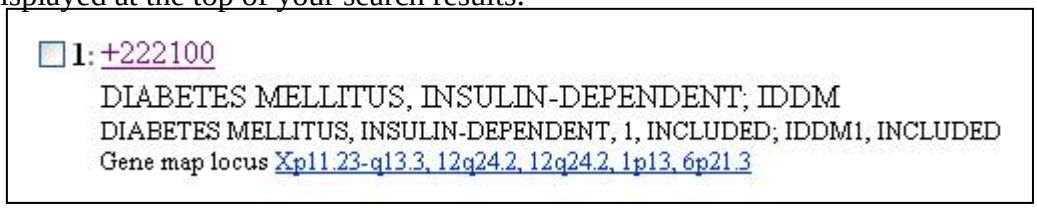
There are two types of diabetes that affect people's ability to process sugar – Type I and II. In Diabetes Type I, an individual may have to take insulin as a medication (insulin-dependant) because their pancreas is not producing enough insulin on its own. Without insulin, sugar may be present in the bloodstream but may not be available for cells to use as energy.

Directions to Complete Web Activity:

1. Using your Internet browser (mozilla, explorer, netscape, etc.) and your favorite search engine (google, yahoo, etc.), search "OMIM" to find the gene map website. What does OMIM stand for?

2. **Follow the link to the OMIM site**. The resulting page should look like this:

3. Search "OMIM" (select in the drop-down menu); **type in "Diabetes Type 1" in the 'for' box and click Go**. You should see something like the following displayed at the top of your search results:



   a) How many articles come up with this search?
   b) What is the name of the first article?
   c) What is the reference number of the first article (6-digit number)?

4. **Link to the first article**, read over it and fill in the following blanks based on the information on the web page:

   Description:

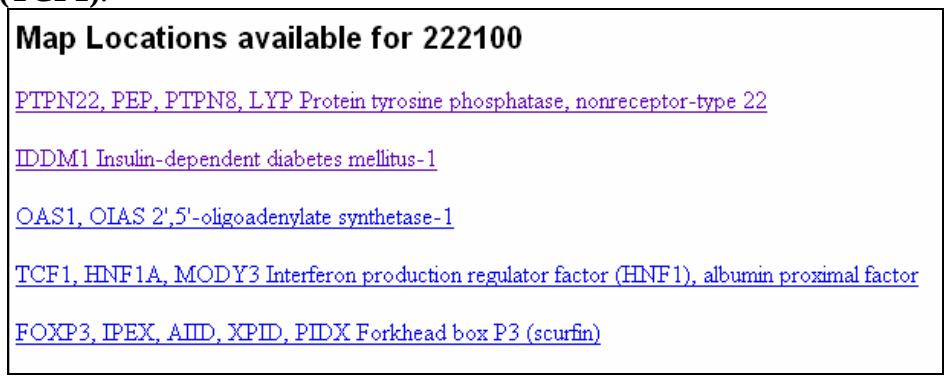
   Phenotype/Symptoms:

   Clinical Features:

5. Look at the **"Gene map locus"** at the top of the page. Which chromosomes play a role in Diabetes Type 1? (*Review chromosome notation)

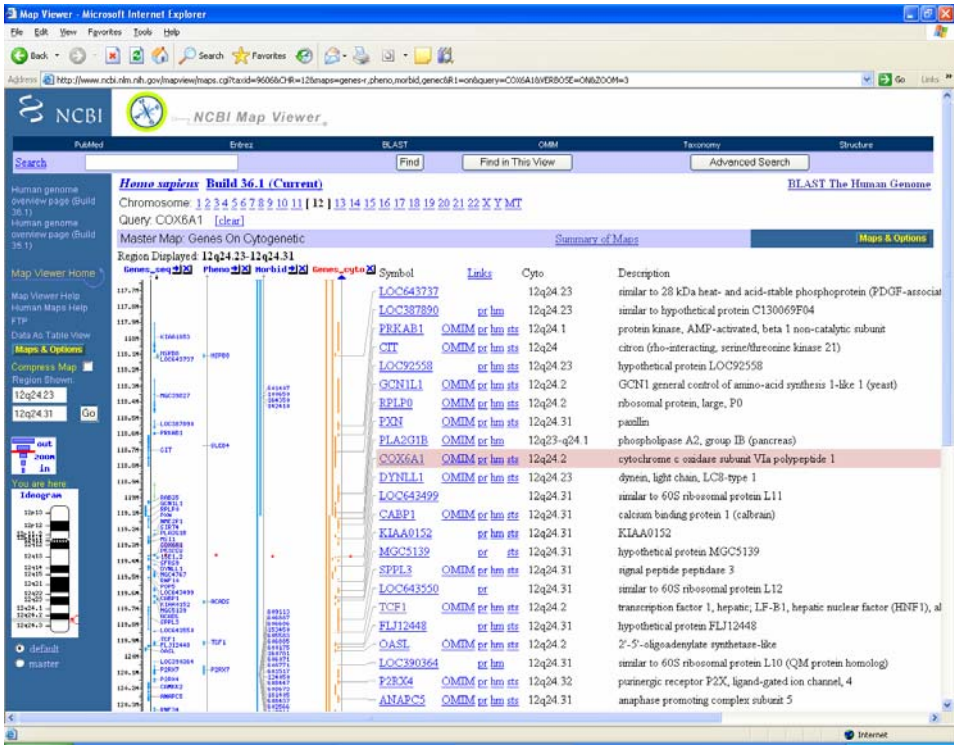
   _____ _____ _____ _____

For the rest of the exercises, we will concentrate on one of the chromosomes in the list above. Specifically, we will explore chromosome 12q.24.2 (the one that appears twice on the list). **Click on the 12q24.2 link and select the fourth location in the list below (TCF1)**.



Now link on the first 'location' (12q.24.2) to see the chromosome ideogram.

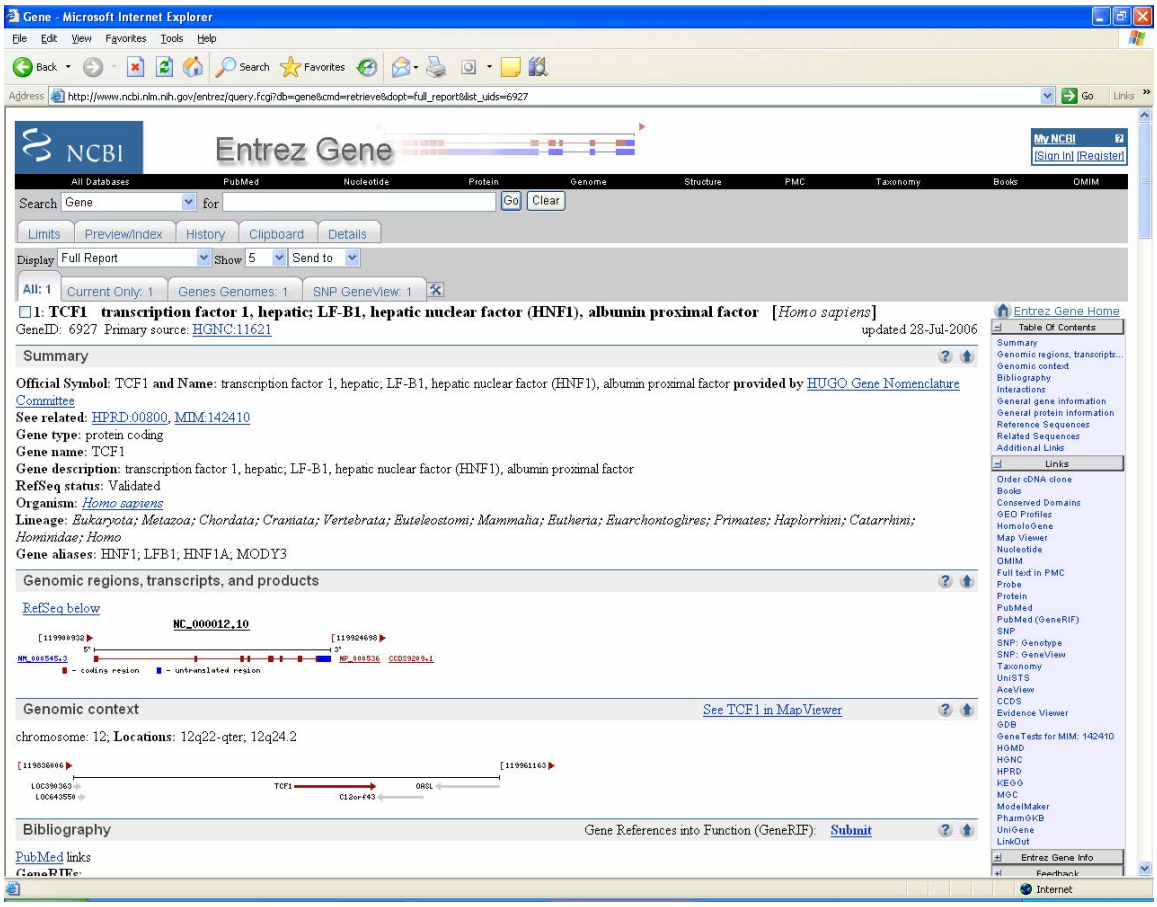
6. Take a look at the ideogram on the left side of the page. On which arm (long or short) of the chromosome does the highlighted gene appear?



7. Scan the column labeled (Cyto) for genes mapped on 12q24.2. Write down the names (from the Symbol column) of the genes mapped to this locus.

8. **Click on the link labeled TCF1**. What does TCF1 stand for?

The following page should now be displayed on your screen. Use the information on this page to answer questions 9 and 10:



9. Look in the section labeled "Genomic regions, transcripts, and products." The gene's reference sequence (RefSeq) is illustrated there. The chromosome number contains several digits, starting with the letters NC. What is the gene's chromosome number?

10. **Click on the number you just wrote down**; you will see a dropdown menu like the one shown below. **Select FASTA from this list**, and you will go to a page showing the nucleotide structure of the gene. At the top of this page is the NC number you just clicked on, and next to it a number in square brackets that begins with the letters gi. Write down this number (the gene index?) below:

11. Examine the nucleotide string shown on the page. In the space below, write down the first 21 nucleotide bases in the string.

12. For the nucleotide sequence you wrote above, list the complementary nucleotides in the space below:

13. For the nucleotide string in number 10, give the mRNA transcript below:

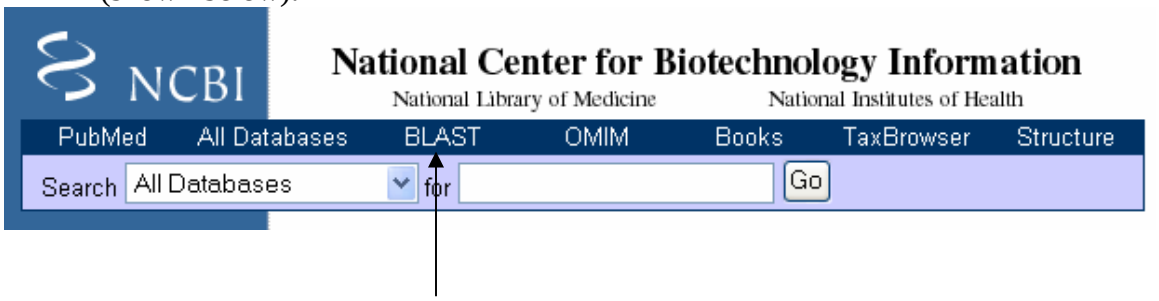
14. List the 7 amino acids (in order) described by the mRNA in number 12, using the chart below:

First letter	Second letter: U	Second letter: C	Second letter: A	Second letter: G	Third letter
U	UUU, UUC: Phe; UUA, UUG: Leu	UCU, UCC, UCA, UCG: Ser	UAU, UAC: Tyr; UAA: Stop; UAG: Stop	UGU, UGC: Cys; UGA: Stop; UGG: Trp	U C A G
C	CUU, CUC, CUA, CUG: Leu	CCU, CCC, CCA, CCG: Pro	CAU, CAC: His; CAA, CAG: Gln	CGU, CGC, CGA, CGG: Arg	U C A G
A	AUU, AUC, AUA: Ile; AUG: Met	ACU, ACC, ACA, ACG: Thr	AAU, AAC: Asn; AAA, AAG: Lys	AGU, AGC: Ser; AGA, AGG: Arg	U C A G
G	GUU, GUC, GUA, GUG: Val	GCU, GCC, GCA, GCG: Ala	GAU, GAC: Asp; GAA, GAG: Glu	GGU, GGC, GGA, GGG: Gly	U C A G

15. **Click Back** to return to the TCF1 page, the top of which is shown below:

16. **Scroll down to the Genomic regions, transcripts, and products section** and find the Nucleotide Messenger RNA number (begins with NM). Write this number below:

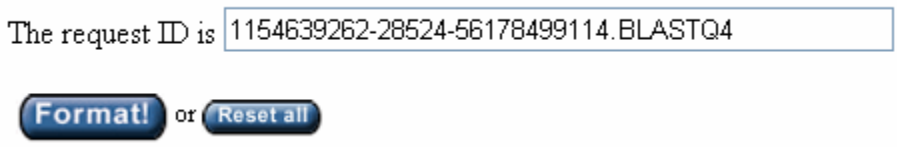
17. **Click on this link and choose FASTA**. You will see a nucleotide sequence display similar to the one you used for steps 10-14.
18. **Copy and paste the entire sequence (**beginning with the character **">"** – very important!) **into a Notepad file**.
19. **Save the Notepad file (named est1) to your desktop.**
20. **In your browser, scroll back to the top of the page and left-click on "NCBI"** to go to the NCBI (National Center for Biotechnology Information) home page (shown below)**.**



21. **In the blue toolbar select BLAST.**
22. **Choose "Nucleotide-nucleotide BLAST (blastn)"** from the list of BLAST programs**.**

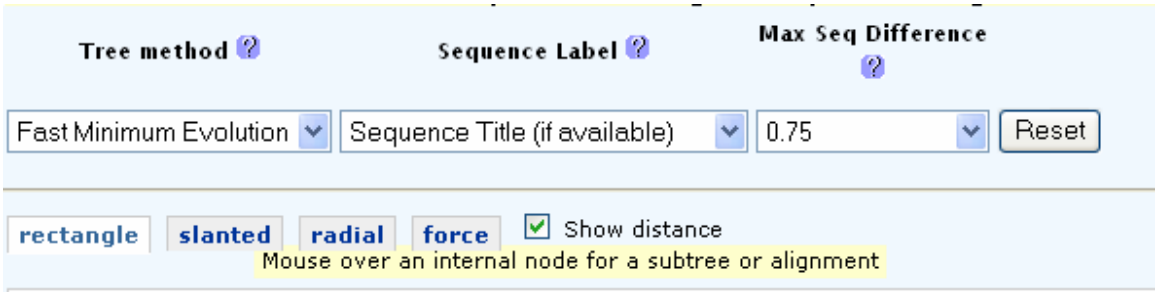
23. **Copy and paste entire text of your Notepad file into the Search box of the BLAST page** (shown below):

24. **Click the BLAST! button**. A new screen appears, a portion of which is shown below. **Click the FORMAT button**.



25. Use the information on the BLAST results page to answer the following questions:
    a) How many BLAST hits did you get?
    b) How many of these were close matches (score of 200 or higher)?
    c) How many very close matches (e-value of 0.0) were found?

26. **Click on the link labeled "Distance tree of results"** to view a phylogenetic tree of our selected gene. A portion of the top of the resulting window is shown below. **Click on the tab labeled "slanted"** for a clearer view of the tree and answer the questions below.



a) How many primate species are represented in the tree?
b) How many human (Homo sapiens) chromosomes are represented?
c) How many rodent chromosomes are represented?

Future plans for Curriculum:

- Have students this exercise using different genes/chromosomes related to Diabetes Type 1
- Have students explore genetic information on a disease of their choice using a similar procedure
- Have students run an ORF and explain what it means
- Exercises to look for ESTs
- BLAST exercises for nucleotides/proteins related to Diabetes Type 1 (or other disease)

PubMed references
(free, full-text articles in English using subject line "TCF1 and Diabetes Type I")

1: Pearson ER, Badman MK, Lockwood CR, Clark PM, Ellard S, Bingham C, Hattersley AT.
Contrasting diabetes phenotypes associated with hepatocyte nuclear factor-1alpha and -1beta mutations.
Diabetes Care. 2004 May;27(5):1102-7.
PMID: 15111528 [PubMed - indexed for MEDLINE]

2: McKinney J, Cao H, Behme MT, Mahon JL, Hegele RA.
Maturity-onset diabetes of the young (MODY) mutation in type 2 diabetes and latent autoimmune diabetes of the adult.
Diabetes Care. 2003 Dec;26(12):3358-9. No abstract available.
PMID: 14633836 [PubMed - indexed for MEDLINE]

3: Vilsboll T, Knop FK, Krarup T, Johansen A, Madsbad S, Larsen S, Hansen T, Pedersen O, Holst JJ.
The pathophysiology of diabetes involves a defective amplification of the late-phase insulin response to glucose by glucose-dependent insulinotropic polypeptide-regardless of etiology and phenotype.
J Clin Endocrinol Metab. 2003 Oct;88(10):4897-903.
PMID: 14557471 [PubMed - indexed for MEDLINE]

4: Tuomi T, Groop L.
Intrauterine hyperglycemia modifying the development of (monogenic) diabetes?
Diabetes Care. 2003 Apr;26(4):1295-6. Review. No abstract available.
PMID: 12663611 [PubMed - indexed for MEDLINE]

5: Bjorkhaug L, Sagen JV, Thorsby P, Sovik O, Molven A, Njolstad PR.
Hepatocyte nuclear factor-1 alpha gene mutations and diabetes in Norway.
J Clin Endocrinol Metab. 2003 Feb;88(2):920-31.
PMID: 12574234 [PubMed - indexed for MEDLINE]

6: Lambert AP, Ellard S, Allen LI, Gallen IW, Gillespie KM, Bingley PJ, Hattersley AT.
Identifying hepatic nuclear factor 1alpha mutations in children and young adults with a clinical diagnosis of type 1 diabetes.
Diabetes Care. 2003 Feb;26(2):333-7.
PMID: 12547858 [PubMed - indexed for MEDLINE]

7: Collet C, Ducorps M, Mayaudon H, Dupuy O, Ceppa F, Boutin P, Froguel P, Bauduceau B.
Prevalence of the missense mutation Gly574Ser in the hepatocyte nuclear factor-1alpha in Africans with diabetes.
Diabetes Metab. 2002 Feb;28(1):39-44.
PMID: 11938027 [PubMed - indexed for MEDLINE]

8: Bingham C, Ellard S, Nicholls AJ, Pennock CA, Allen J, James AJ, Satchell SC, Salzmann MB, Hattersley AT.
The generalized aminoaciduria seen in patients with hepatocyte nuclear factor-1alpha mutations is a feature of all patients with diabetes and is associated with glucosuria.
Diabetes. 2001 Sep;50(9):2047-52.
PMID: 11522670 [PubMed - indexed for MEDLINE]

9: Pontoglio M, Prie D, Cheret C, Doyen A, Leroy C, Froguel P, Velho G, Yaniv M, Friedlander G.
HNF1alpha controls renal glucose reabsorption in mouse and man.
EMBO Rep. 2000 Oct;1(4):359-65.
PMID: 11269503 [PubMed - indexed for MEDLINE]

10: Cha JY, Kim H, Kim KS, Hur MW, Ahn Y.
Identification of transacting factors responsible for the tissue-specific expression of human glucose transporter type 2 isoform gene. Cooperative role of hepatocyte nuclear factors 1alpha and 3beta.
J Biol Chem. 2000 Jun 16;275(24):18358-65.
PMID: 10748140 [PubMed - indexed for MEDLINE]

11: Kawasaki E, Sera Y, Yamakawa K, Abe T, Ozaki M, Uotani S, Ohtsu N, Takino H, Yamasaki H, Yamaguchi Y, Matsuura N, Eguchi K.
Identification and functional analysis of mutations in the hepatocyte nuclear factor-1alpha gene in anti-islet autoantibody-negative Japanese patients with type 1 diabetes.
J Clin Endocrinol Metab. 2000 Jan;85(1):331-5.
PMID: 10634407 [PubMed - indexed for MEDLINE]

12: Doria A, Yang Y, Malecki M, Scotti S, Dreyfus J, O'Keeffe C, Orban T, Warram JH, Krolewski AS.
Phenotypic characteristics of early-onset autosomal-dominant type 2 diabetes unlinked to known maturity-onset diabetes of the young (MODY) genes.
Diabetes Care. 1999 Feb;22(2):253-61.
PMID: 10333942 [PubMed - indexed for MEDLINE]

13: Hathout EH, Cockburn BN, Mace JW, Sharkey J, Chen-Daniel J, Bell GI.
A case of hepatocyte nuclear factor-1 alpha diabetes/MODY3 masquerading as type 1 diabetes in a Mexican-American adolescent and responsive to a low dose of sulfonylurea.
Diabetes Care. 1999 May;22(5):867-8. No abstract available.
PMID: 10332709 [PubMed - indexed for MEDLINE]

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