Lab 6: Sequence Characteristics

Q0: What's your name?

B. Predict characteristics of your protein

First we'll retreive a protein sequence from NCBI.

In [1]:
```
from Bio import Entrez # we'll import a set of code written for common biological applications
from Bio import SeqIO
```

In [2]:
```
prot_id = "NP_000302" #the accession number of our protein of interest
```

In [3]:
```
Entrez.email = "" # let NCBI know who you are
handle = Entrez.efetch(db="protein", rettype="gb", retmode="text", id=prot_id) # db = database to query, rettype = retrieval type, retmode = retrieval mode
seq_record = SeqIO.read(handle, "gb") # read the contents "fetched" from NCBI
handle.close()
```

In [4]:
```
protdes = seq_record.description # assign the protein's description to protdes
# print the protein  description
print protdes
protseq = seq_record.seq # pull out the sequence associated with this record
# print the amino acid sequence
print protseq
# print the number of amino acids in the sequence
print len(protseq)
```

```
major prion protein preproprotein [Homo sapiens].
MANLGCWMLVLFVATWSDLGLCKKRPKPGGWNTGGSRYPGQGSPGGNRYPPQGGGGWGQPHGGGWGQPHGGGW
GQPHGGGWGQPHGGGWGQGGGTHSQWNKPSKPKTNMKHMAGAAAAGAVVGGLGGYMLGSAMSRPIIHFGSDYE
DRYYRENMHRYPNQVYYRPMDEYSNQNNFVHDCVNITIKQHTVTTTTKGENFTETDVKMMERVVEQMCITQYE
RESQAYYQRGSSMVLFSSPPVILLISFLIFLIVG
253
```

Biopython's SeqUtils module contains functions for estimating properties of your protein.
You can find more information on the class ProteinAnalysis here:
http://biopython.org/DIST/docs/api/Bio.SeqUtils.ProtParam.ProteinAnalysis-class.html.

In [5]:
```
from Bio.SeqUtils.ProtParam import ProteinAnalysis
from Bio.SeqUtils import ProtParamData
from Bio import SeqIO

prot = ProteinAnalysis(str(protseq))

print "The predicted molecular weight of", prot_id, "is", prot.molecular_
weight(), "Daltons" # molecular weight of your protein in Daltons
print "The relative frequency of phenylalanine, tryptophan, and tyrosine
(\"aromaticity\") in", prot_id, "is", prot.aromaticity() # the relative f
requency of Phe+Trp+Tyr
print "The predicted isoelectric point of", prot_id, "is", prot.isoelectr
ic_point() # the pH at which the protein has no net charge
print "The instability index of", prot_id, "is", prot.instability_index()
  # predicts the instability of the protein, values over 40 suggest an un
stable structure

print round(prot.secondary_structure_fraction()[0]*100, 1), "% of amino a
cids in", prot_id, "tend to form helices." # returns a tuple of the fract
ion of amino acids which tend to form helixes, turns or sheets.
print round(prot.secondary_structure_fraction()[1]*100, 1), "% of amino a
cids in", prot_id, "tend to form turns."
print round(prot.secondary_structure_fraction()[2]*100, 1), "% of amino a
cids in", prot_id, "tend to form sheets."
```

```
The predicted molecular weight of NP_000302 is 27659.6898 Daltons
The relative frequency of phenylalanine, tryptophan, and tyrosine ("aroma
ticity") in NP_000302 is 0.114624505929
The predicted isoelectric point of NP_000302 is 9.12603759766
The instability index of NP_000302 is 43.1098814229
25.3 % of amino acids in NP_000302 tend to form helices.
35.2 % of amino acids in NP_000302 tend to form turns.
17.0 % of amino acids in NP_000302 tend to form sheets.
```

Q6: Briefly discuss the relevance of the above characteristics to the biology of your protein.

Q7: If the amino acids were evenly distributed in your protein, how many aromatic amino acids (phenylalanine, tryptophan, and tyrosine) would you expect to see in a protein the length of your protein? Show your work below.

In [6]:
```
prot.aromaticity() * len(protseq)
```

Out[6]:
```
29.0
```

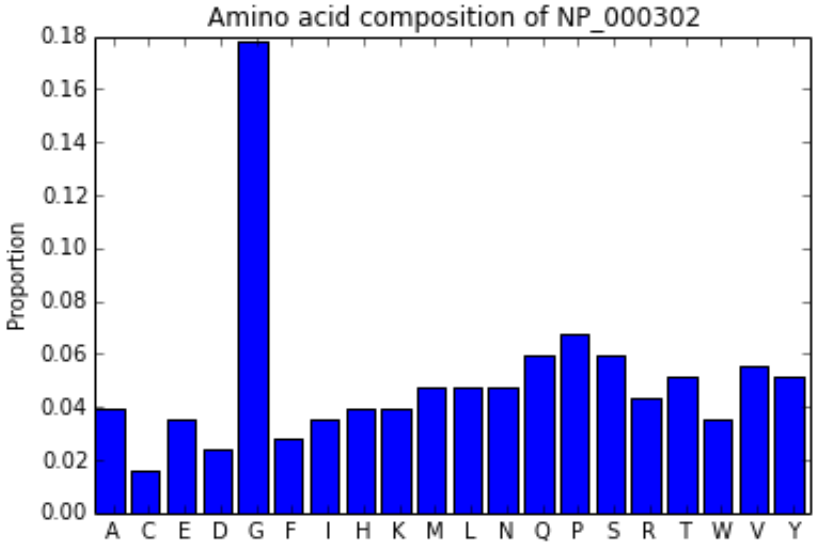
Next generate a graph, with the number of times each amino acid occurs displayed as a percentage of your protein's overall length. The code is executable as written, read through and make sure you understand what is happening at each step.

In [7]:
```
import matplotlib.pyplot as plt # import a set of code written for genera
ting graphics and tell the computer how to display graphs

%pylab inline

percaa = prot.get_amino_acids_percent() # returns a dictionary of the num
ber of times each aa occurs in your protein sequence
amino_a = arange(20) # the x-axis has twenty positions, one for each of t
he 20 amino acids
bar(amino_a, percaa.values()) # chart will be a bar chart, containing ami
no acids on the x-axis and count as percent of length of the sequence on
the y-axis
xticks(amino_a + 0.5, percaa.keys()) # label the x-axis with the amino ac
ids in the same order as in the counts list
title("Amino acid composition of " + prot_id) #add a title to the graph
ylabel("Proportion") #label the y-axis
show()
```

```
Populating the interactive namespace from numpy and matplotlib
```



Q8: Based on the graph you've generated, do the amino acids appear with equal frequency within your protein? What type of amino acids appear over or under represented?

Next generate a graph, with the predicted flexibility values plotted over the length of your protein sequence. Regions with a flexibility index less than 1 are classified as rigid.

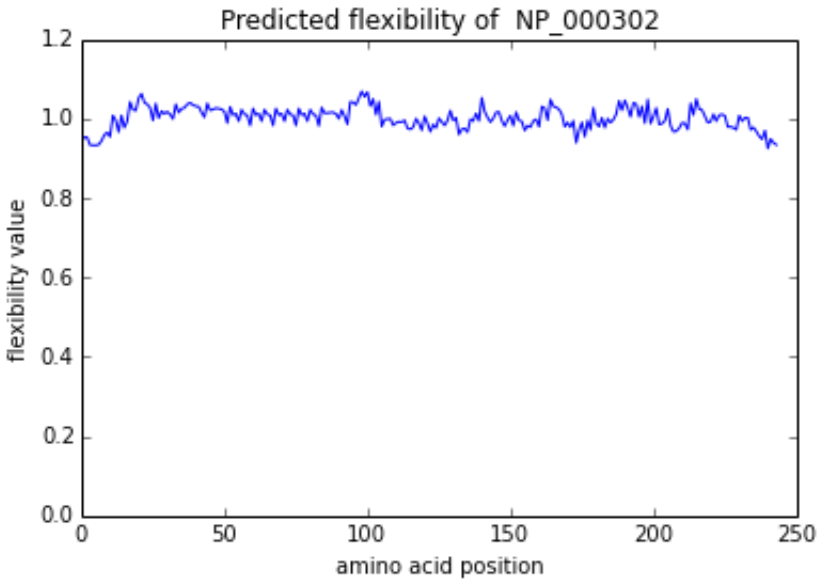
In [7]:
```
import matplotlib.pyplot as plt # import a set of code written for genera
ting graphics and tell the computer how to display graphs
%pylab inline

flex = prot.flexibility()

plt.plot(flex) # plot the flexibility values
plt.axhline(0, color='black') # forces the y-axis to start at 0
plt.ylabel("flexibility value") # label the y-axis
plt.xlabel("amino acid position") # label the x-axis

plt.title("Predicted flexibility of  " + prot_id) # add a title
plt.show()
```

```
Populating the interactive namespace from numpy and matplotlib
```



Q9: Do the predicted flexibility scores differ over the length of your protein?

The distribution of hydrophobic/hydrophilic amino acids throughout a protein sequence is associated with a protein's three-dimensional structure.

We'll loop over the amino acids in the protein, look up the hydrophobicity value of the amino acid, and generate a graph of the hydrophobicity values across the length of the protein sequence.

First, we'll generate a dictionary of the amino acids and their hydrophobicity values . . .

In [9]:
```
hydro = {"I":4.5, "V":4.2, "L":3.8, "F":2.8, "C":2.5, "M":1.9, "A":1.8, "
G":-0.4, "T":-0.7, "S":-0.8, "W":-0.9, "Y":-1.3, "P":-1.6, "H":-3.2, "E":
-3.5, "Q":-3.5, "D":-3.5, "N":-3.5, "K":-3.9, "R":-4.5}
print len(hydro)
```

```
20
```

The more positive the number the more hydrophobic the amino acid.
The hydrophobicity values were derived from Kyte J, Doolittle RF. (1982) A simple method for displaying the hydropathic character of a protein. J Mol Biol. 157(1):105-32.

Loop over the amino acids in the protein sequence, then look up the amino acid in the hydrophobicity dictionary.

In []:
```
# hvalues is a empty list to hold the hydrophobicity values along the len
gth of the protein
hvalues = []
# for each amino acid in the protein sequence, protseq
for aa in protseq:
    # ask if the amino acid is in the dictionary's keys
    if aa in hydro.keys():
        # if so, print the amino acid and it's hydrophobicity value
        print aa, "hydrophobicity:", hydro[aa]
        # add the hydrophobicity value to the list hvalues
        hvalues.append(hydro[aa])
    # if the amino acid is NOT in the dictionary's keys
    else:
        print "Amino acid", aa, "not in dictionary." # print out a warnin
g
        break # stop the flow of the for loop
```

Now, let's generate a graph.

Ploting the hydrophobicity of each position in the protein sequence results in a noisy graph. Let's calculate the average hydrophobicity over a sliding window 20 aa in length, and include a red line on the graph at 1.6 on the hydrophobicity scale. Regions with hydrophobicity values greater than 1.6 are more likely to be transmembrane domains.

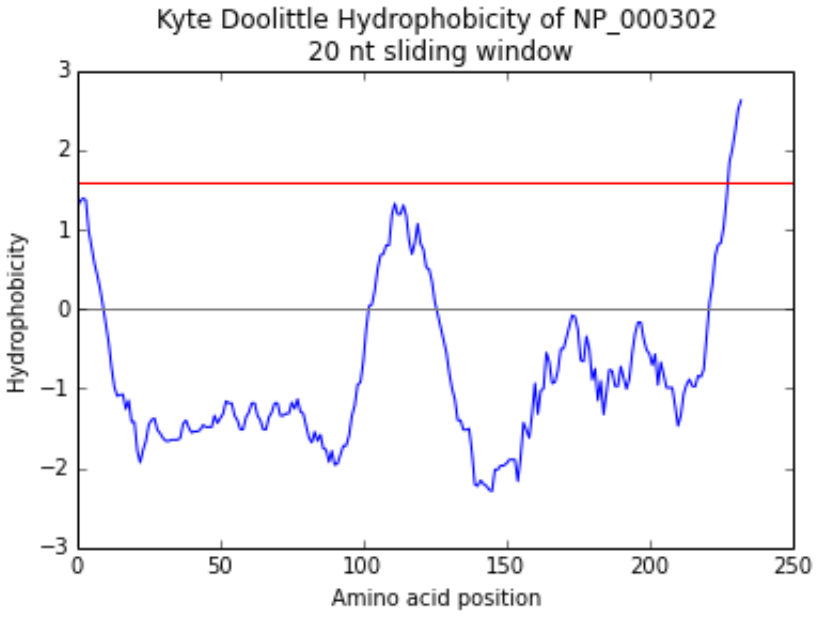
In [13]:
```
window_len = 20 # define the length of the window
smooth_hydro = []# smooth_hydro = a list to hold the moving average value
s
for i in range(len(hvalues)-window_len): # for each number in a list
    # set the hydrophobicity value, hval, to 0
    hval = 0
    for v in hvalues[i:i+window_len]:# for each aa in window
        # increment the value of hval by v
        hval += v
    avh = hval/len(protseq[i:i+window_len]) # when the loop over the slic
e is complete, calculate the average hydrophobicity value of the region
    # add the average to the list smooth_hydro
    smooth_hydro.append(avh)
```

We'll generate a graph of the smoothed values.

In [14]:
```
import matplotlib.pyplot as plt # import a set of code written for genera
ting graphics and tell the computer how to display graphs
%pylab inline

plt.plot(smooth_hydro) # plot the value of smooth_hydro
plt.axhline(0, color='grey') # add a horizontal grey line at y=0
plt.axhline(1.6, color='red')# add a horizontal red line at y=1.6
plt.ylabel('Hydrophobicity') # add a label to the y-axis
plt.xlabel('Amino acid position') # add a label to the x-axis
plt.title("Kyte Doolittle Hydrophobicity of " + prot_id + "\n 20 nt slidi
ng window") # add a title to the graph
plt.show()
```

```
Populating the interactive namespace from numpy and matplotlib
```



Q10: Based on the graph you've generated, do you think there are any transmembrane domains in your protein? Explain.

That's all folks!

Please save your notebook and upload the notebook to Blackboard.