## Making Sequence logos

**Q1)** Below is a multiple alignment of 35 human sequences. The sequences have been aligned around a donor splice. That site is indicated as the boundary between the 'Dark blue' and 'Dark red' colours.

----Exon|intron----01234567890123456789 tatcacaATGGTAGGTAACT **TCAACCAGGAGTAAGTCTTG GTTGCACCCTGTAAGTCTCA** tatcacaATGGTAGGTAACT TCAACCAGGAGTAAGTCTTG **CTTGCGAGAGGTGTGACATG GCTCTACTCGGTAAGGTGAC GCCTGGAGAGGTAATGACCC** CAAAACCATTGTGAGTAATC **GCCAGAGCAGGTAAAATATC** GAACAGTCAGGTCTGTTGCT GAAGGCCCAGGTGAGCATAA TCCTCTACAGGTGGGTACAT **GGCGTCCCGCGTAAGTATGG CCTCGTGCAGGTAAGATTAA** TGCATGACAGGTGAGTGTTA **GAAATGTACAGTAAGTCTCT GGTTCTCTGGGTAAGTAGAG AAATGTACAGGTGAGTACTG ACCTCGCTTGGTACGTGGGA AATCAGACAGGTATAGAAAC AGGACAGAAGGTAATTTTCT AACTATTTGGGTAGCA AAACTTGAAGGTATGTTGTT CTGGGATAAGGTAAAAGTAT** TTGCACCCAGGTTAGTGGAT ACTTCAATCGGTATGTTTTC **ACAGAGAAAAGTAAATTCCT** AATGGGAAAGGTAACAACAA CATGCTACAGGTAGGTGAAT ggctaggATGGTGAGGGCGC **CGACGCGGGCGTGAGAGGCG** CATTGAGAATGTGAGTTATT **AACAGAGCAGGTACTTGTAT TGAACCAAAGGTGAAGACAT** 

Calculate the counts and frequencies (P) for positions 6-5. You have each been assigned one column on the upper right corner of the handout.

Position	6	7	8	9	0	1	2	3	4	5
Counts A										
Counts T										
Counts C										
Counts G										
P(A)										
P(T)										
P(C)										
P(G)										

Note P(A) is the frequency of amino acid A, this number of between 0 and 1, and the sum of P over the four nucleotides is 1.

Q2) Calculate the Entropy (S) and Information Content (I) using the formula below

Eq.1 
$$S(p) = -\sum_{a} p_a \log_2(p_a) = -\frac{1}{\log(2)} \sum_{a} p_a \log(p_a)$$

where log<sub>2</sub> is the logarithm with base 2, and log is the logarithm with base 10 (or any base for that sake)

Eq.2 
$$I = 2.0 - S(p)$$

position	6	7	8	9	0	1	2	3	4	5
Entropy										
Information content										

- Q3) Where does the constant 2.0 come from in Eq.2?
- Q4) Draw an approximate Logo Plot by hand on the White board

If you have internet-access

Q5) Submit the multiple alignment to the WebLogo server <a href="http://weblogo.berkeley.edu/">http://weblogo.berkeley.edu/</a>

Make both the Logo plot and a frequency plot Explain what you see on the two plots.