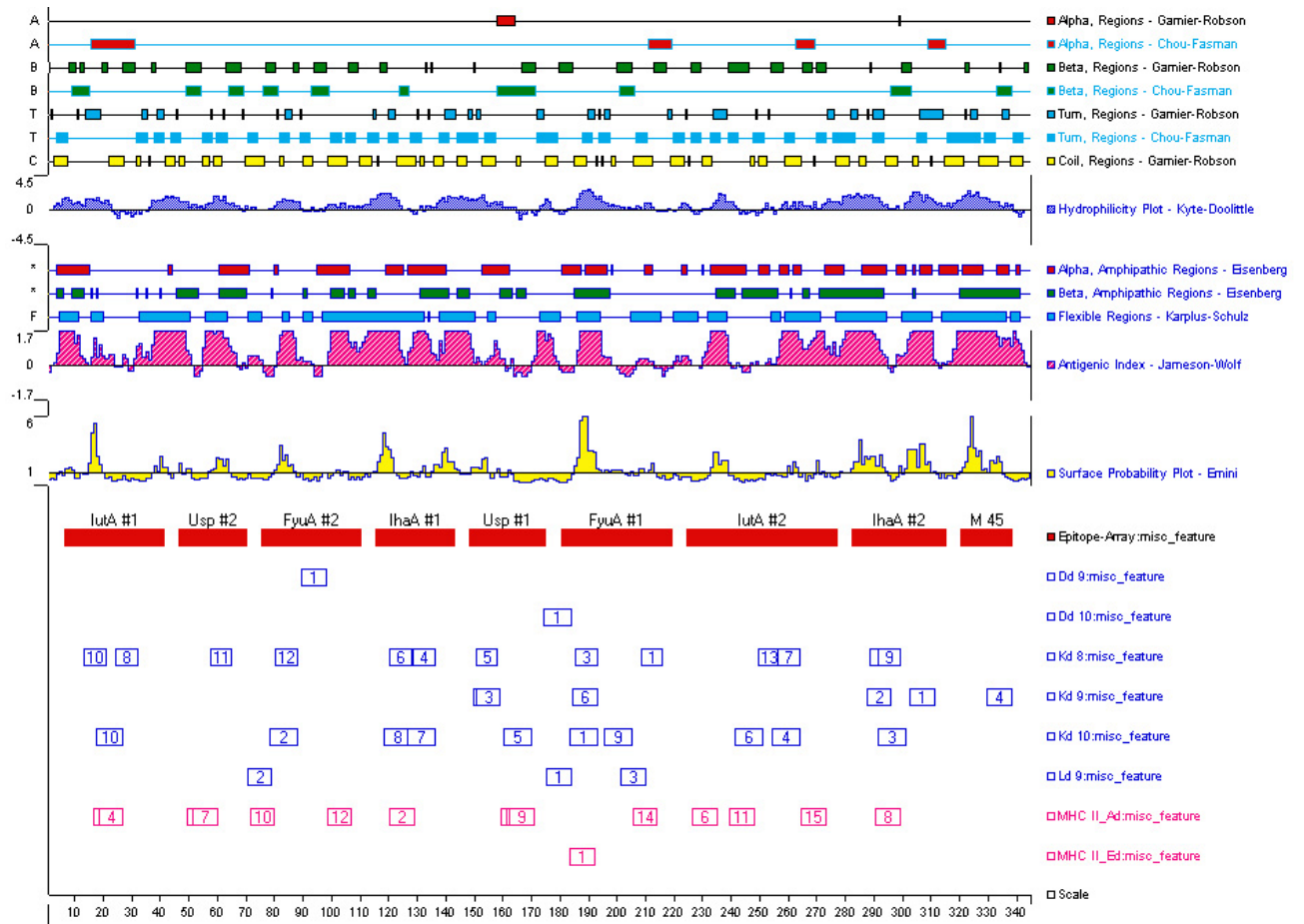
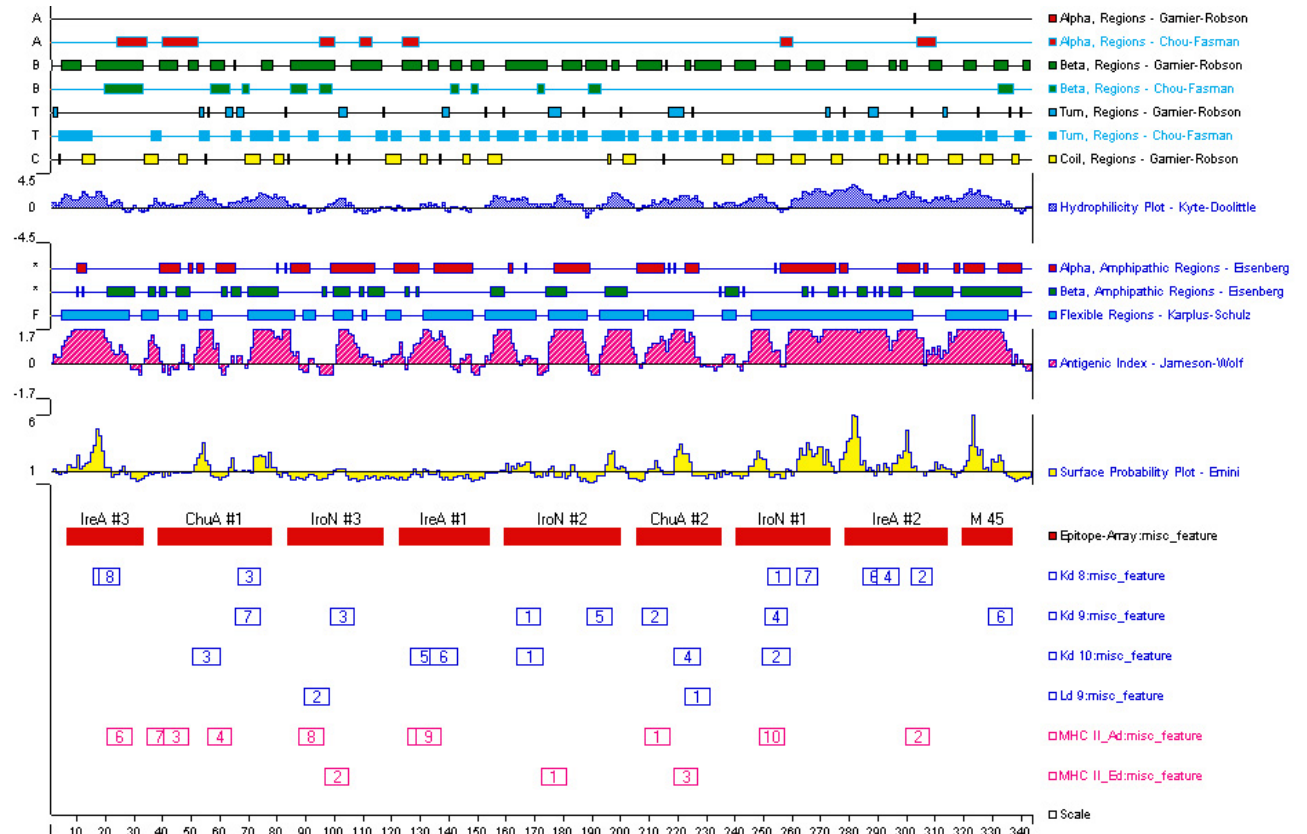


S1:

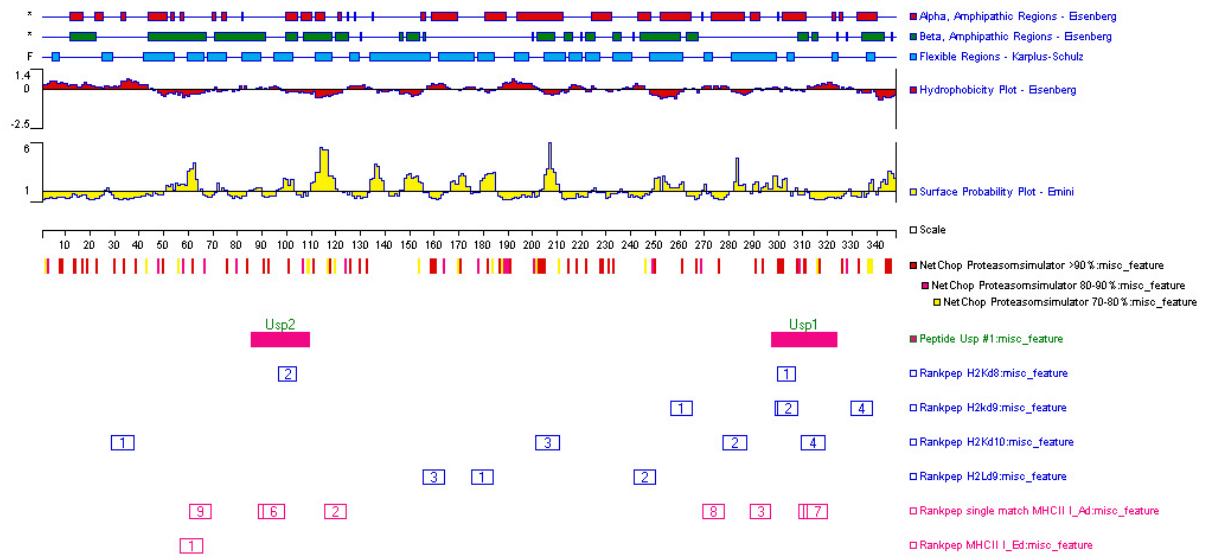


Vol1

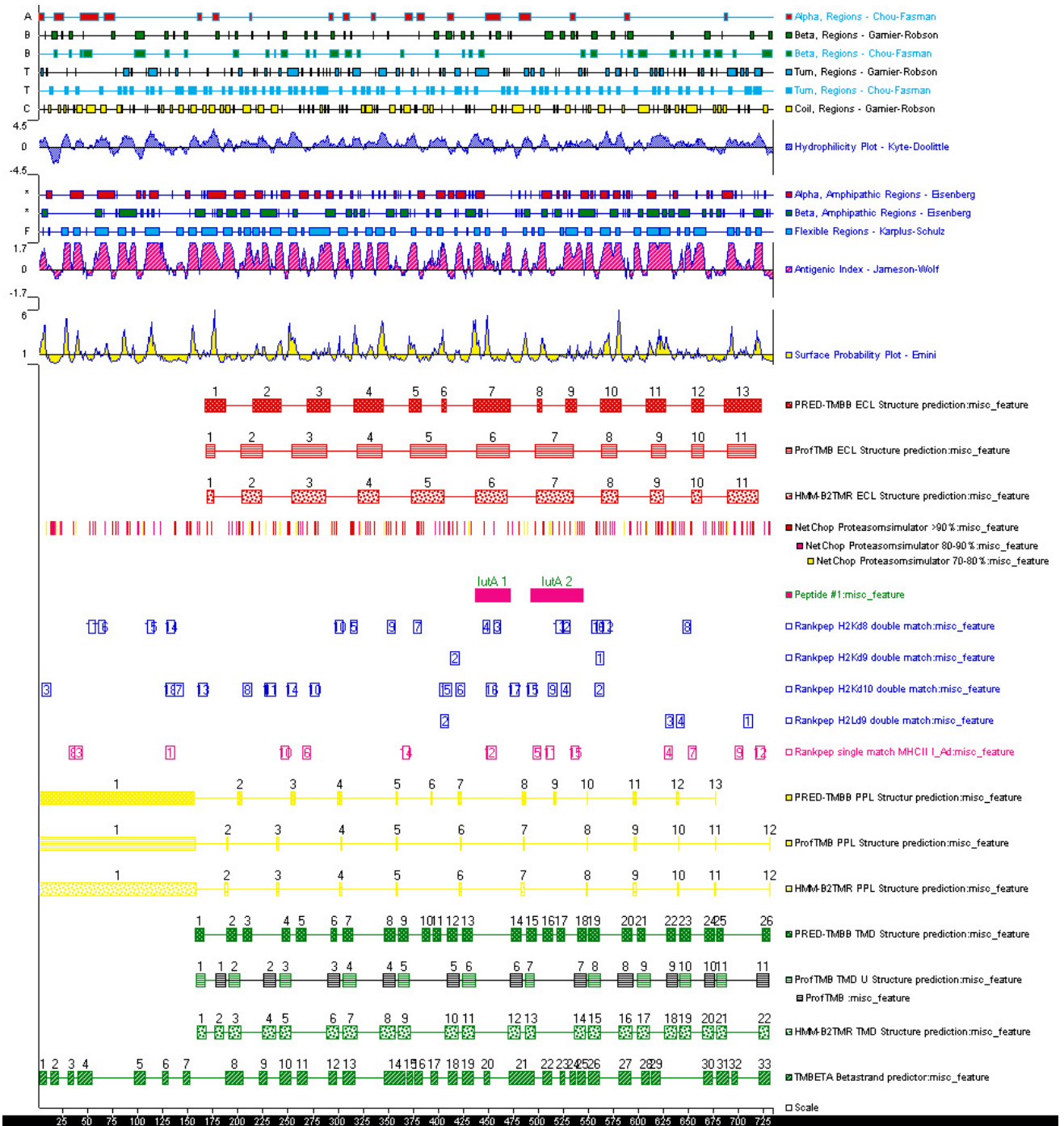


Vol2

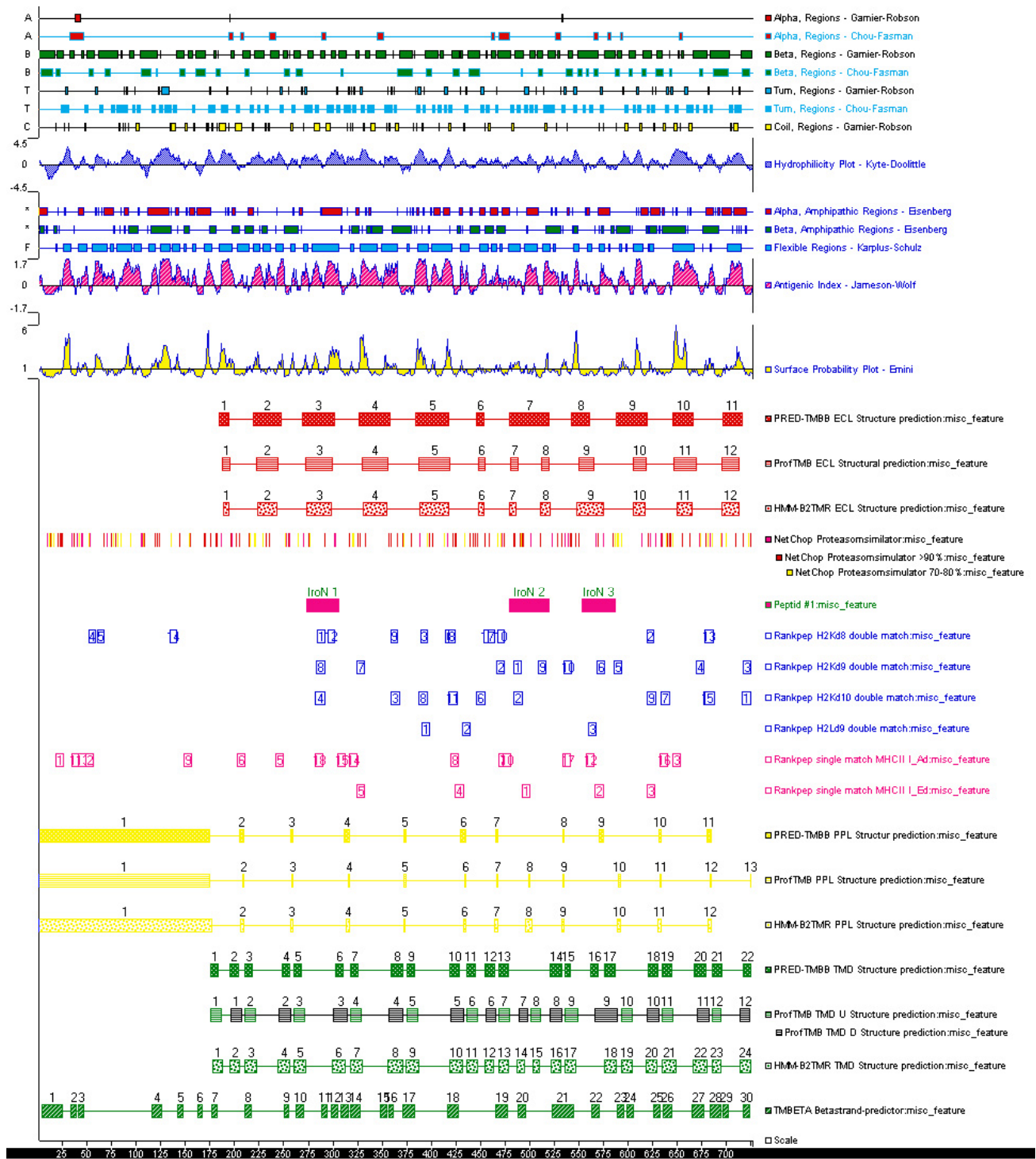
S2:



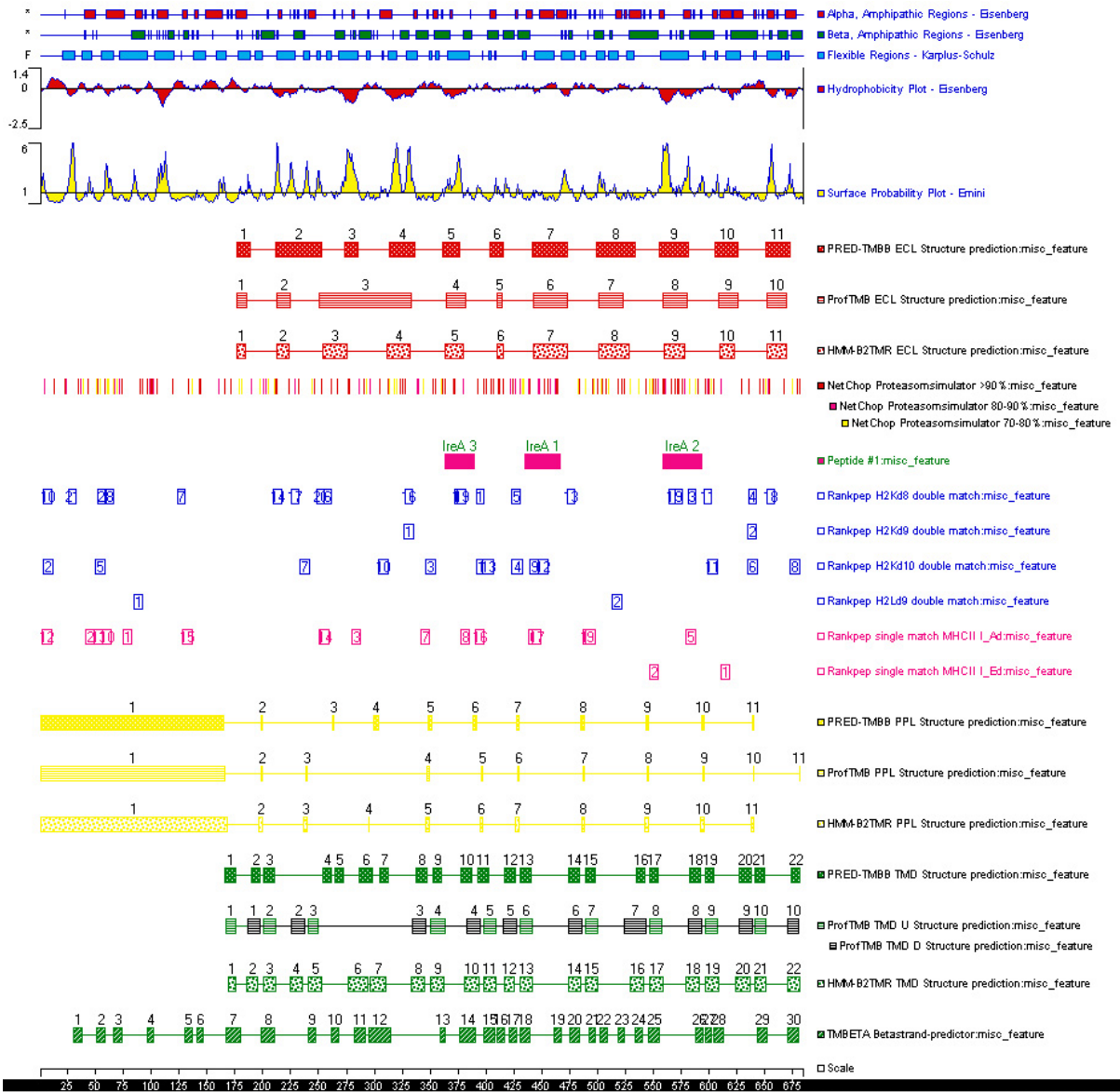
UspA



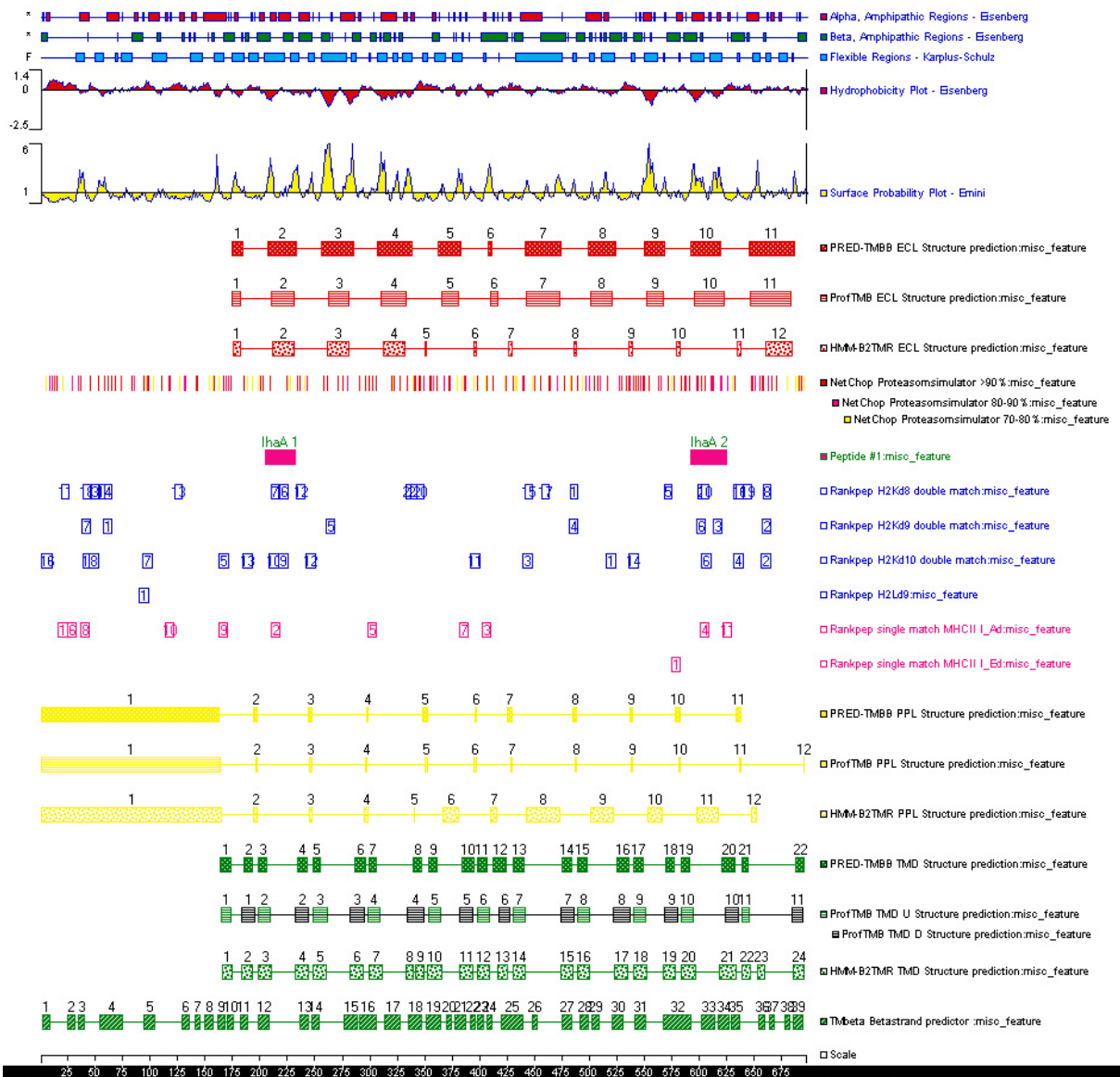
IutA



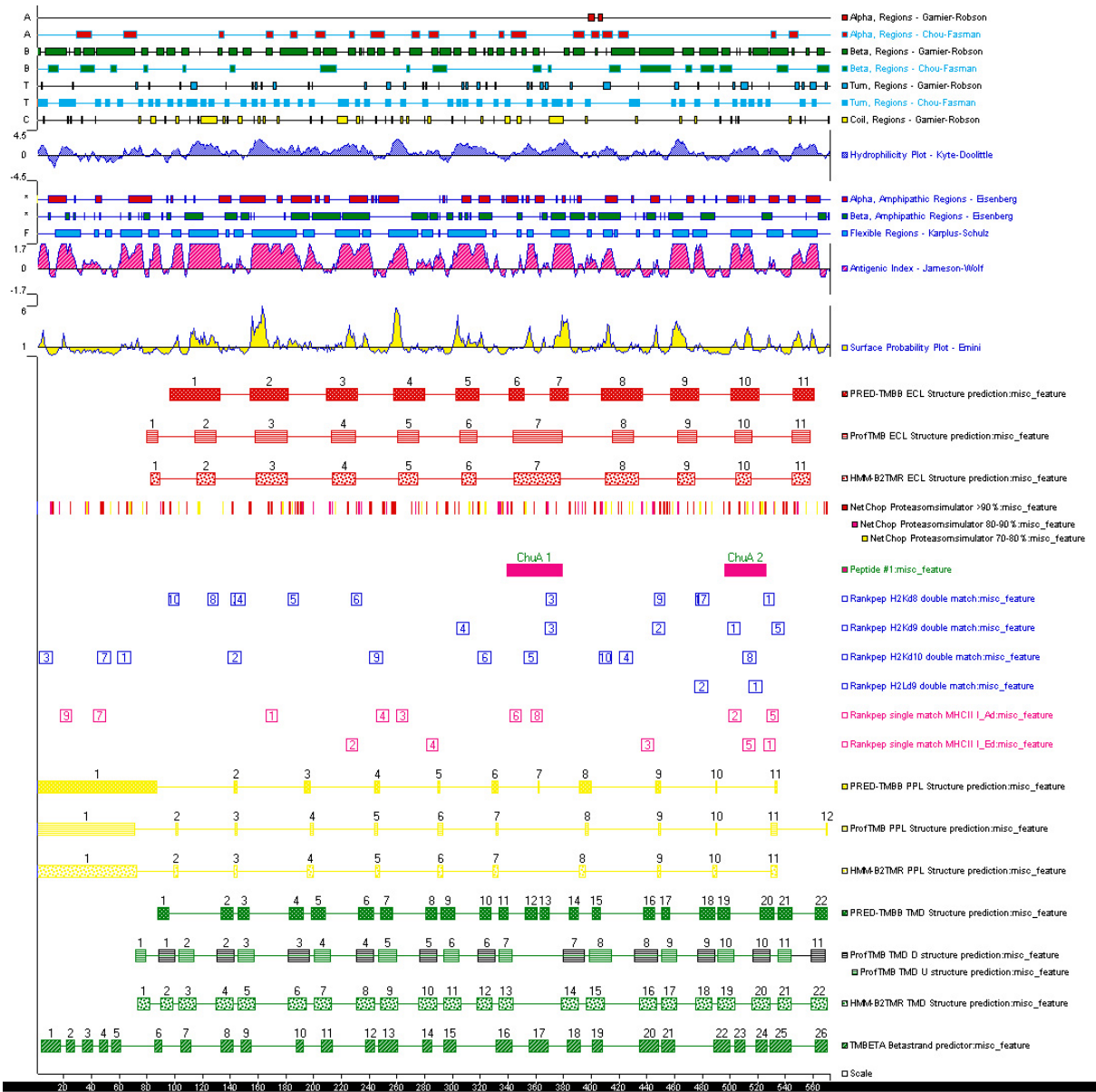
IroN



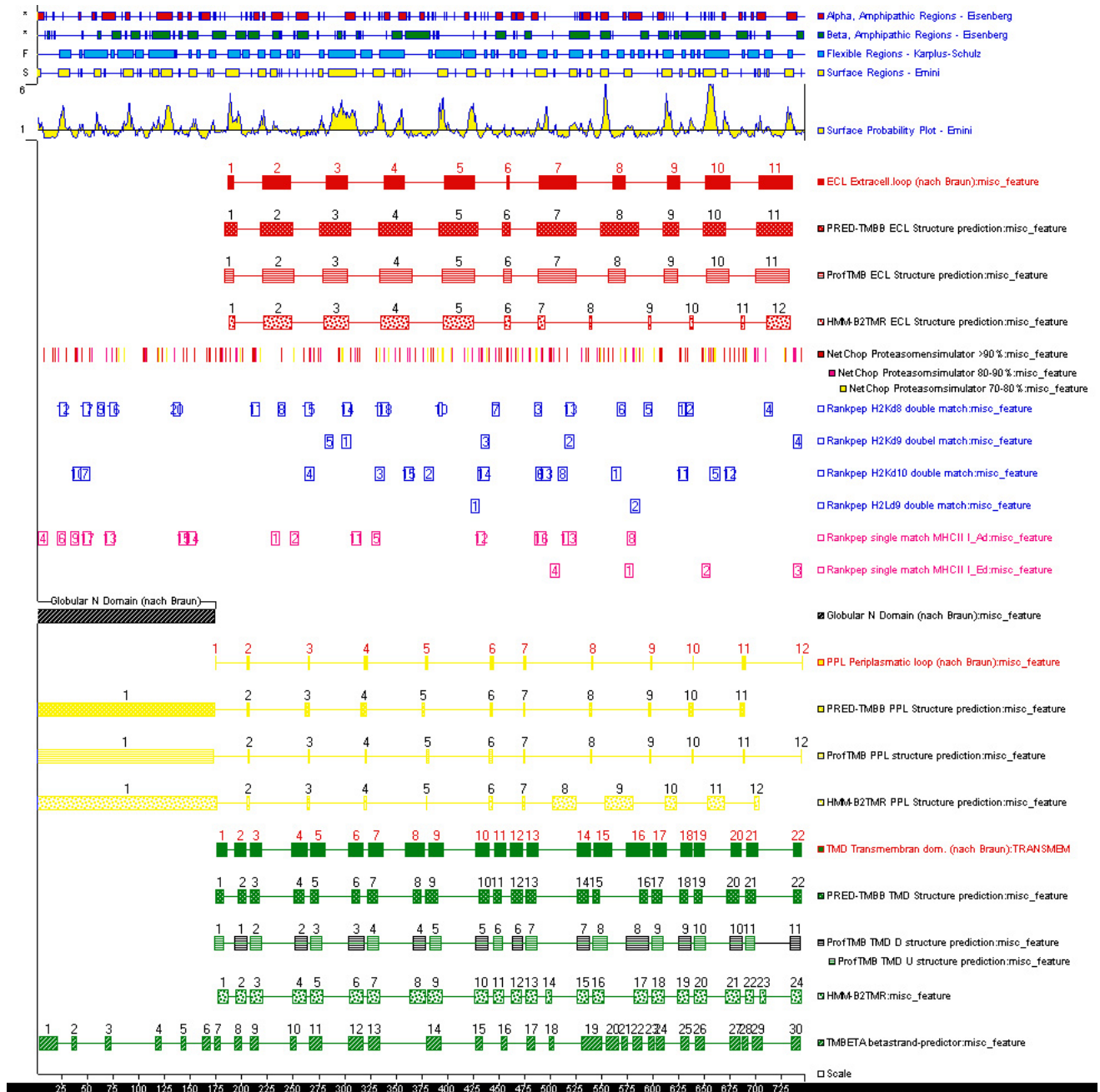
IreA



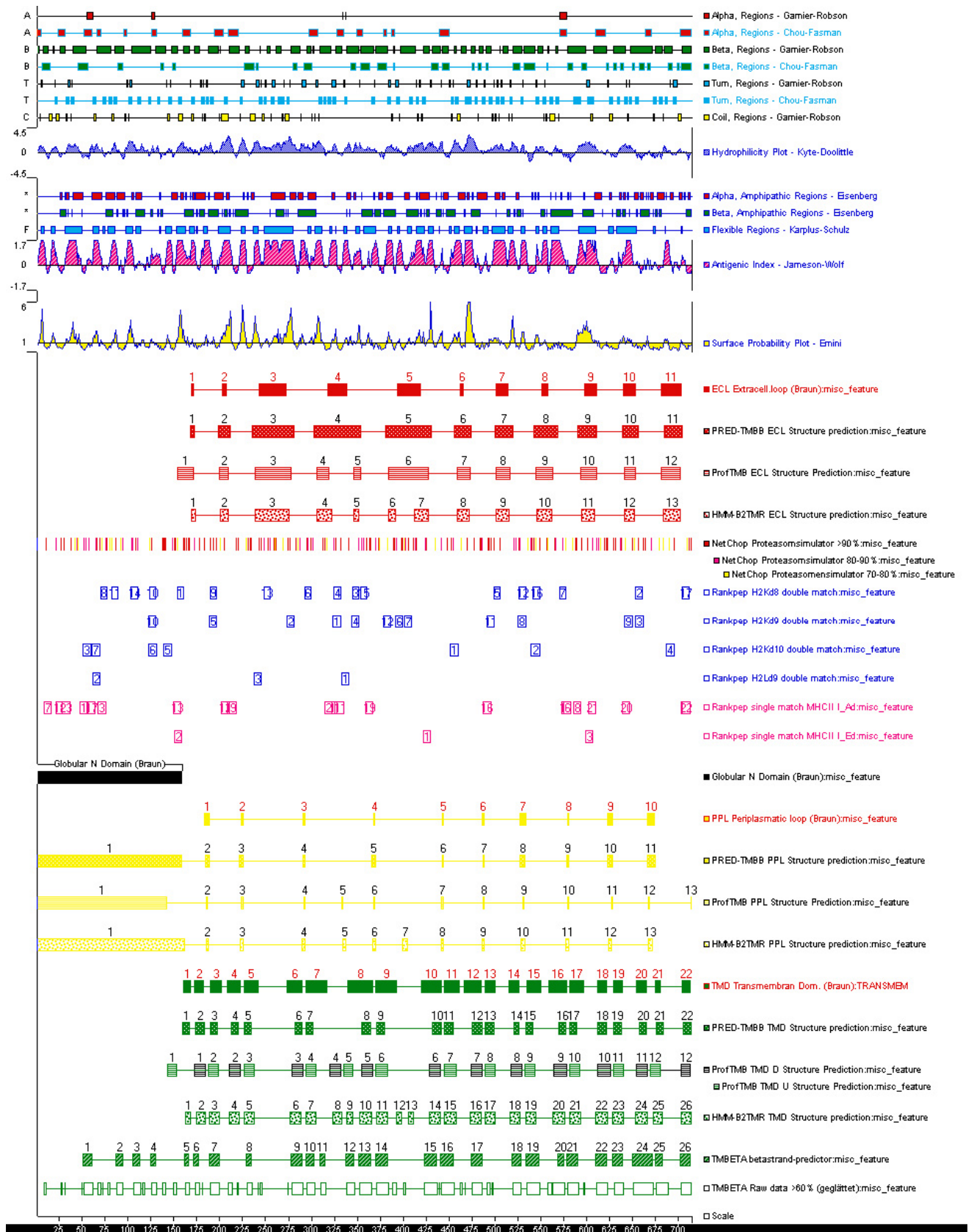
IhaA



ChuaA



FepA



FhuA

S3:

Properties of vaccine proteins	Vol1:	Vol2:
Molecular weight:	36172.11 Da	36116.96 Da
AA-residues	343	342
Basic AA: Lys, Arg	32	37
Acidic AA: Asp, Glu	30	32
hydrophobias AA: Ala, Ile, Leu, Phe, Trp, Val	81	78
polar AA: Asn, Cys, Gln, Ser, Thr, Tyr	100	101
Isoelectric point: pH:	8.571	8.941
Charge at pH 7,0:	2.920	5.364

S4:

Sequence of the synthetic gene

Vol1:
CTCGGATCCAACCGCGTGGACGACTTTATTGATTACACTCAGCAGCAGAAAATTGCAGC GGGCAAAGCTATCTCGGCGGACGCAATCCCGGGTGGCTCTGTAGATTATGACAATGGCC CTGGTCCGGGTGGCAAACCGCAAGTATACGGCGTCCATACTGGTGAGGGCACCCTTAC GAAAACGTTTCGCGTCGCCAACATGGGTCCCTGGTCCGGGAGCGGCGACGTTTTATACCA CACTAAAGACATGCAGCTTTACTCCGGTCCGGTTGGCATGCAGACTCTGAGCAACGCGG GTAAGGCGGACGCGACTGGTGTGCGAAGGCCCGGGTCCGGGTTCTACGCAGCAGCGCCAG GGTTCCTCCGTGACCTCCTTGAGCGACACCGCAGCAACGCGCATTCCCTATCCTACCGAG TCCCAGGGTCCGGGTCCCGGTGAAGGTTACTATTATGGCCCGAACGAAATCGTTAAGAA ATTCCAAATCCACCACGTTGTGGCTATTGAACATGGAGGTGGCGGTCCGGGTCCGGGTG TTTTTGGTCTGTACCGTCAAACACCCGCGAAAAGCTGAATAGCGCATATAACATGCCG ACGATGCCGTATCTGTCTTCCACTGGTTATACTACCGCCGAAACCCTCGCGGCATACGGC CCGGGTCCCTGGCTTCAGCCAGGGCGTGGCACTGCCTGACCCAGGTAAATACTATGGTCG CGGTATCTACGGCGCTGCGGTCAACGGTCACCTGCCGCTCACCAAATCTGTAAACGTTTC GGACTCCAAGCTGGAAGGTGTTAAAGTGGATAGCTACGAGCTGGGCTGGCGTGGCCCCG GTCCGGGCCGTGGTAAACTCCGCGCTTACCCAGAATACTCATCTCTGTCTGTCTGTTCA GAAAAGGTGTACGACGAGAAAGGTGAGTACCTGAAAGCCTGGACTGGTCCGGGCCAG GTATGGATCGTTCCCGTGACCGTCTGCCTCCATTCGAAACCGAAACCCGCATCCTGCTCCA GCTGCAGTAAGGTACCGTTCGAC
Vol2:
CTCGGATCCCTGCGTGACGATAGCGCCACTGGTAAAAAGACTACTGAAACTCAAAGCG TATCTATCAAACAGAAAGCGGTGTTTATCGAAGGTCCTGGCCCAGGCTATGCACAGGC CTTTCGCGCTCCTACCATGGGTGAGATGTACAATGATTCTAAGCATTCTCTATCGGTCG CTTCTACACAAACTACTGGGTTCCGAACCCAAACCTGCGGCCAGAACTGGCCCCGGGTC CTGGCGGCGACAACGTAATTGGTCAGACCGCTTCGGGTGCGTACATCCTGAAATGGCAA AACGGCGGCAAAGCTCTGGTGGATGGTATCGAAGCGTCTATGTCGTTTGGTCCAGGCC GGGTGGTATCGCCAAAGCATTTCGTGCCCCGTCCATCCGTGAAGTGTCCCCGGGGTTCG GTACTCTGACCCAGGGTGGTGTAGCATCATGTACGGCAATGGCCCCGGGCCGGTAAA GCACCCAACCTGTACCAATCCTCGGAAGGTTACCTGTTGTATTCCAAAGGTAACGGTTG TCCGAAAGATATCACCTCTGGGGGCTGCTACCTGATCGGCAATAAGGACCTGGACCCAG AAGGTCCGGGTCCCTGGCGTCGGAACCTTCGCGGACCGCAGCACTCACATTAGTTCTTCTT ATAGCAAACAGCCGGGTTACGGAGTTAACGATTTCTACGTTTCCCTACGGTCCGGGCCCA GGCAATATCTATGCTGGTGATACTCAAATTCCTCCTCCAGCGCCGTGACCGAATCTCTG GCCAAGAGCGGCAAAGAAACCAACCGCCTGTACCGCCAGAACGGCCCCGGGCCGGGTC

GCCGCAAAGCGATGATGAATCCCTGAACGGCAAGTCCCTTAAGGGCGAGCCGCTGGA
ACGTACCCCGCGTCACGCGGCAAACGCAAACTGGAATGGGACTACACTGGCCCAGGTC
CAGGTATGGATCGTAGCCGAGATCGTCTGCCACCGTTCGAACTGAAACGCGCATCCTGT
TGCAGCTGCAGTAAGGTACCGTCGAC

Supplemental Fig. S1: Analysis of the vaccine proteins Vol1 and Vol2 with Garnier-Robson and Chou-Fasman (α -; β -; turn-regions), Kyte Doolittle hydrophobicity plot and Jameson-Wolf (antigenic index). Newly simulated putative MHC-I and -II epitopes are indicated for each allele of the Balb/c mouse.

Supplemental Fig. S2: Analysis of the vaccine target proteins Usp, IutA, IroN, IreA, IhaA and ChuA plus the control proteins FepA and FhuA. Plots with Garnier-Robson and Chou-Fasman (α ; β ; turn- regions), Kyte Doolittle (hydrophobicity) and Jameson-Wolf (antigenic index) as well as Emini (surface probability) and Karplus-Schulz (α ; β ; flexible) are shown. Results of structure prediction algorithms Pred TMBB, PROFtmb, HMM-B2 TMR and TM-Beta are indicated for all proteins except Usp, which has no outer membrane portion. Putative MHC-I and -II epitopes are indicated for each allele of the Balb/c mouse and putative proteasome cleavage sites (Net Chop) are also marked. The subunits chosen for the vaccine protein are indicated as pink boxes and numbered according to the content of predicted epitopes.

Supplemental Fig. S3: Properties and amino acid composition of the vaccine proteins Vol1 and Vol2.

Supplemental Fig. S4: Nucleotide sequence of the synthetic gene used to express multi-epitope vaccine proteins Vol1 and Vol2.