Comp 555 - BioAlgorithms - Spring 2021



Jumping into Genomes

A simple genome





Characteristics of Viral genomes:

- Small, dense, and tricky
- Viral genomes code for functional proteins in order to "live", but rely on a host's machinery to perform essential functions
- Small genomes (3K 30K bases) with a few "key" genes

Today's Virus





SARS-CoV-2, the virus that causes COVID-19

- 29903 bases of the original Wuhan isolate
- 10 (11?) genes, 4 structural, 2 with primary functions

How viral life works



Youtube: https://www.youtube.com/watch?v=Xv3TxtFtCNE CORONAVIRUS

Comp 555 - Spring 2021



Time to get serious

- By next Tuesday's class meeting everyone should set up a Jupyter Notebook environment
- Recommend using Anaconda

https://www.anaconda.com/products/individual

- \circ $\,$ $\,$ Includes an isolated environment, an IDE, common packages, and a package manager $\,$
- Will need it for problem sets and exams
- Next Wednesday's office hours will focus on helping folks install Jupyter
- COMP555 accounts should be up by next Tuesday
- We'll start using Python and Jupyter today.
 - You should go back through today's Notebook to verify your setup



Let's look at it



FASTA is a common format for biological sequences

- Each sequence is preceded by a header line that starts with '>'
- Followed by multiple lines of sequence data from a standard alphabet
 - For DNA, alphabet = "ACGT"
 - For RNA, alphabet = "ACGU"
 - For Proteins, alphabet = "ACDEFGHIKLMNOPQRSTUVWY"
- A sequence ends when either another header line is reached or the end-of-file
- Multiple sequences per file are allowed
- Sequences are 1-indexed rather than 0-indexed!



TTCTGCAGGCTGCTTACGGTTTCGTCCGTGTTGCAGCCGATCATCAGCACATCTAGGTTT CGTCCGGGTGTGACCGAAAGGTAAGATGGAGAGCCTTGTCCCTGGTTTCAACGAGAAAAC ACACGTCCAACTCAGTTTGCCTGTTTTACAGGTTCGCGACGTGCTCGTACGTGGCTTTGG

In [123]:

In [125]:

TATTGACGCATACAAAACATTCCCACCAACAGAGCCTAAAAAGGACAAAAAGAAGAAGAGGC TGATGAAACTCAAGCCTTACCGCAGAGACAGAAGAAACAGCAAACTGTGACTCTTCTTCC TGCTGCAGATTTGGATGATTTCTCCAAACAATTGCAACAATCCATGAGCAGTGCTGACTC AACTCAGGCCTAAACTCATGCAGACCACACAAGGCAGATGGGCTATATAAACGTTTTCGC ACAAGTAGATGTAGTTAACTTTAATCTCACATAGCAATCTTTAATCAGTGTGTAACATTA GGGAGGACTTGAAAGAGCCACCACATTTTCACCGAGGCCACGCGGAGTACGATCGAGTGT ACAGTGAACAATGCTAGGGAGAGCTGCCTATATGGAAGAGCCCTAATGTGTAAAATTAAT

GTTCTCTAAACGAACTTTAAAATCTGTGTGGCTGTCACTCGGCTGCATGCTTAGTGCACT

CACGCAGTATAATTAATAACTAATTACTGTCGTTGACAGGACACGAGTAACTCGTCTATC

AGACTCCGTGGAGGAGGTCTTATCAGAGGCACGTCAACATCTTAAAGATGGCACTTGTGG CTTAGTAGAAGTTGAAAAAGGCGTTTTGCCTCAACTTGAACAGCCCTATGTGTTCATCAA ACGTTCGGATGCTCGAACTGCACCTCATGGTCATGTTATGGTTGAGCTGGTAGCAGAACT

7

>NC_045512.2 |Severe acute respiratory syndrome coronavirus 2 isolate Wuhan-Hu-1, complete genome

An Example

!head data/SARS-CoV-2.fa

!tail data/SARS-CoV-2.fa







A little code for reading FASTA



In [129]: import gzip def loadFasta(filename): """ Parses a classically formatted and possibly compressed FASTA file into two lists. One of headers and a second list of sequences. The ith index of each list correspond.""" if (filename.endswith(".gz")): fp = gzip.open(filename, 'r') else: "splits" the file at every header fp = open(filename, 'r') line. Then each of those sections # split at headers data = fp.read().split('>') is split at each return '\n'. "pop()" fp.close() is used to remove the header line. # ignore whatever appears before the 1st header The sequence is formed by joining data.pop(0) headers = [] together the remaining lines of sequences = [] sequences. A "+" is added to the for sequence in data: front to give the string an offset lines = sequence.split('\n') headers.append(lines.pop(0)) of1 # add an extra "+" to make string "1-referenced" sequences.append('+' + ''.join(lines)) return (headers, sequences)

```
In [130]: header, seq = loadFasta("data/SARS-CoV-2.fa")
```

```
for i in range(len(header)):
    print(header[i])
    print(len(seq[i])-1, "bases", seq[i][:30], "...", seq[i][-30:])
    print()
```

Let's take a minute to explore



Genome sequences are best understood by examining subsequences

Often we examine all subsequences of length k, called k-mers.

The statististics and patterns of k-mers can shed light on a genome's organization and local function.

Two simple rules to consider:

- 1) There are 4^k possible DNA k-mers
- 2) A linear sequence of length N has N k + 1 k-mers

ATGGAGAGCCTTGTCCCTGGTTTCAACGAGAAAACA ATGGAG CCTTGT CTGGTT AACGAG TGGAGA CTTGTC TGGTTT ACGAGA GGAGAG TTGTCC GGTTTC CGAGAA GAGAGC TGTCCC GTTTCA GAGAAA TTTCAA AGAGCC GTCCCT AGAAAA GAGCCT TCCCTG TTCAAC GAAAAC AGCCTT CCCTGG TCAACG AAAACA CCTGGT CAACGA GCCTTG

A 36 base sequence has 31, 6-mers

$$(36 - 6 + 1) = 31$$

Genome "k-mer" statistics



In [104]:	def kme kme for ret	erCounts(sec erDict = {} i in range kmer = sec kmerDict[k curn kmerDic	<pre>1, k): ((1,len(seq)-k+1): [[i:i+k] imer] = kmerDict.get it</pre>	(kmer,0) +	1			
In [139]:	<pre>print(' k k-mers 4^k N-k+1 missing repeated') for k in range(3,25): kmers = kmerCounts(seq[0], k) print("%3d %10d %20d %10d" % (k,len(kmers),4**k,(len(seq[0])-1)-k+1,4**k-len(kmers),(len(seq[0])-1)-k+1-len(kmers)))</pre>							
	k	k-mers	4^k	N-k+1	missing	repeated		
	3	64	64	29901	0	29837	There is one 5-mer '(GGGG'	
	4	256	256	29900	0	29644		
	5	1023	1024	29899	1	28876	missing from this genome	
	6	3756	4096	29898	340	26142		
	7	10696	16384	29897	5688	19201	X ^s	
	8	20185	65536	29896	45351	9711		
	9	26360	262144	29895	235784	3535		
	10	28789	1048576	29894	1019787	1105		
	11	29566	4194304	29893	4164738	327		
	12	29777	16777216	29892	16747439	115		
	13	29835	67108864	29891	67079029	56		
	14	29855	268435456	29890	268405601	35		
	15	29861	1073741824	29889	1073711963	28		
	16	29866	4294967296	29888	4294937430	22		
	17	29869	17179869184	29887	17179839315	18	There are nine 2.4-merc that are	
	18	29871	68719476736	29886	68719446865	15		
	19	29871	274877906944	29885	274877877073	14	repeats of another. (BTW, they	
	20	29871	1099511627776	29884	1099511597905	13	/ are copies of a sinale 24-mer,	
	21	29871	4398046511104	29883	4398046481233	12	· AAAAAAAAAAAAAAAAAAAAAAA	
	22	29871	17592186044416	29882	17592186014545	11		
	23	29871	70368744177664	29881	70368744147793	10	which appears 10 times.)	
	24	29871	281474976710656	29880	281474976680785	9	K	
- Sprina 2	2021						()	

What do k-mer statistics look like?

```
import matplotlib.pyplot as plot
%matplotlib inline
# Compute a histogram of kmer-counts (i.e. how many kmers appear 1 time, 2 times, 3 times ...)
k = 6
maxcount = 50
kmers = kmerCounts(seq[0], k)
hist = [0 for i in range(maxcount)]
for kmer in kmers:
    count = kmers[kmer]
    if (count < maxcount):</pre>
        hist[count] += 1
fig = plot.figure(figsize=(10,6))
plot.plot([i for i in range(maxcount)], hist)
plot.show()
 400
 300
 200
 100
```

20

30

40

50



Okay, there are 432 G-mers that appear only once, 430 that are repeated twice, and the fewer and fewer are repeated 3, 4, 5, and so on.

Meanwhile there are two 6-mers that are repeated more than 40 times ("TTGTTA" 42 times, and "TGTTAA" 41 times)

But are these counts typical?

Comp 555 - Spring 2021

10

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In [90]: M import matplotlib

How does it compare to a random sequence?



In [131]: ▶ import random

```
fig = plot.figure(figsize=(10,6))
for j in range(20):
    # Make a fake genome of random nucleotides
    fake = '+' + ''.join(random.choices("ACGT", k=len(seq[0])-1))
    k = 6
    maxcount = 50
    kmers = kmerCounts(fake, k)
    hist = [0 for i in range(maxcount)]
    for kmer in kmers:
        count = kmers[kmer]
        if (count < maxcount):</pre>
            hist[count] += 1
        if (count > 25):
            print(kmer, count)
    plot.plot([i for i in range(maxcount)], hist)
plot.show()
```



In a random sequence of the same length as SARS-CoV-2, there would be far fewer unique G-mers (typically around 20). Also, most G-mers would appear approximately 7 times (roughly 29903/4096 = 7.3 times).

Also it would be rare for any 6-mer to be repeated more than 25 times.

Conclusion ... virus sequences aren't random patterns



Let's look at some key genes

The "Spikes" of the viral envelope seek out the ACE2 recptors in order to infect a cell.

Eventually, an immune response is set off.

B-cells use knowlege (acquired from T-cells) about the Spike sequence to generate antibodies that target the virus to inactive it.

The key point is learning to recognize the spike sequence.



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How an mRNA vaccine works

https://www.youtube.com/watch?v=LcTEmHlvY10





It we introduce a proxy that "looks" sufficently like the Spike, then we can set off the immune reaction, without having to go through the infection.

From "Pfizer-BioNTech COVID-19 vaccine" wikipedia page:

Sequence [edit]

The modRNA sequence of tozinameran, the active ingredient in the Pfizer-BioNTech COVID-19 vaccine, is 4,284 nucleotides long, with a molecular weight of approximately 1388 kDa.^{[50][51]} It consists of a five-prime cap; a five prime untranslated region derived from the sequence of human alpha globin: a codon-optimized gene of the full-length spike protein of SARS-CoV-2 (bases 55–3879), including the signal peptide (bases 55–102) and two proline substitutions (K986P and V987P, designated "2P") that cause the spike to adopt a prefusion-stabilized conformation reducing the membrane fusion ability, increasing expression and stimulating neutralizing antibodies;^{[13][52]} followed by a three prime untranslated region (bases 3880–4174) combined from *AES* and mtRNR1 selected for increased protein expression and mRNA stability^[53] and a poly(A) tail comprising 30 adenosine residues, a 10-nucleotide linker sequence, and 70 other adenosine residues (bases 4175–4284).^[51] The sequence contains no uridine residues; they are replaced by 1-methyl-3'-pseudouridine.^[51]

A look at the Spike, 'S', gene sequence







A look at the Spike, 'S', gene sequence

ATGTTTGTTTTCTTGTTTTATTGCCACTAGTCTCTAGTCAGTGTGTTAATCTTACAACCAGAACTCAATTACCCCCTGCATACACTAATTCTTTCACACGTGGTGTTTATTACC GAGGTTTGATAACCCTGTCCTACCATTTAATGATGGTGTTTATTTTGCTTCCACTGAGAAGTCTAACATAATAAGAGGCTGGATTTTTGGTACTACTTTAGATTCGAAGACCCAG TCCCTACTTATTGTTAATAACGCTACTAATGTTGTTATTAAAGTCTGTGAATTTCAATTTTGTAATGATCCATTTTTGGGTGTTTATTACCACAAAAACAACAACAAAGTTGGATGG AAAGTGAGTTCAGAGTTTATTCTAGTGCGAATAATTGCACTTTTGAATATGTCTCTCAGCCTTTTCTTATGGACCCTTGAAGGAAAACAGGGTAATTTCAAAAATCTTAGGGAATT TGTGTTTAAGAATATTGGTGGTTATTTTAAAATATATTCTAAGCACACGCCTATTAATTTAGTGCGTGATCTCCCTCAGGGTTTTTCGGCTTTAGAACCATTGGTAGATTTGCCA ATAGGTATTAACATCACTAGGTTTCAAACTTTACTTGCTTTACATAGAAGTTATTTGACTCCTGGTGATTCTTCTTCAGGTTGGACAGCTGGTGCTGCAGCTTATTATGTGGGTT ATCTTCAACCTAGGACTTTTCTATTAAAATATAATGAAAATGGAACCATTACAGATGCTGTAGACTGTGCACTTGACCCTCTCCAGAAACAAAGTGTACGTTGAAATCCTTCAC CTACTAAATTAAATGATCTCTGCTTTACTAATGTCTATGCAGATTCATTTGTAATTAGAGGTGATGAAGTCAGACAAATCGCTCCAGGGCAAACTGGAAAGATTGCTGATTATAA TTATAAATTACCAGATGATTTTACAGGCTGCGTTATAGCTTGGAATTCTAACAATCTTGATTCTAAGGTTGGTGGTAATTATAATTACCTGTATAGATTGTTTAGGAAGTCTAAT CTCAAACCTTTTGAGAGAGATATTTCAACTGAAATCTATCAGGCCGGTAGCACACCTTGTAATGGTGTTGAAGGTTTTAATTGTTACTTTCCTTTACAATCATATGGTTTCCAAC ATGTGTCAATTTCAACTTCAATGGTTTAACAGGCACAGGTGTTCTTACTGAGTCTAACAAAAAGTTTCTGCCTTTCCAACAATTTGGCAGAGACATTGCTGACACTACTGATGCT GTCCGTGATCCACAGACACTTGAGATTCTTGACATTACACCATGTTCTTTTGGTGGTGTCAGTGTTATAACACCAGGAACAAATACTTCTAACCAGGTTGCTGTTCTTTATCAGG ATGTTAACTGCACAGAAGTCCCTGTTGCTATTCATGCAGATCAACTTACTCCTACTTGGCGTGTTTATTCTACAGGTTCTAATGTTTTTCAAACACGTGCAGGCTGTTTAATAGG GGCTGAACATGTCAACAACTCATATGAGTGTGACATACCCATTGGTGCAGGTATATGCGCTAGTTATCAGACTCAGACTAATTCTCCTCGGCGGGCACGTAGTGTAGCTAGTCACA TCCATCATTGCCTACACTATGTCACTTGGTGCAGAAAATTCAGTTGCTTACTCTAATAACTCTATTGCCATACCCACAAATTTTACTATTAGTGTTACCACAGAAATTCTACCAG TGTCTATGACCAAGACATCAGTAGATTGTACAATGTACATTTGTGGTGATTCAACTGAATGCAGCAATCTTTTGTTGCAATATGGCAGTTTTTGTACACAATTAAACCGTGCTTT TTACCAGATCCATCAAAACCAAGCAAGAGGTCATTTATTGAAGATCTACTTTTCAACAAAGTGACACTTGCAGATGCTGGCTTCATCAAACAATATGGTGATTGCCTTGGTGATA TTGCTGCTAGAGACCTCATTTGTGCACAAAAGTTTAACGGCCTTACTGTTTTGCCACCTTTGCTCACAGATGAAATGATTGCTCAATACACTTCTGCACTGTTAGCGGGTACAAT CACTTCTGGTTGGACCTTTGGTGCAGGTGCTGCATTACAAATACCATTTGCTATGCAAATGGCTTATAGGTTTAATGGTATTGGAGTTACACAGAATGTTCTCTATGAGAACCAA AAATTGATTGCCAACCAATTTAATAGTGCTATTGGCAAAATTCAAGACTCACTTTCTTCCACAGCAAGTGCACTTGGAAAACTTCAAGATGTGGTCAACCAAAATGCACAAGCTT TAAACACGCTTGTTAAACAACTTAGCTCCAATTTTGGTGCAATTTCAAGTGTTTTAAATGATATCCTTTCACGTCTTGACAAAGTTGAGGCTGAAGTGCAAATTGATAGGTTGAT ACTTCACAACTGCTCCTGCCATTTGTCATGATGGAAAAGCACACTTTCCTCGTGAAGGTGTCTTTGTTTCAAATGGCACACACTGGTTTGTAACACAAAGGAATTTTTATGAACC ACAAATCATTACTACAGACAACACACTTTGTGTCTGGTAACTGTGATGTTGTAATAGGAATTGTCAACAACAACACAGTTTATGATCCTTTGCAACCTGAATTAGACTCATTCAAGGAG GAGTTAGATAAATATTTTAAGAATCATCACCAGATGTTGATTTAGGTGACATCTCTGGCATTAATGCTTCAGTTGTAAACATTCAAAAAGAAATTGACCGCCTCAATGAGG TTGCCAAGAATTTAAATGAATCTCTCATCGATCTCCAAGAACTTGGAAAGTATGAGCAGTATATAAAATGGCCATGGTACATTTGGCTAGGTTTTATAGCTGGCTTGATTGCCAT AGTAATGGTGACAATTATGCTTTGCTGTATGACCAGTTGCTGTAGTTGTCTCAAGGGCTGTTGTTCTTGTGGATCCTGCAAATTTGATGAAGACGACCACTCTGAGCCAGTGCTC AAAGGAGTCAAATTACATTACACATAA 3822



Why are there Us in this table?

Before a DNA sequence is translated into a protein, a copy is first made. This copy is made from RNA. In RNA, the nucleotide "Uracil" replaces "Thymine". Uracil and Thymine are both chemically and structurally very similar.

Comp 555 - Spring 2021

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Maping to Amino Acid Residues



In [138]:	▶ codon = { # Maps an RNA triplet of nucelotides to a 1-letter Amino Acid Abbrevation							
	"AAA": 'K', "AAG": 'K', "AAC": 'N', "AAT": 'N',							
	"AGA": 'R', "AGG": 'R', "AGC": 'S', "AGT": 'S',							
	"ACA": 'T', "ACG": 'T', "ACC": 'T', "ACT": 'T',							
	"ATA": 'I', "ATG": 'M', "ATC": 'I', "ATT": 'I',							
	"GAA": 'E', "GAG": 'E', "GAC": 'D', "GAT": 'D',							
	"GGA": 'G', "GGG": 'G', "GGC": 'G', "GGT": 'G',							
	"GCA": 'A', "GCG": 'A', "GCC": 'A', "GCT": 'A',							
	"GTA": 'V', "GTG": 'V', "GTC": 'V', "GTT": 'V',							
	"CAA": '0', "CAG": '0', "CAC": 'H', "CAT": 'H',							
	"CGA": 'R', "CGG": 'R', "CGC": 'R', "CGT": 'R',							
	"CCA": 'P'. "CCG": 'P'. "CCC": 'P'. "CCT": 'P'.							
	"CTA": 'L'. "CTG": 'L'. "CTC": 'L'. "CTT": 'L'.							
	"TAA": '*', "TAG": '*', "TAC": 'Y', "TAT": 'Y',							
	"TGA" '*' "TGG" 'W' "TGC" 'C' "TGT" 'C'							
	"TCA": 'S', "TCG": 'S', "TCC": 'S', "TCT": 'S',							
	"TTA": 'L' "TTG": 'L' "TTC": 'E' "TTT": 'E'							
	L							
	AminoAcid = { # Maps 1-letter Amino Acid Abbrevations to their full name							
	'A': 'Alanina' 'C': 'Cystaina' 'D': 'Aspartic acid' 'E': 'Glutamic acid' 'E': 'Dhanylalanina'							
	A. Alanine, C. Cysterne, D. Aspartic actu, E. Stutamic actu, F. Phenylalanine,							
	Why Apparential I Dealine, I. ISofeucine, K. Lysine, L. Leucine, M. Methionine,							
	N: Asparagine, P: Proiine, V: Giucamine, R: Arginine, S: Serine,							
	T: Theronine, V: Vaiine, W: Tryptophan, Y: Tyrosine, ": STOP							
	1							

"Spike" as a peptide sequence



In [139]:

peptide = ''.join([codon[spike[i:i+3]] for i in range(0,len(spike),3)])
print(peptide)

MEVFLVLLPLVSSQCVNLTTRTQLPPAYTNSFTRGVYYPDKVFRSSVLHSTQDLFLPFFSNVTWFHAIHVSGTNGTKRFDNPVLPFNDGVYFASTEKSNIIRGWIFGTTLDSKTQSLL IVNNATNVVIKVCEFQFCNDPFLGVYYHKNNKSWMESEFRVYSSANNCTFEYVSQPFLMDLEGKQGNFKNLREFVFKNIDGYFKIYSKHTPINLVRDLPQGFSALEPLVDLPIGINIT RFQTLLALHRSYLTPGDSSSGWTAGAAAYYVGYLQPRTFLLKYNENGTITDAVDCALDPLSETKCTLKSFTVEKGIYQTSNFRVQPTESIVRFPNITNLCPFGEVFNATRFASVYAWN RKRISNCVADYSVLYNSASFSTFKCYGVSPTKLNDLCFTNVYADSFVIRGDEVRQIAPGQTGKIADYNYKLPDDFTGCVIAWNSNNLDSKVGGNYNYLYRLFRKSNLKPFERDISTEI YQAGSTPCNGVEGFNCYFPLQSYGFQPTNGVGYQPYRVVVLSFELLHAPATVCGPKKSTNLVKNKCVNFNFNGLTGTGVLTESNKKFLPFQQFGRDIADTTDAVRDPQTLEILDITPC SFGGVSVITPGTNTSNQVAVLYQDVNCTEVPVAIHADQLTPTWRVYSTGSNVFQTRAGCLIGAEHVNNSYECDIPIGAGICASYQTQTNSPRRARSVASQSIIAYTMSLGAENSVAYS NNSIAIPTNFTISVTTEILPVSMTKTSVDCTMYICGDSTECSNLLLQYGSFCTQLNRALTGIAVEQDKNTQEVFAQVKQIYKTPPIKDFGGFNFSQILPDPSKPSKRSFIEDLLFNKV TLADAGFIKQYGDCLGDIAARDLICAQKFNGLTVLPPLLTDEMIAQYTSALLAGTITSGWTFGAGAALQIPFAMQMAYRFNGIGVTQNVLYENQKLIANQFNSAIGKIQDSLSSTASA LGKLQDVVNQNAQALNTLVKQLSSNFGAISSVLNDILSRLDKVEAEVQIDRLITGRLQSLQTYVTQQLIRAAEIRASANLAATKMSECVLGQSKRVDFCGKGYHLMSFPQSAPHGVVF LHVTYVPAQEKNFTTAPAICHDGKAHFPREGVFVSNGTHWFVTQRNFYEPQIITTDNTFVSGNCDVVIGIVNNTVYDPLQPELDSFKEELDKYFKNHTSPDVDLGDISGINASVVNIQ KEIDRLNEVAKNLNESLIDLQELGKYEQYIKWPWIWLGFIAGLIAIVMVTIMLCCMTSCCSCLKGCCSCGSCCKFDEDDSEPVLKGVKLHYT*

Next time



We'll go hunting for virus fossils.

