Applied Comparative Genomics

Michael Schatz

August 29, 2022

Lecture 1: Course Overview

Welcome!

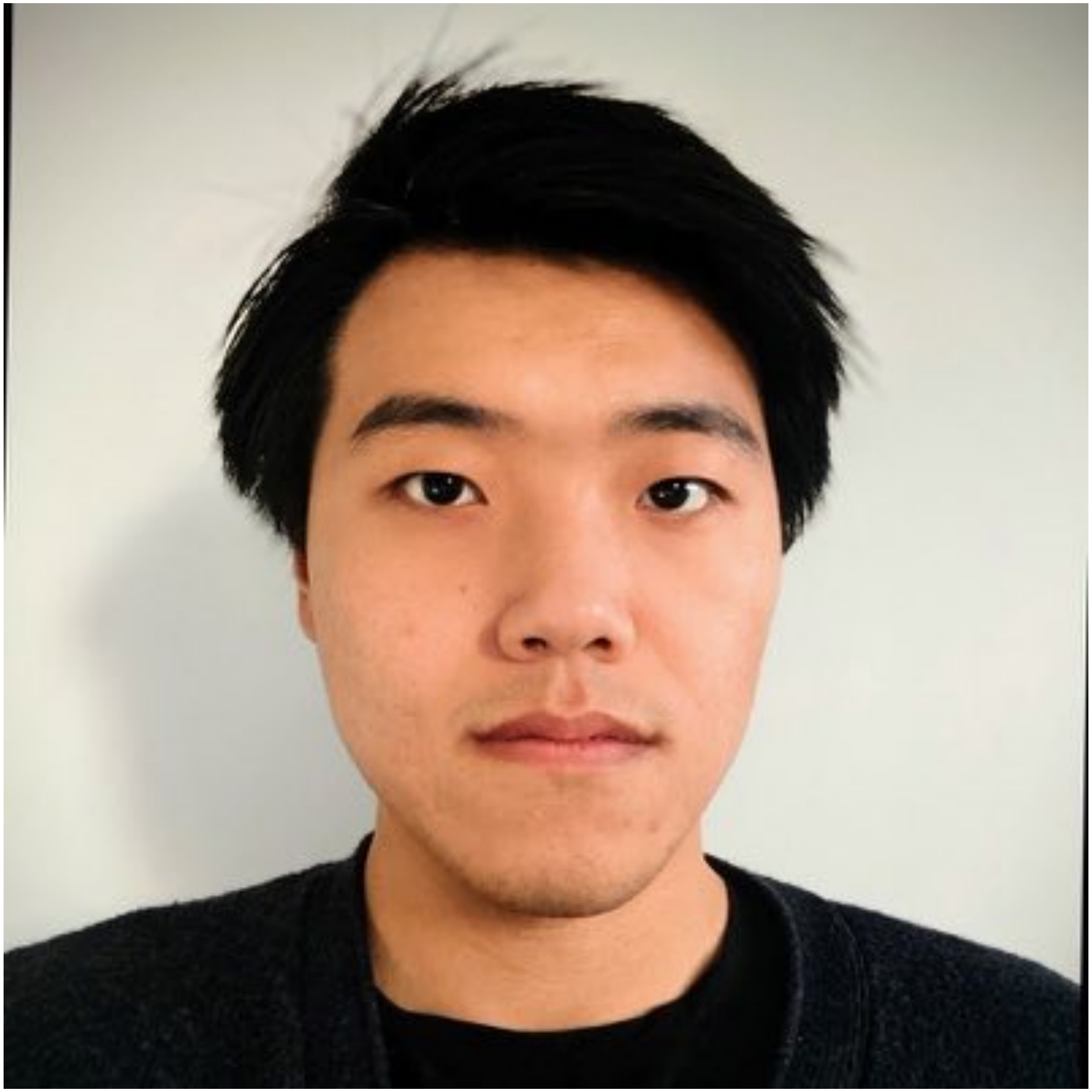
The primary goal of the course is for students to be grounded in theory and leave the course empowered to conduct independent genomic analyses.

- We will study the leading computational and quantitative approaches for comparing and analyzing genomes starting from raw sequencing data.
- The course will focus on human genomics and human medical applications, but the techniques will be broadly applicable across the tree of life.
- The topics will include genome assembly & comparative genomics, variant identification & analysis, gene expression & regulation, personal genome analysis, and cancer genomics.

Course Webpage:	https://github.com/schatzlab/appliedgenomics2022
Course Discussions:	http://piazza.com
Class Hours:	Mon + Wed @ 1:30p – 2:45p, Gilman 17
Schatz Office Hours:	TBD and by appointment
Ni Office Hours:	TBD and by appointment

Please try Piazza first!

TA: Bohan Ni



Prerequisites and Resources

Prerequisites

- No formal course requirements
- Access to an Apple or Linux Machine, or Install VirtualBox
- Familiarity with the Unix command line for exercises
  - bash, ls, grep, sed, + install published genomics tools
- Familiarity with a major programming language for project
  - C/C++, Java, R, Perl, Python

Primary Texts

- None! We will be studying primary research papers

Other Resources:

- Google, SEQanswers, Biostars, StackOverflow

- Applied Computational Genomics Course at UU: Spring 2018/2020
- https://github.com/quinlan-lab/applied-computational-genomics

- Ben Langmead's teaching materials:
- http://www.langmead-lab.org/teaching-materials/

Grading Policies

Assessments:

- 5 Assignments: 30% Due at 11:59pm a week later

  Practice using the tools we are discussing

- 1 Exam: 30% Take Home (Tentatively Nov 2)

  Assess your performance, focusing on the methods

- 1 Class Project: 40% Presented last week of class

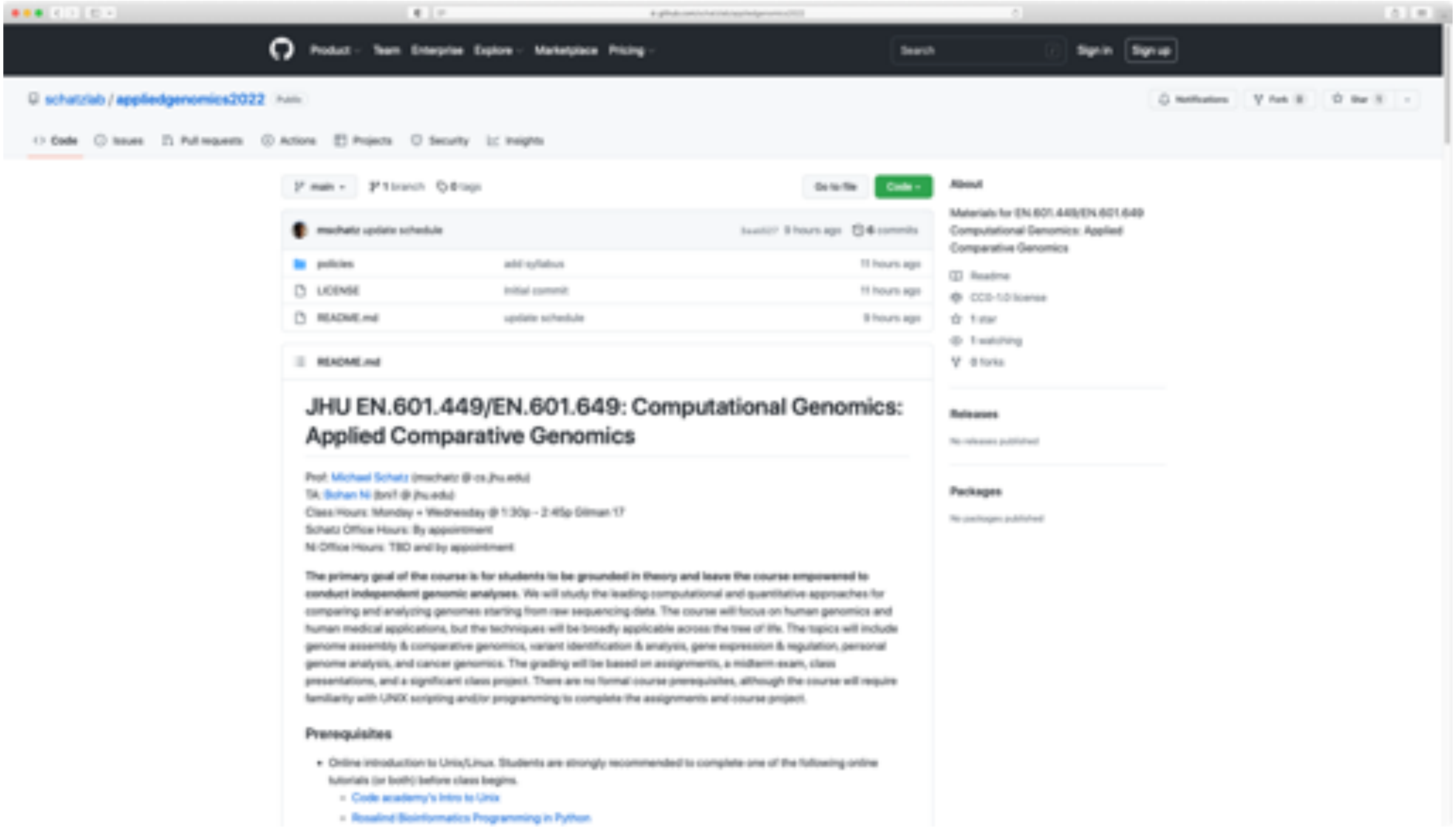
  Significant project developing a novel analysis/method

- In-class Participation: Not graded, but there to help you!

Policies:

- Scores assigned relative to the highest points awarded
- Automated testing and grading of assignments
- ***Late Days:***
  - A total of 96 hours (24 x 4) can be used to extend the deadline for assignments, but not the class project, without any penalty; after that time assignments will not be accepted

Course Webpage



mschatz update schedule · 5aa6027 9 hours ago · 6 commits

policies	add syllabus	11 hours ago
LICENSE	Initial commit	11 hours ago
README.md	update schedule	9 hours ago

README.md

JHU EN.601.449/EN.601.649: Computational Genomics: Applied Comparative Genomics

Prof: Michael Schatz (mschatz @ cs.jhu.edu)
TA: Bohan Ni (bni1 @ jhu.edu)
Class Hours: Monday + Wednesday @ 1:30p - 2:45p Gilman 17
Schatz Office Hours: By appointment
Ni Office Hours: TBD and by appointment

The primary goal of the course is for students to be grounded in theory and leave the course empowered to conduct independent genomic analyses. We will study the leading computational and quantitative approaches for comparing and analyzing genomes starting from raw sequencing data. The course will focus on human genomics and human medical applications, but the techniques will be broadly applicable across the tree of life. The topics will include genome assembly & comparative genomics, variant identification & analysis, gene expression & regulation, personal genome analysis, and cancer genomics. The grading will be based on assignments, a midterm exam, class presentations, and a significant class project. There are no formal course prerequisites, although the course will require familiarity with UNIX scripting and/or programming to complete the assignments and course project.

Prerequisites

- Online introduction to Unix/Linux. Students are strongly recommended to complete one of the following online tutorials (or both) before class begins.
  - Code academy's Intro to Unix
  - Rosalind Bioinformatics Programming in Python

About

Materials for EN.601.449/EN.601.649 Computational Genomics: Applied Comparative Genomics

Readme
CC0-1.0 license
1 star
1 watching
0 forks

Releases

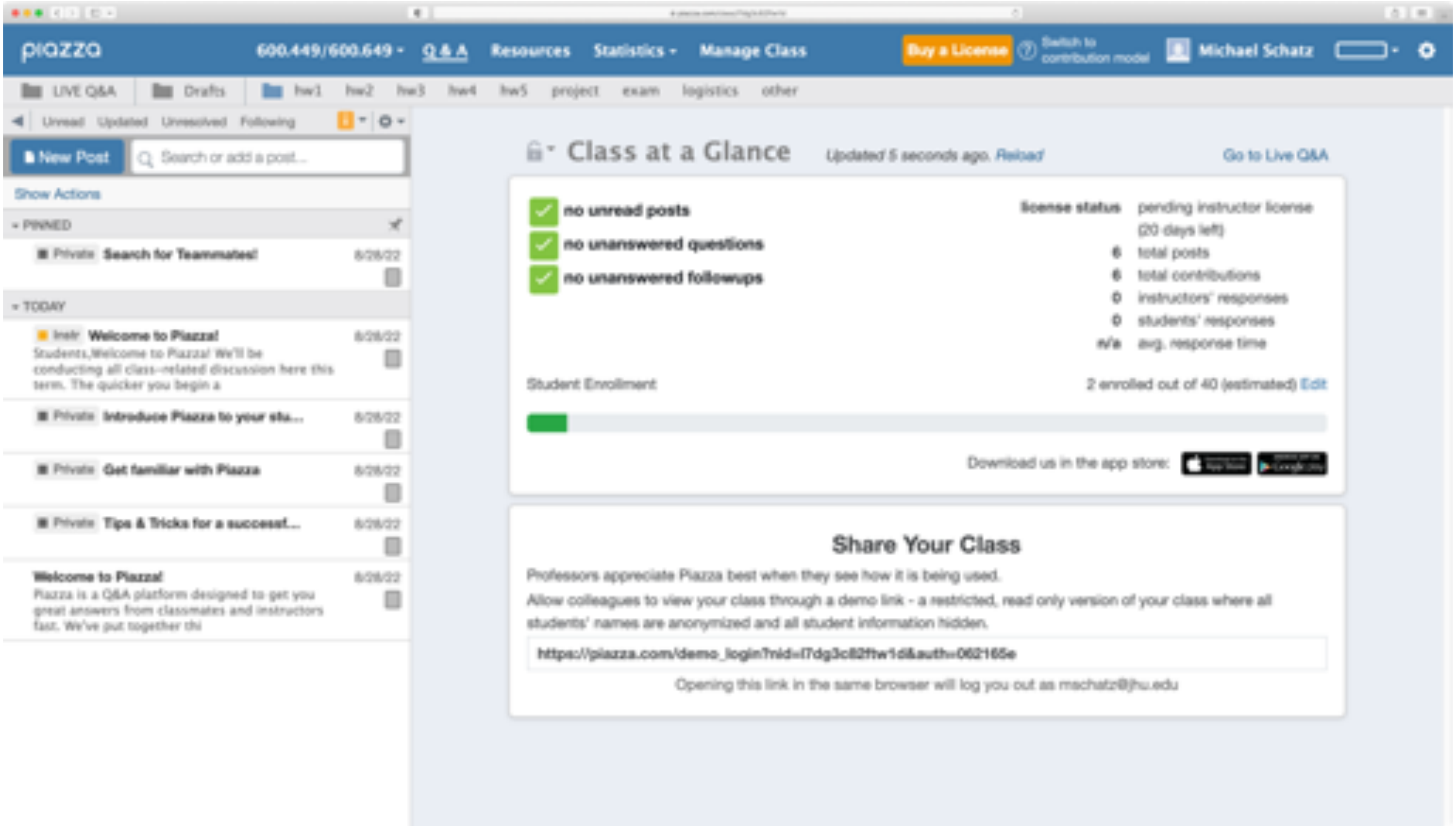
No releases published

Packages

No packages published

https://github.com/schatzlab/appliedgenomics2022

Piazza



PINNED

Private **Search for Teammates!** 8/28/22

TODAY

Instr **Welcome to Piazza!** 8/28/22
Students,Welcome to Piazza! We'll be conducting all class-related discussion here this term. The quicker you begin a

Private **Introduce Piazza to your stu...** 8/28/22

Private **Get familiar with Piazza** 8/28/22

Private **Tips & Tricks for a successf...** 8/28/22

Welcome to Piazza! 8/28/22
Piazza is a Q&A platform designed to get you great answers from classmates and instructors fast. We've put together thi

Class at a Glance

Updated 5 seconds ago. Reload

✓ **no unread posts**

✓ **no unanswered questions**

✓ **no unanswered followups**

license status	pending instructor license (20 days left)
6	total posts
6	total contributions
0	instructors' responses
0	students' responses
n/a	avg. response time

Student Enrollment — 2 enrolled out of 40 (estimated) Edit

Share Your Class

Professors appreciate Piazza best when they see how it is being used.

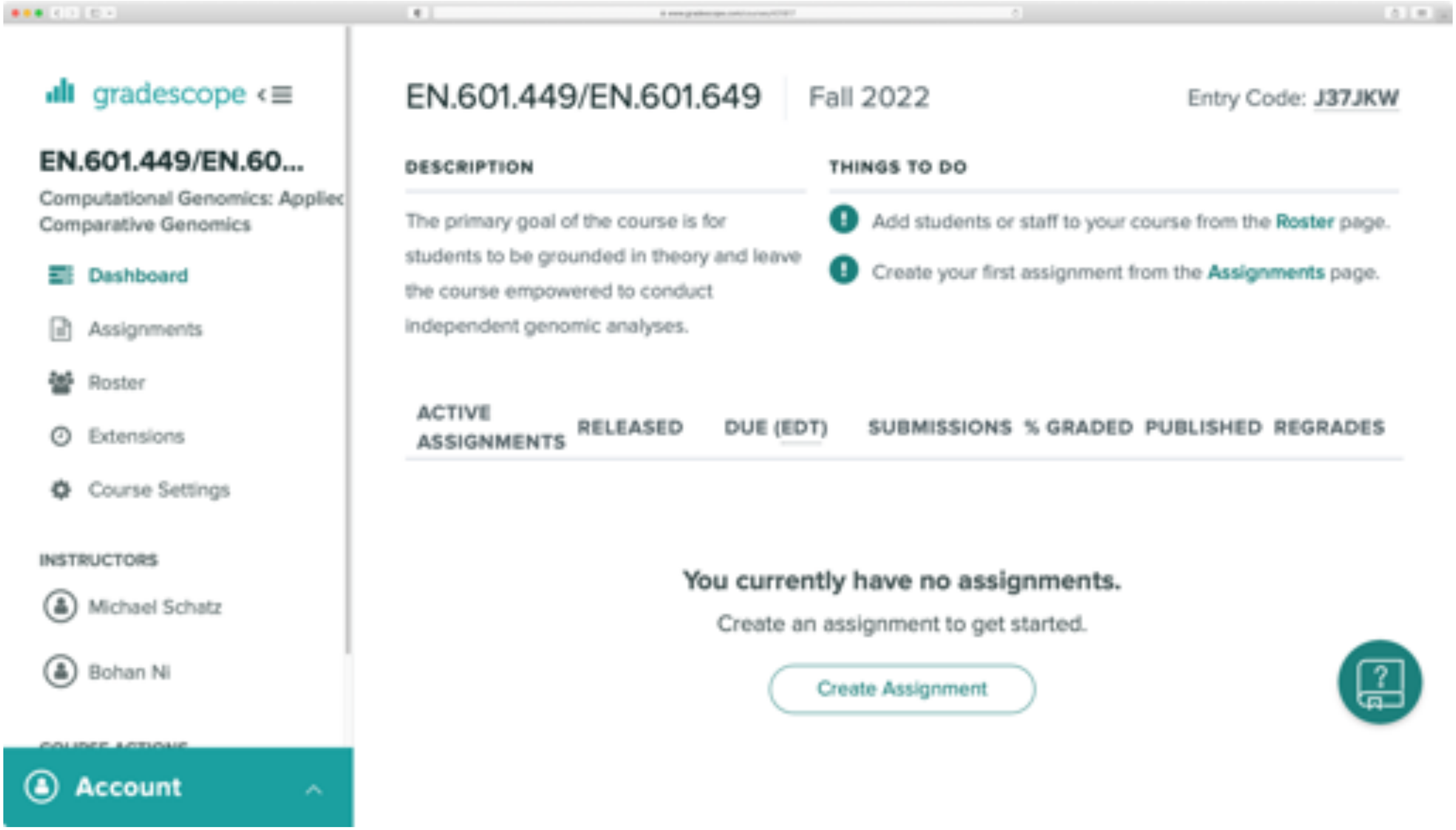
Allow colleagues to view your class through a demo link - a restricted, read only version of your class where all students' names are anonymized and all student information hidden.

https://piazza.com/demo_login?nid=l7dg3c82ftw1d&auth=062165e

Opening this link in the same browser will log you out as mschatz@jhu.edu

https://piazza.com/class/l7dg3c82ftw1d/

GradeScope



gradescope

EN.601.449/EN.60...

Computational Genomics: Applied Comparative Genomics

INSTRUCTORS

Michael Schatz

Bohan Ni

EN.601.449/EN.601.649 Fall 2022 Entry Code: J37JKW

DESCRIPTION

The primary goal of the course is for students to be grounded in theory and leave the course empowered to conduct independent genomic analyses.

THINGS TO DO

- Add students or staff to your course from the Roster page.
- Create your first assignment from the Assignments page.

ACTIVE ASSIGNMENTS	RELEASED	DUE (EDT)	SUBMISSIONS	% GRADED	PUBLISHED	REGRADES

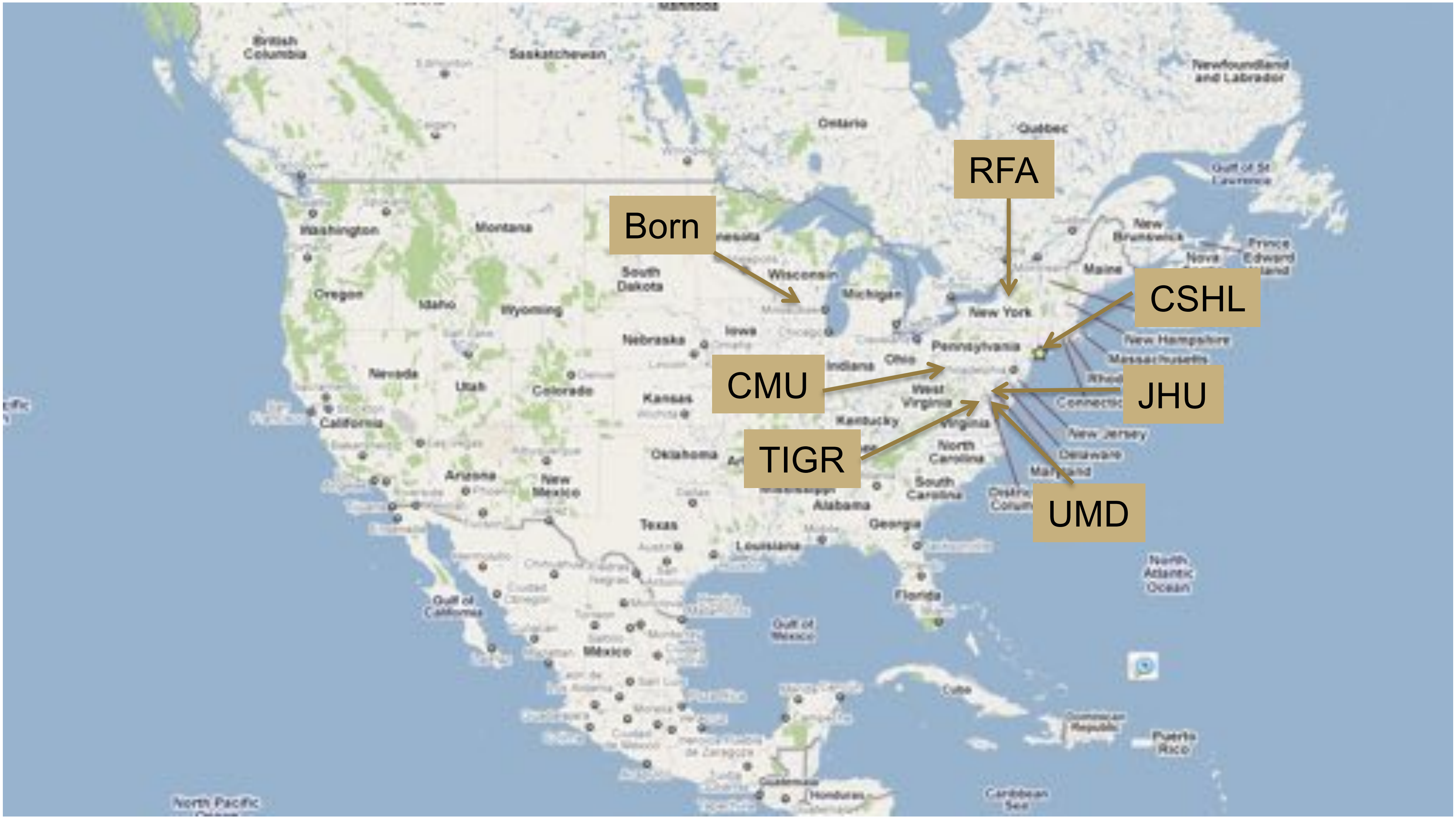
You currently have no assignments.

Create an assignment to get started.

https://www.gradescope.com/

Entry Code: **J37JKW**

A Little About Me



Born

RFA

CSHL

CMU

JHU

TIGR

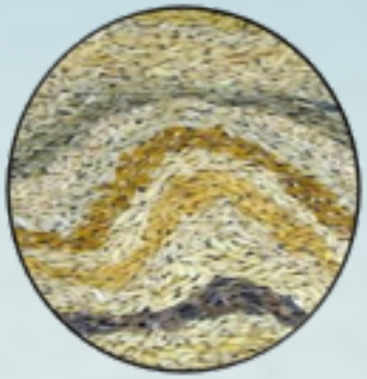
UMD

Schatzlab Overview

Human Genetics

Role of mutations in disease

Nurk et al. (2022)
Aganezov *et al.* (2020)



Agricultural Genomics

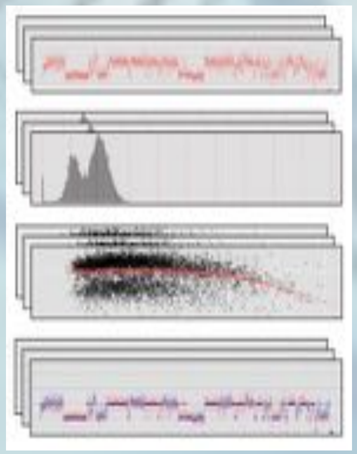
Genomes & Transcriptomes

Alonge *et al.* (2020)
Soyk *et al.* (2019)

Algorithmics & Systems Research

Ultra-large scale biocomputing

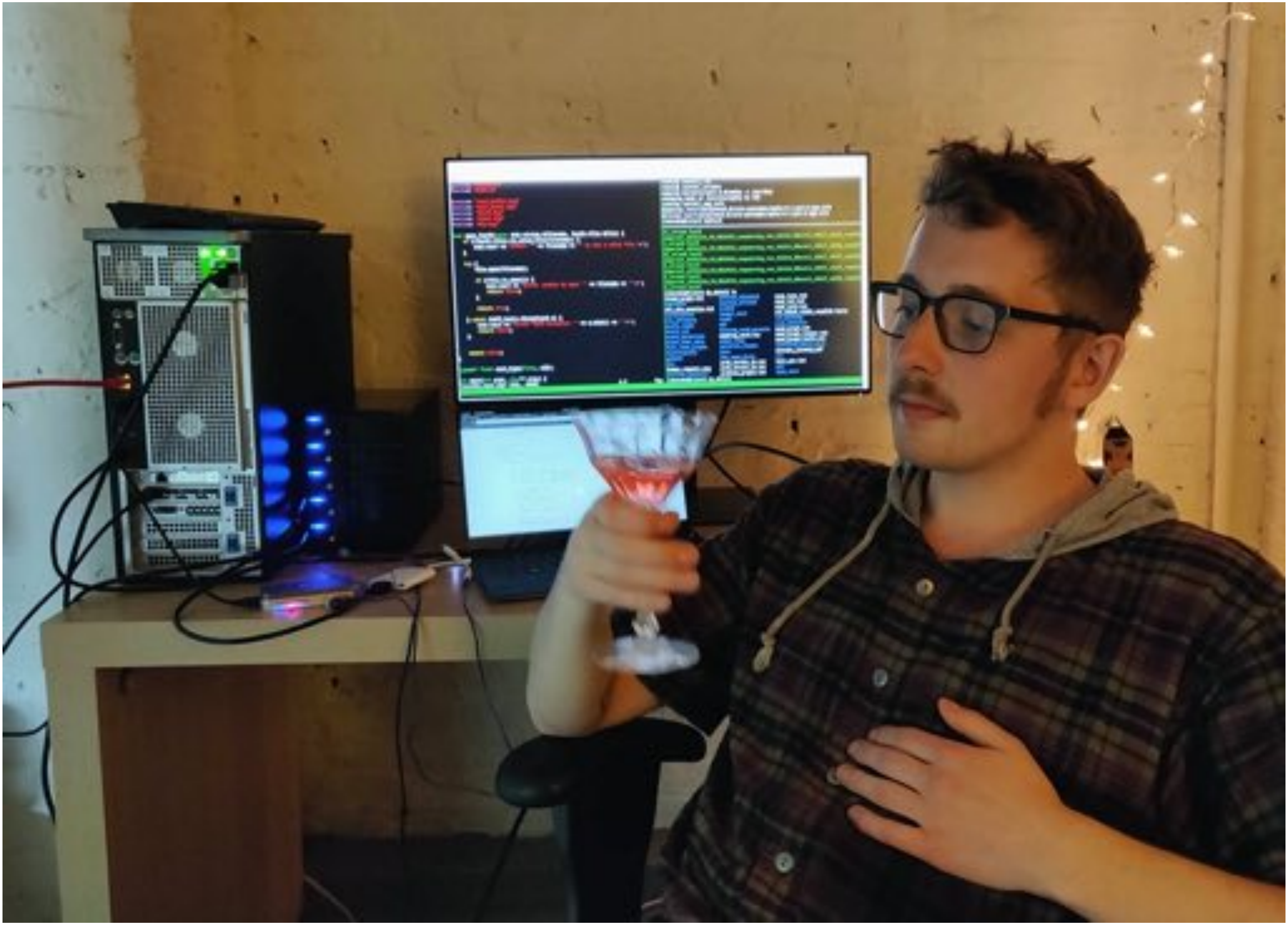
Schatz et al. (2022)
Kirsche *et al.* (2020)

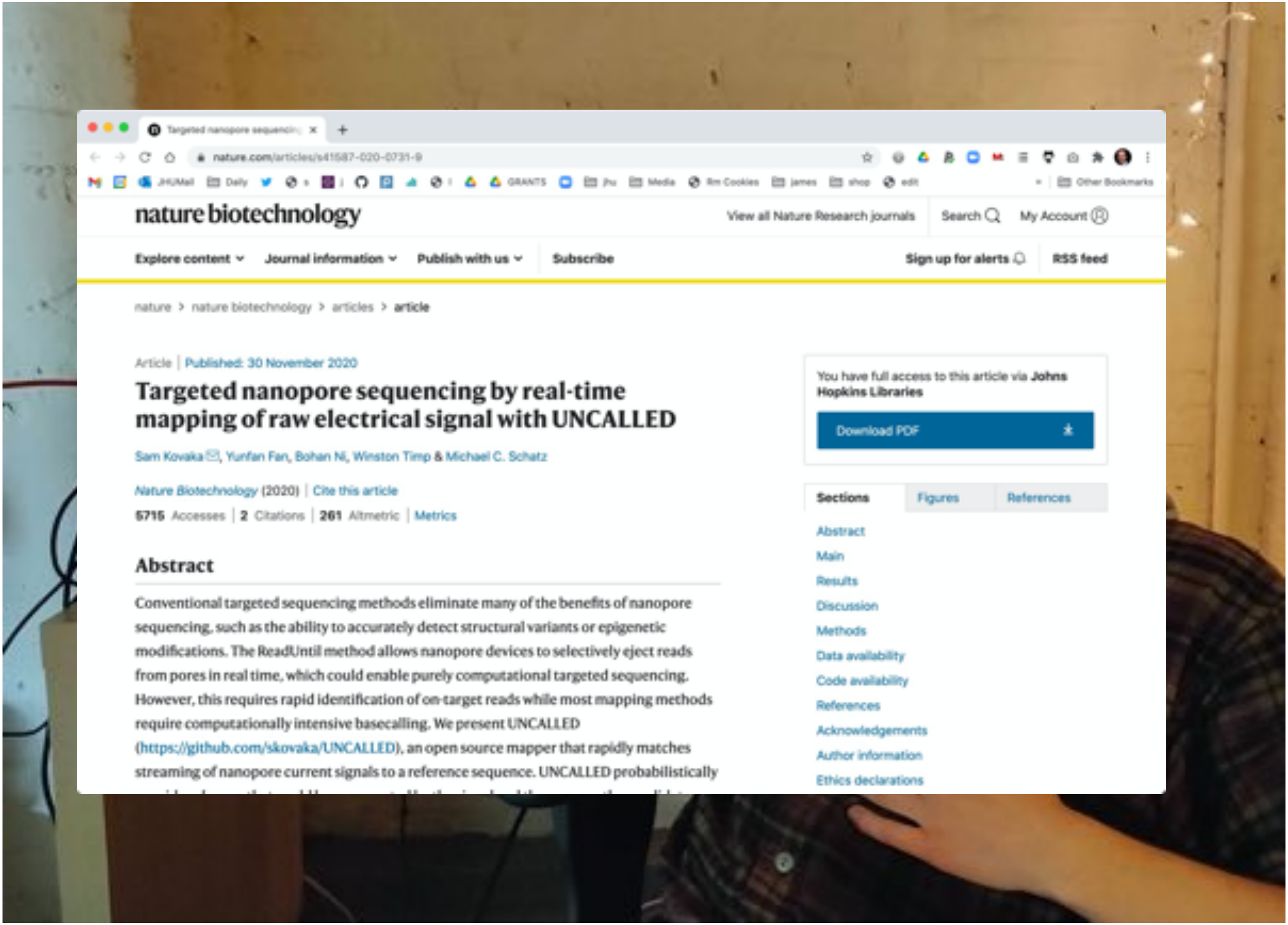


Biotechnology Development

Single Cell + Single Molecule Sequencing

Kovaka *et al.* (2020)
Sedlazeck *et al.* (2018)





Article | Published: 30 November 2020

Targeted nanopore sequencing by real-time mapping of raw electrical signal with UNCALLED

Sam Kovaka, Yunfan Fan, Bohan Ni, Winston Timp & Michael C. Schatz

Nature Biotechnology (2020) | Cite this article

5715 Accesses | 2 Citations | 261 Altmetric | Metrics

You have full access to this article via **Johns Hopkins Libraries**

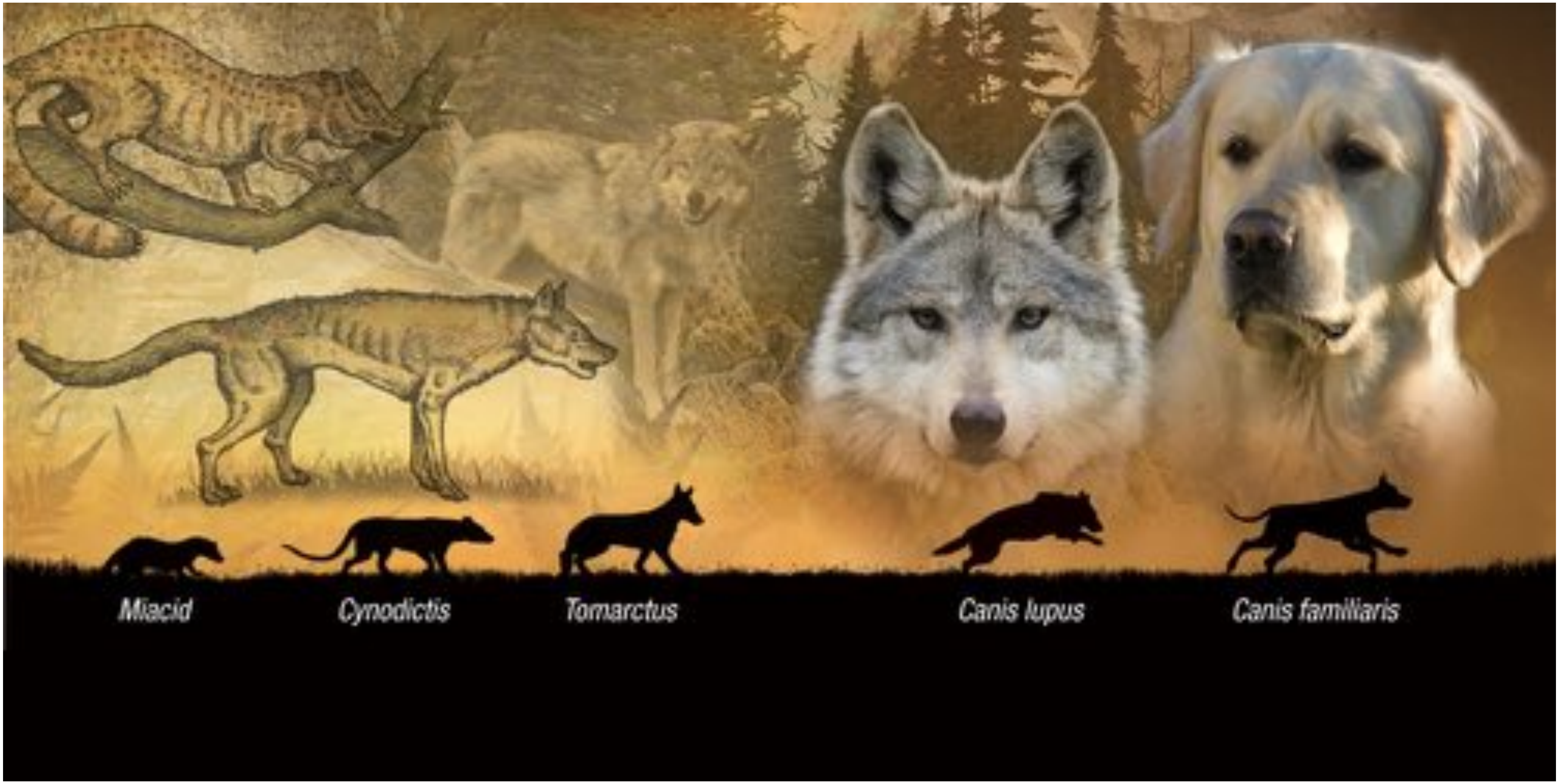
Abstract

Conventional targeted sequencing methods eliminate many of the benefits of nanopore sequencing, such as the ability to accurately detect structural variants or epigenetic modifications. The ReadUntil method allows nanopore devices to selectively eject reads from pores in real time, which could enable purely computational targeted sequencing. However, this requires rapid identification of on-target reads while most mapping methods require computationally intensive basecalling. We present UNCALLED (https://github.com/skovaka/UNCALLED), an open source mapper that rapidly matches streaming of nanopore current signals to a reference sequence. UNCALLED probabilistically

Earliest Genomics

Any Guesses?

Earliest Genomics

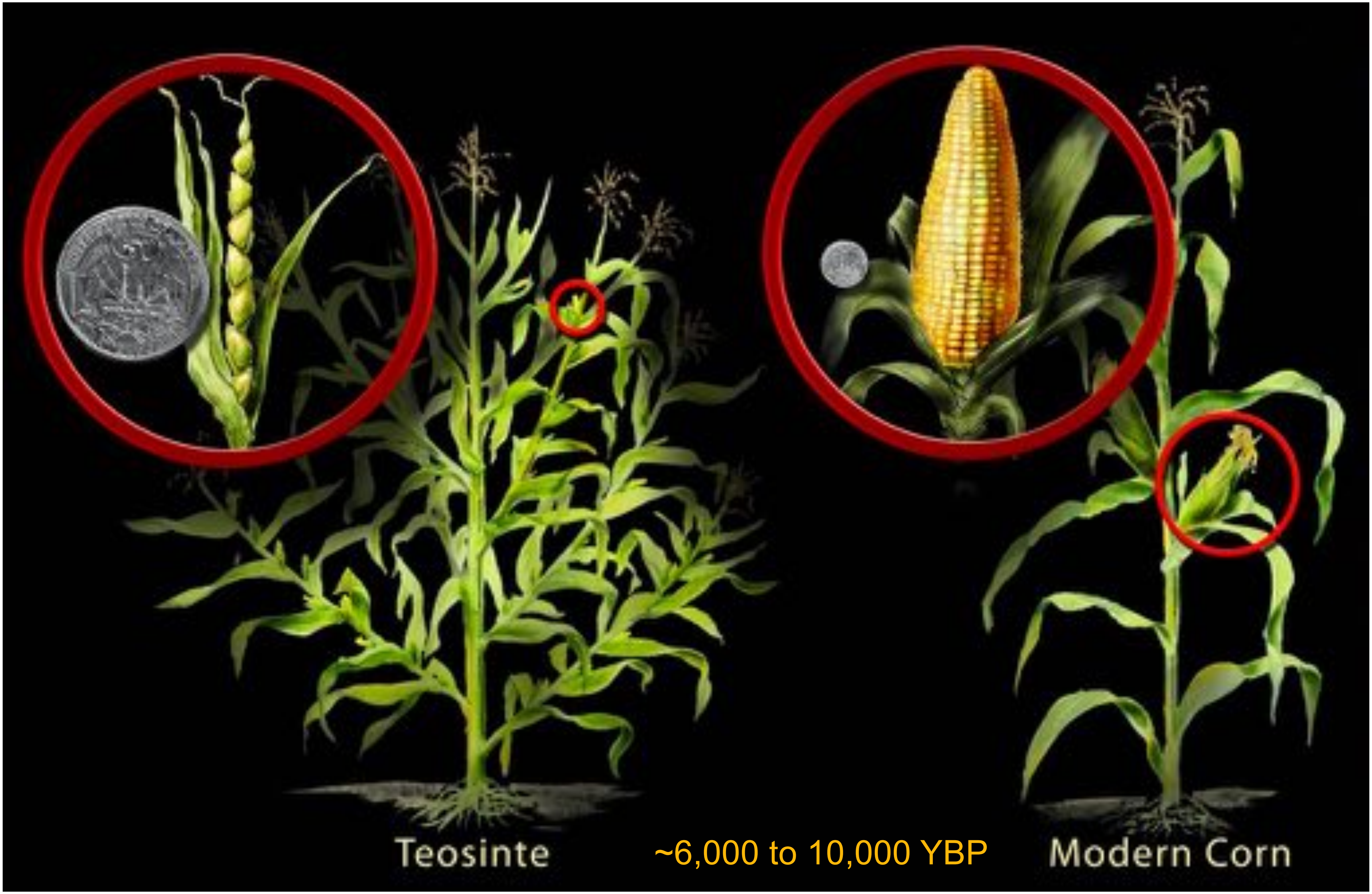


15,000 to 35,000 YBP

Earliest Genomics

~1,000 to 10,000 YBP

Earliest Genomics



~6,000 to 10,000 YBP

Angiosperms (Flowering Plants)

~130 Ma

Discovery of Chromosomes

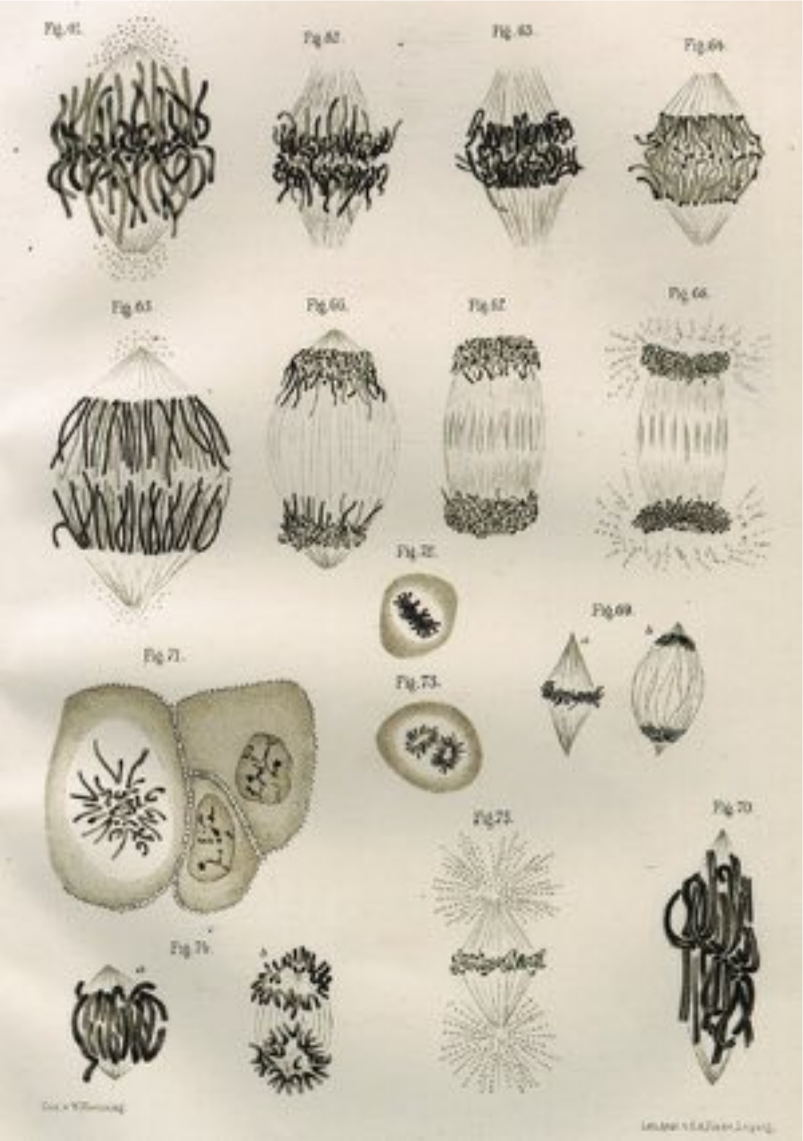
By the mid-1800s, microscopes were powerful enough to observe the presence of unusual structures called “chromosomes” that seemed to play an important role during cell division.

It was only possible to see the chromosomes unless appropriate stains were used

“Chromosome” comes from the Greek words meaning “color body”

Today, we have much higher resolution microscopes, and a much richer varieties of dies and dying techniques so that we can visualize particular sequence elements.

When you see something unexpected that you think might be interesting, give it a name

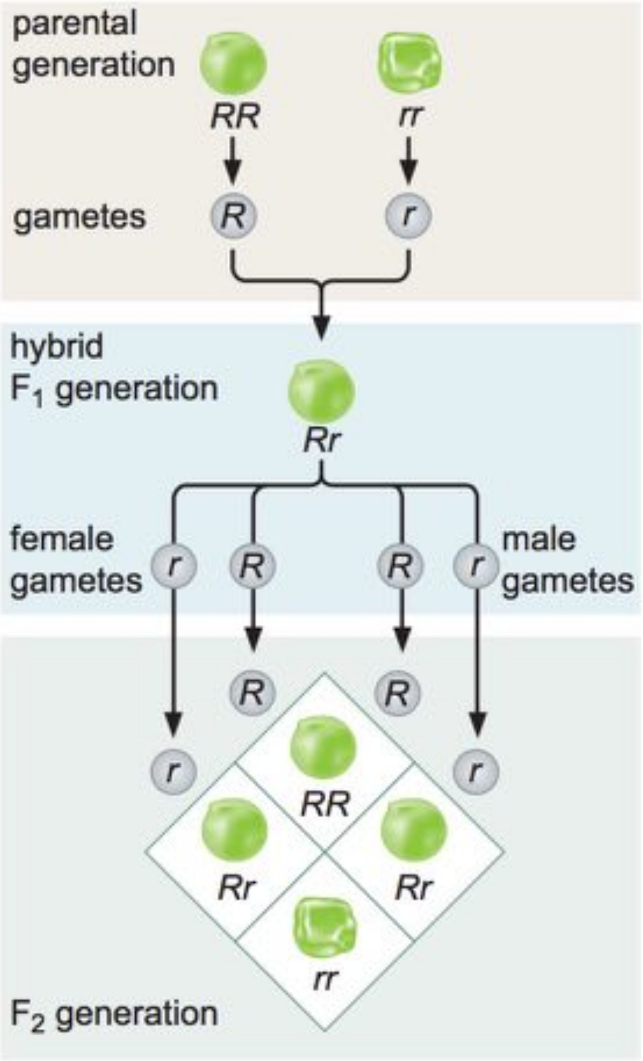


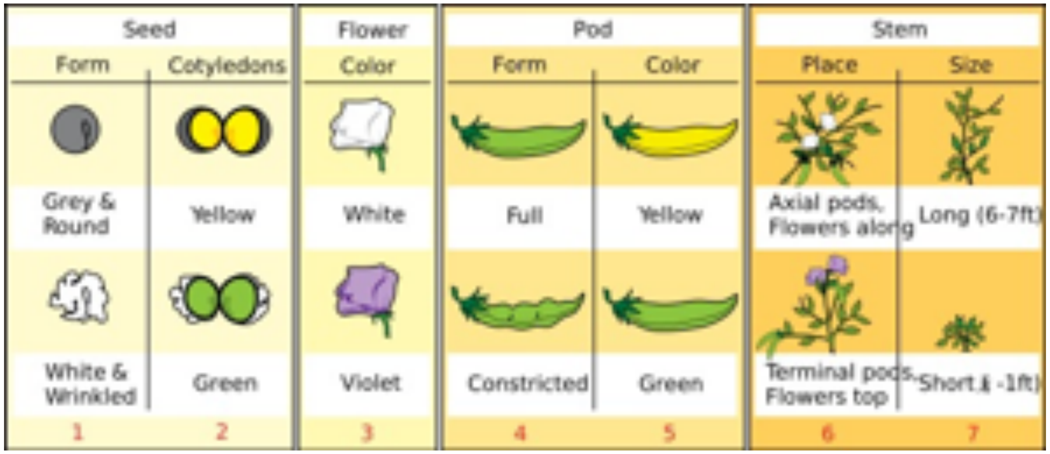
Drawing of mitosis by Walther Flemming.
Flemming, W. Zellsubstanz, Kern und Zelltheilung (F. C. W. Vogel, Leipzig, 1882).

The “first” quantitative biologist

Any Guesses?

Laws of Inheritance

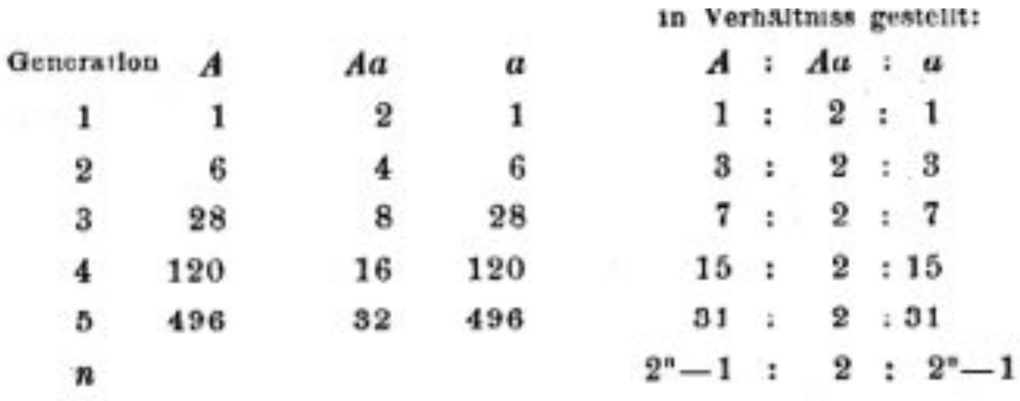




Seed		Flower	Pod		Stem	
Form	Cotyledons	Color	Form	Color	Place	Size
Grey & Round	Yellow	White	Full	Yellow	Axial pods, Flowers along	Long (6-7ft)
White & Wrinkled	Green	Violet	Constricted	Green	Terminal pods, Flowers top	Short (¾ -1ft)
1	2	3	4	5	6	7

http://en.wikipedia.org/wiki/Experiments_on_Plant_Hybridization

Observations of 29,000 pea plants and 7 traits



				in Verhältniss gestellt:
Generation	A	Aa	a	A : Aa : a
1	1	2	1	1 : 2 : 1
2	6	4	6	3 : 2 : 3
3	28	8	28	7 : 2 : 7
4	120	16	120	15 : 2 : 15
5	496	32	496	31 : 2 : 31
n				2^n-1 : 2 : 2^n-1

Versuche über Pflanzen-Hybriden. Verh. Naturforsch (Experiments in Plant Hybridization)
Mendel, G. (1866). Ver. Brünn 4: 3–47 (in English in 1901, J. R. Hortic. Soc. 26: 1–32).

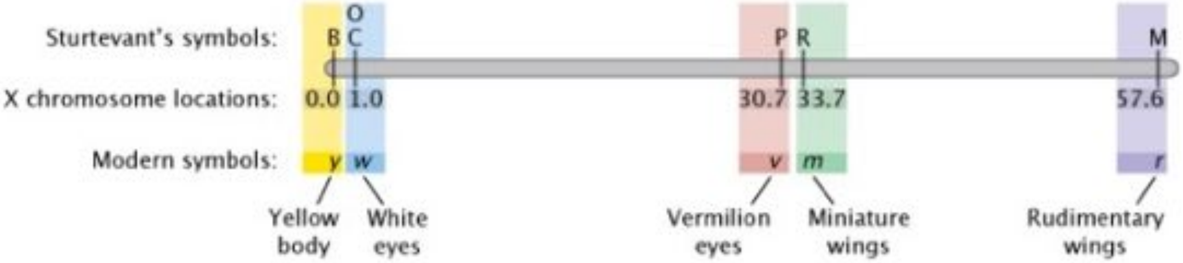
The first genetic map

Mendel's Second Law (The Law of Independent Assortment) states alleles of one gene sort into gametes independently of the alleles of another gene: ***Pr(smooth/wrinkle) is independent of Pr(yellow/green)***

Morgan and Sturtevant noticed that the probability of having one trait given another was **not** always 50/50– those traits are ***genetically linked***

http://www.caltech.edu/news/first-genetic-linkage-map-38798

Sturtevant realized the probabilities of co-occurrences could be explained if those alleles were arranged on a linear fashion: traits that are most commonly observed together must be locates closest together

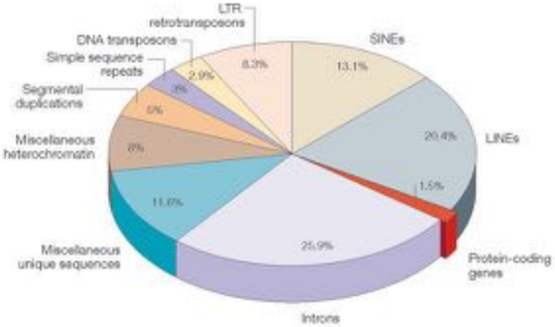


The Linear Arrangement of Six Sex-Linked Factors in Drosophila as shown by their mode of Association
Sturtevant, A. H. (1913) *Journal of Experimental Zoology*, 14: 43-59

Jumping Genes

Previously, genes were considered to be stable entities arranged in an orderly linear pattern on chromosomes, like beads on a string

Careful breeding and cytogenetics revealed that some elements can move (cut-and-paste, DNA transposons) or copy itself (copy-and-paste, retrotransposons)



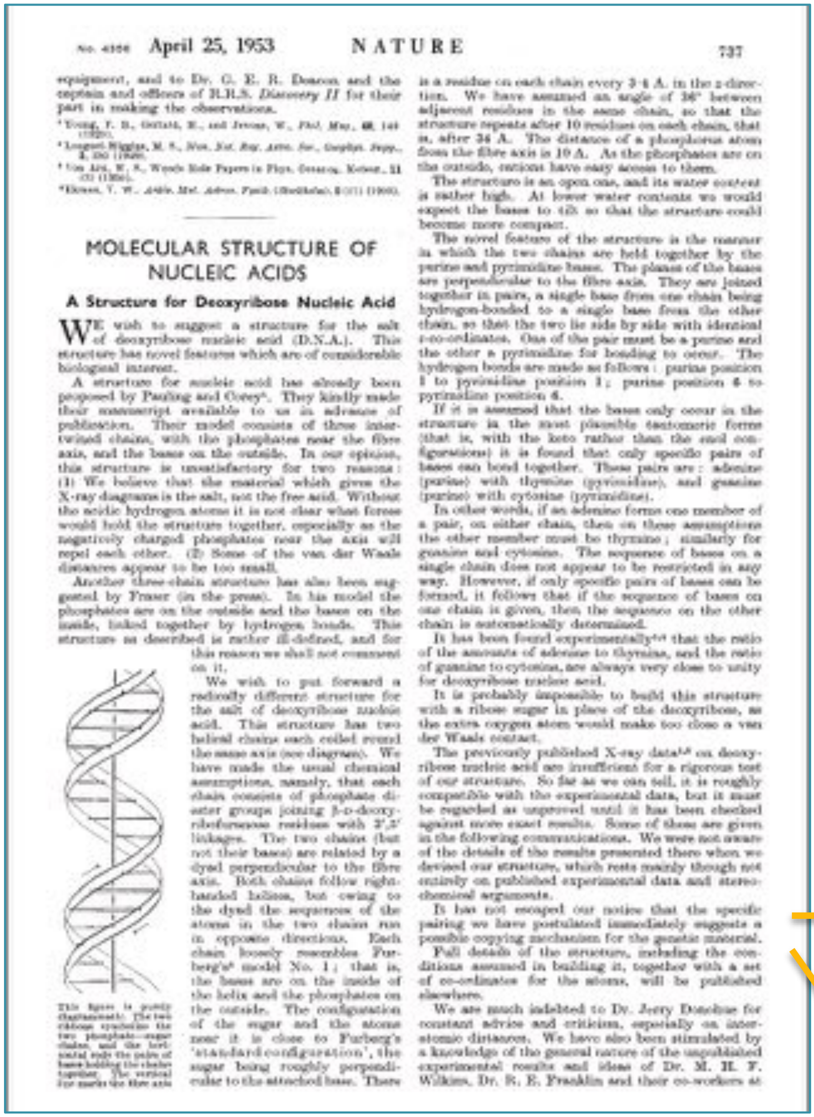
(Gregory, 2005, Nature Reviews Genetics)

(Much) later analysis revealed that nearly 50% of the human genome is composed of transposable elements, including LINE and SINE elements (long/short interspersed nuclear elements) which can occur in 100k to 1M copies

"The genome is a graveyard of ancient transposons"

The origin and behavior of mutable loci in maize.
McClintock, B. (1950) *PNAS.* 36(6):344–355.
Nobel Prize in Physiology or Medicine in 1983

Discovery of the Double Helix



No. 4356 April 25, 1953 NATURE 737

equipment, and to Dr. G. E. R. Deacon and the captain and officers of R.R.S. *Discovery II* for their part in making the observations.

[1] Young, F. B., Gerrard, H., and Jevons, W., *Phil. Mag.*, **40**, 149 (1920).

[2] Longuet-Higgins, M. S., *Mon. Not. Roy. Astro. Soc., Geophys. Supp.*, **5**, 285 (1949).

[3] Von Arx, W. S., Woods Hole Papers in Phys. Oceanog. Meteor., **11** (3) (1950).

[4] Ekman, V. W., *Arkiv. Mat. Astron. Fysik.* (Stockholm), **2** (11) (1905).

MOLECULAR STRUCTURE OF NUCLEIC ACIDS

A Structure for Deoxyribose Nucleic Acid

WE wish to suggest a structure for the salt of deoxyribose nucleic acid (D.N.A.). This structure has novel features which are of considerable biological interest.

A structure for nucleic acid has already been proposed by Pauling and Corey[1]. They kindly made their manuscript available to us in advance of publication. Their model consists of three intertwined chains, with the phosphates near the fibre axis, and the bases on the outside. In our opinion, this structure is unsatisfactory for two reasons: (1) We believe that the material which gives the X-ray diagrams is the salt, not the free acid. Without the acidic hydrogen atoms it is not clear what forces would hold the structure together, especially as the negatively charged phosphates near the axis will repel each other. (2) Some of the van der Waals distances appear to be too small.

Another three-chain structure has also been suggested by Fraser (in the press). In his model the phosphates are on the outside and the bases on the inside, linked together by hydrogen bonds. This structure as described is rather ill-defined, and for this reason we shall not comment on it.

We wish to put forward a radically different structure for the salt of deoxyribose nucleic acid. This structure has two helical chains each coiled round the same axis (see diagram). We have made the usual chemical assumptions, namely, that each chain consists of phosphate diester groups joining β-D-deoxyribofuranose residues with 3′,5′ linkages. The two chains (but not their bases) are related by a dyad perpendicular to the fibre axis. Both chains follow right-handed helices, but owing to the dyad the sequences of the atoms in the two chains run in opposite directions. Each chain loosely resembles Furberg's[2] model No. 1; that is, the bases are on the inside of the helix and the phosphates on the outside. The configuration of the sugar and the atoms near it is close to Furberg's 'standard configuration', the sugar being roughly perpendicular to the attached base. There is a residue on each chain every 3·4 A. in the z-direction. We have assumed an angle of 36° between adjacent residues in the same chain, so that the structure repeats after 10 residues on each chain, that is, after 34 A. The distance of a phosphorus atom from the fibre axis is 10 A. As the phosphates are on the outside, cations have easy access to them.

This figure is purely diagrammatic. The two ribbons symbolize the two phosphate—sugar chains, and the horizontal rods the pairs of bases holding the chains together. The vertical line marks the fibre axis

The structure is an open one, and its water content is rather high. At lower water contents we would expect the bases to tilt so that the structure could become more compact.

The novel feature of the structure is the manner in which the two chains are held together by the purine and pyrimidine bases. The planes of the bases are perpendicular to the fibre axis. They are joined together in pairs, a single base from one chain being hydrogen-bonded to a single base from the other chain, so that the two lie side by side with identical z-co-ordinates. One of the pair must be a purine and the other a pyrimidine for bonding to occur. The hydrogen bonds are made as follows: purine position 1 to pyrimidine position 1; purine position 6 to pyrimidine position 6.

If it is assumed that the bases only occur in the structure in the most plausible tautomeric forms (that is, with the keto rather than the enol configurations) it is found that only specific pairs of bases can bond together. These pairs are: adenine (purine) with thymine (pyrimidine), and guanine (purine) with cytosine (pyrimidine).

In other words, if an adenine forms one member of a pair, on either chain, then on these assumptions the other member must be thymine; similarly for guanine and cytosine. The sequence of bases on a single chain does not appear to be restricted in any way. However, if only specific pairs of bases can be formed, it follows that if the sequence of bases on one chain is given, then the sequence on the other chain is automatically determined.

It has been found experimentally[3,4] that the ratio of the amounts of adenine to thymine, and the ratio of guanine to cytosine, are always very close to unity for deoxyribose nucleic acid.

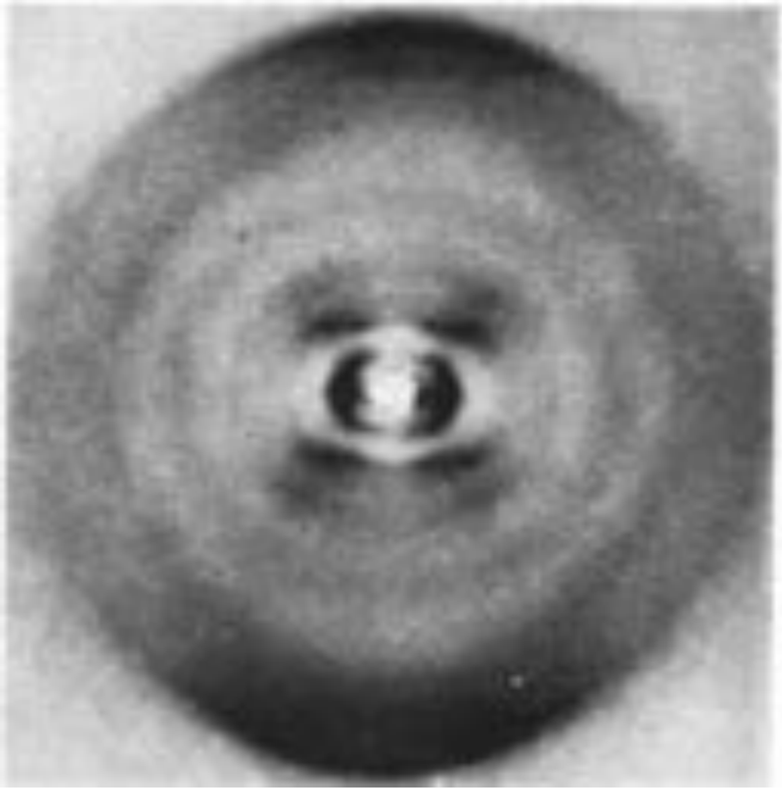
It is probably impossible to build this structure with a ribose sugar in place of the deoxyribose, as the extra oxygen atom would make too close a van der Waals contact.

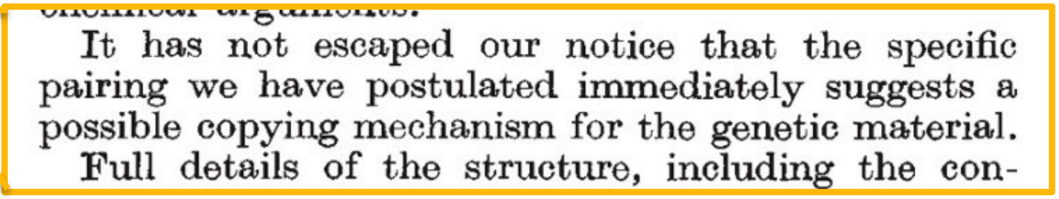
The previously published X-ray data[5,6] on deoxyribose nucleic acid are insufficient for a rigorous test of our structure. So far as we can tell, it is roughly compatible with the experimental data, but it must be regarded as unproved until it has been checked against more exact results. Some of these are given in the following communications. We were not aware of the details of the results presented there when we devised our structure, which rests mainly though not entirely on published experimental data and stereochemical arguments.

It has not escaped our notice that the specific pairing we have postulated immediately suggests a possible copying mechanism for the genetic material.

Full details of the structure, including the conditions assumed in building it, together with a set of co-ordinates for the atoms, will be published elsewhere.

We are much indebted to Dr. Jerry Donohue for constant advice and criticism, especially on interatomic distances. We have also been stimulated by a knowledge of the general nature of the unpublished experimental results and ideas of Dr. M. H. F. Wilkins, Dr. R. E. Franklin and their co-workers at





chemical arguments.

It has not escaped our notice that the specific pairing we have postulated immediately suggests a possible copying mechanism for the genetic material.

Full details of the structure, including the con-

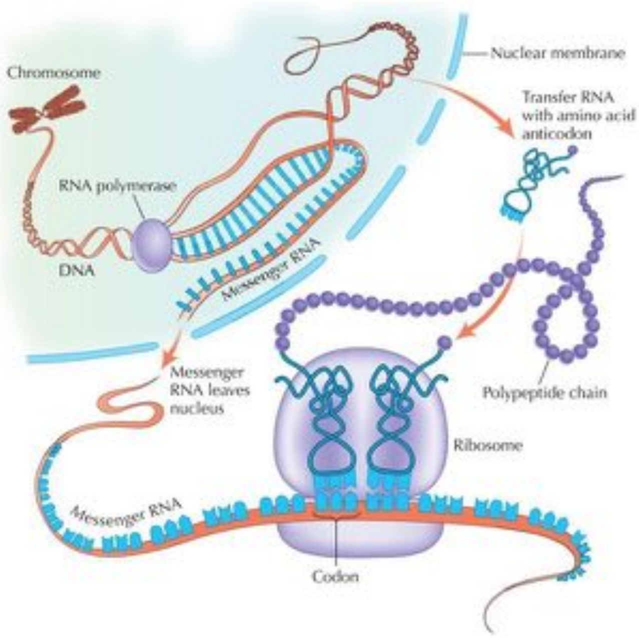
Molecular structure of nucleic acids; a structure for deoxyribose nucleic acid

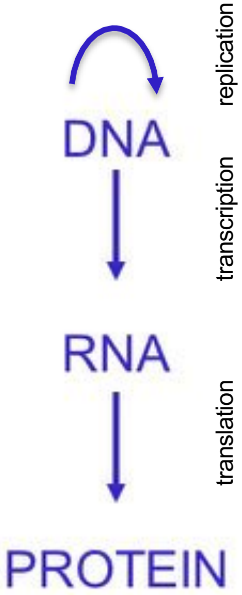
Watson JD, Crick FH (1953). *Nature* 171: 737–738.

Nobel Prize in Physiology or Medicine in 1962

Central Dogma of Molecular Biology

"Once 'information' has passed into protein it cannot get out again. In more detail, the transfer of information ***from nucleic acid to nucleic acid, or from nucleic acid to protein may be possible***, but transfer from protein to protein, or from protein to nucleic acid is impossible. Information means here the precise determination of sequence, either of bases in the nucleic acid or of amino acid residues in the protein"





replication

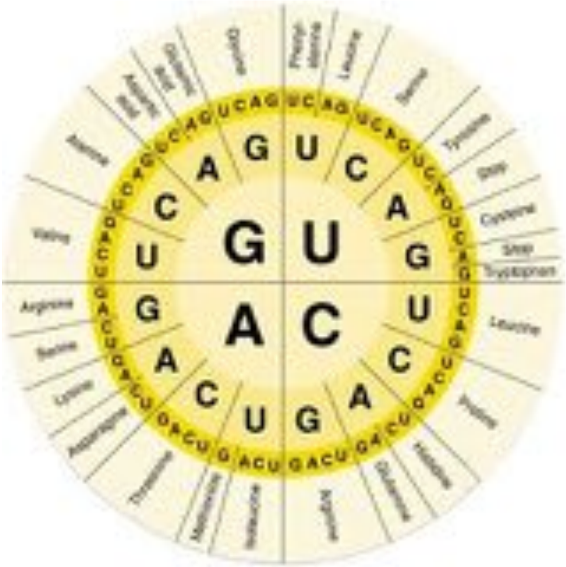
DNA

transcription

RNA

translation

PROTEIN

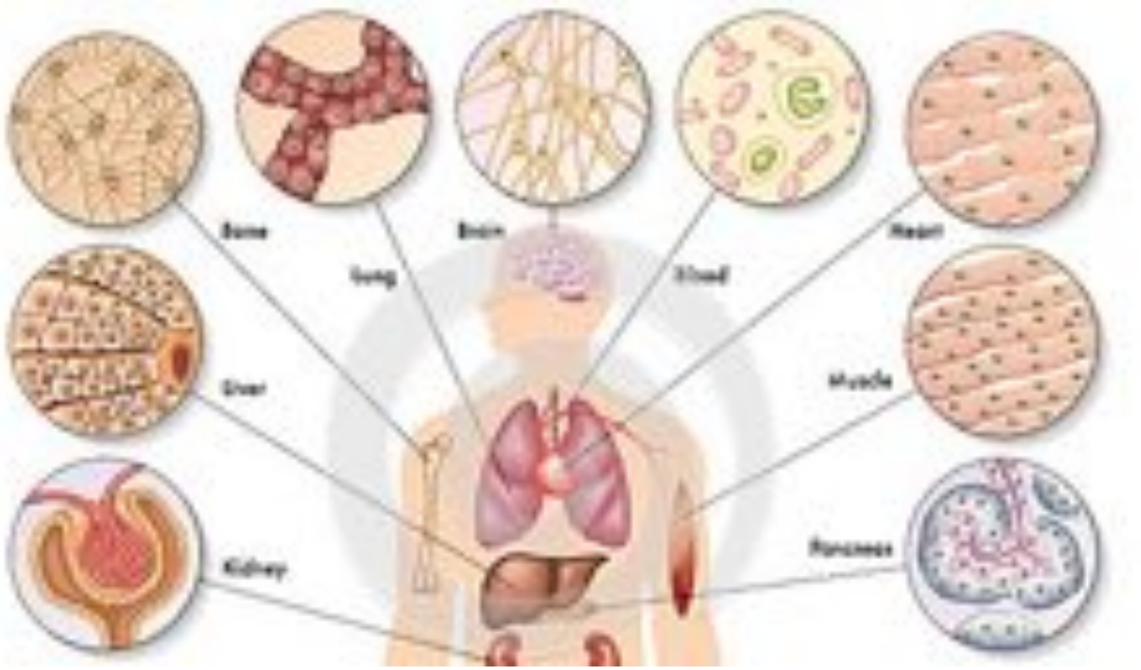


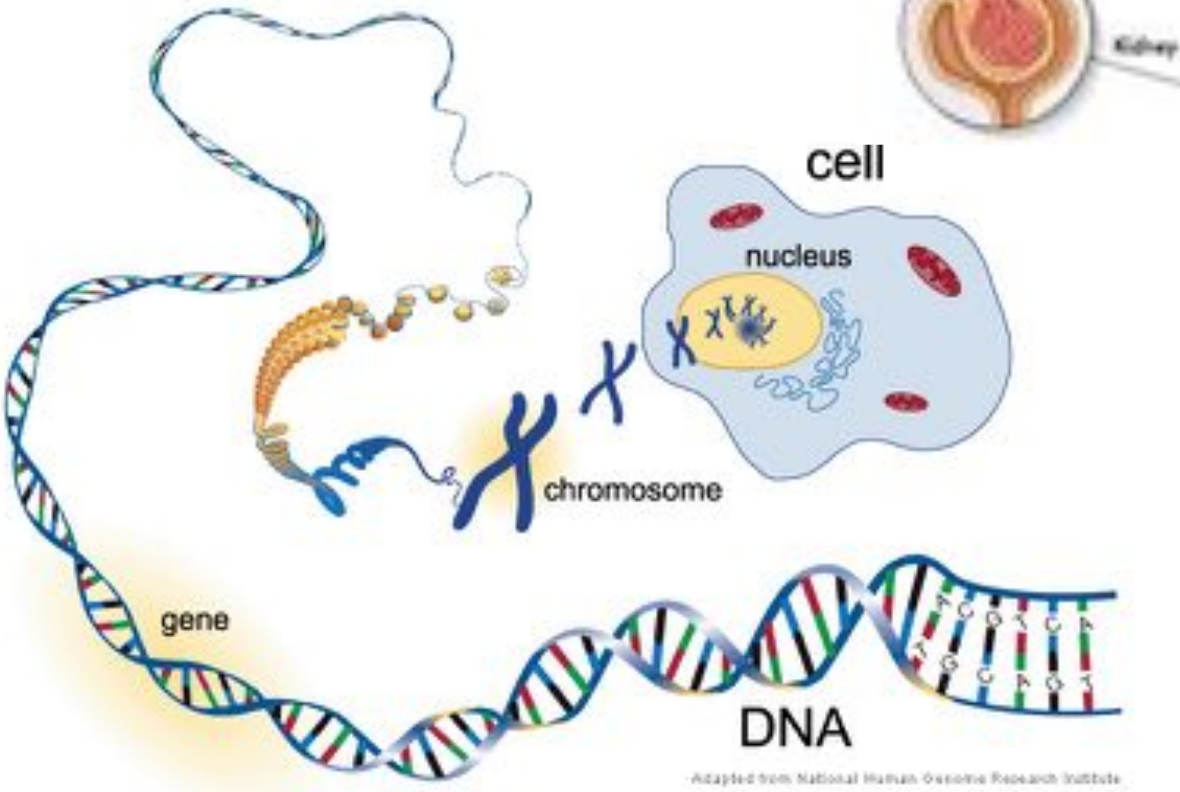
On Protein Synthesis

Crick, F.H.C. (1958). Symposia of the Society for Experimental Biology pp. 138–163.

One Genome, Many Cell Types

Each cell of your body contains an exact copy of your 3 billion base pair genome.

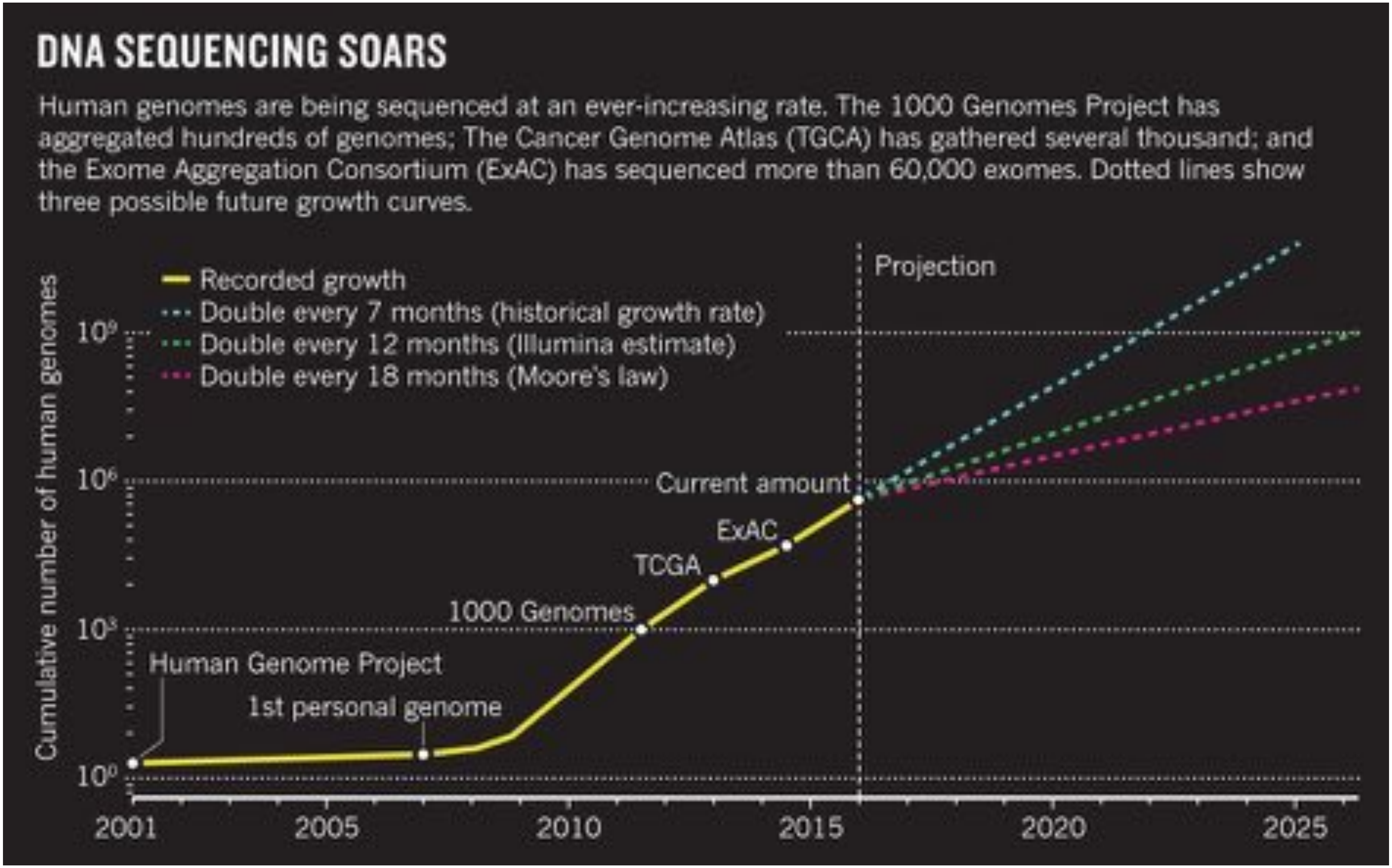




Adapted from National Human Genome Research Institute

Your body has a few hundred (thousands?) major cell types, largely defined by the gene expression patterns

Sequencing Capacity



DNA SEQUENCING SOARS

Human genomes are being sequenced at an ever-increasing rate. The 1000 Genomes Project has aggregated hundreds of genomes; The Cancer Genome Atlas (TGCA) has gathered several thousand; and the Exome Aggregation Consortium (ExAC) has sequenced more than 60,000 exomes. Dotted lines show three possible future growth curves.

Big Data: Astronomical or Genomical?

Stephens, Z, et al. (2015) PLOS Biology DOI: 10.1371/journal.pbio.1002195

Unsolved Questions in Biology

- What is your genome sequence?

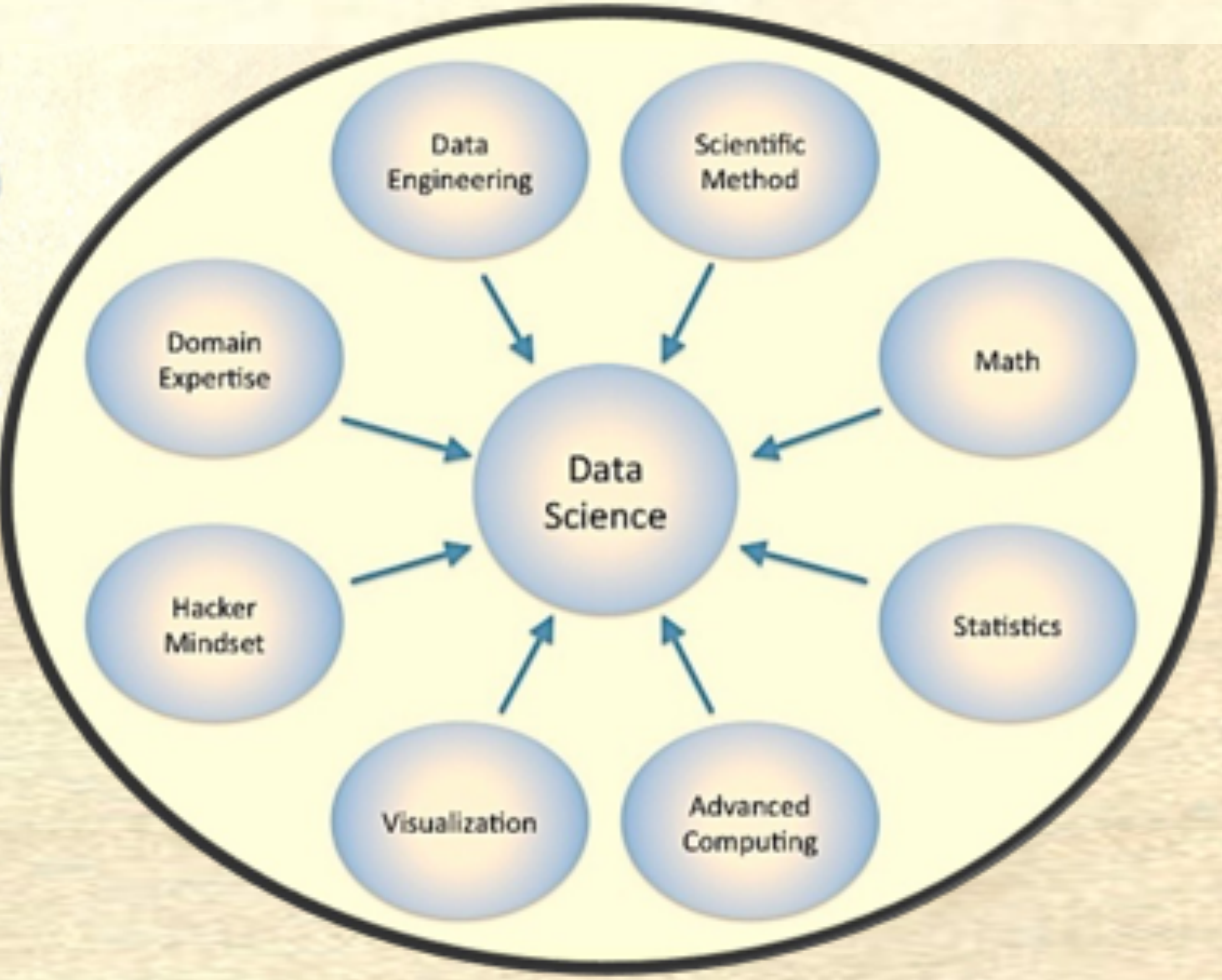
The instruments provide the data, but none of the answers to any of these questions.

What software and systems will?

And who will create them?

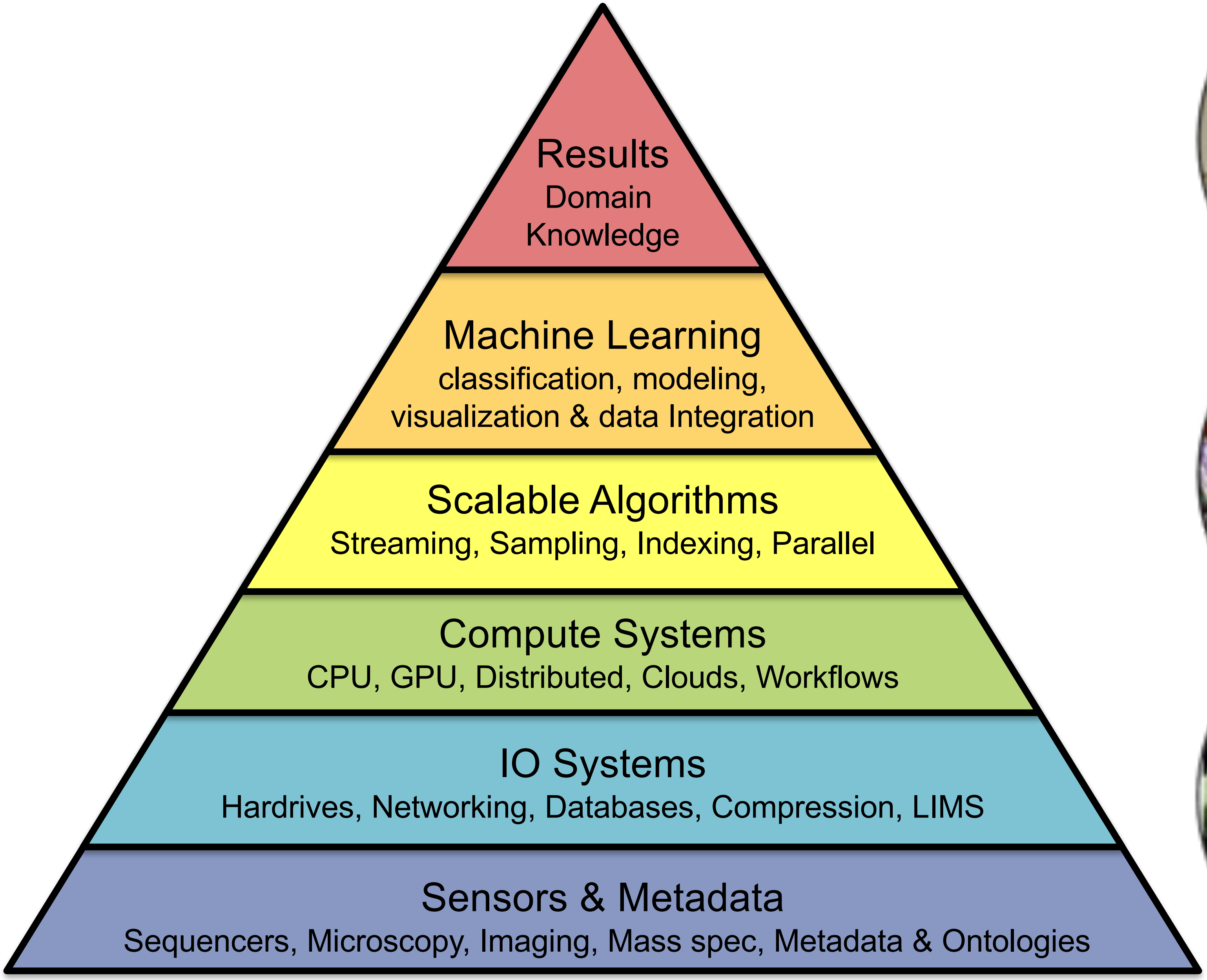
- ***Plus thousands and thousands more***

Who is a Data Scientist?



http://en.wikipedia.org/wiki/Data_science

Comparative Genomics Technologies



Results
Domain Knowledge

Machine Learning
classification, modeling, visualization & data Integration

Scalable Algorithms
Streaming, Sampling, Indexing, Parallel

Compute Systems
CPU, GPU, Distributed, Clouds, Workflows

IO Systems
Hardrives, Networking, Databases, Compression, LIMS

Sensors & Metadata
Sequencers, Microscopy, Imaging, Mass spec, Metadata & Ontologies

Next Steps

1. Reflect on the magic and power of DNA ☺

2. Check out the course webpage

3. Register on Piazza

4. Get Ready for assignment 1

    1. Set up Linux, set up Docker
    2. Set up Dropbox for yourself!
    3. Get comfortable on the command line