

“Next-Generation Sequencing (NGS):future perspectives”

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Biologists modus operandi

- ✦ Observing a phenomenon that is in some way interesting or puzzling.
- ✦ Making a guess as to the explanation of the phenomenon.
- ✦ Devising a test to show how likely this explanation is to be true or false.
- ✦ Carrying out the test, and, on the basis of the results, deciding whether the explanation is a good one or not. In the latter case, a new explanation will (with luck) 'spring to mind' as a result of the first test.

The Observed phenomenon



Selection of test times



But what is the real event?



Sometimes you could be lucky



“Positive” results are used “negative” rejected
Why?

Only positive results are publishable

Things have changed: NGS

- ✦ 454 (Pyrosequencing)
- ✦ Illumina
- ✦ Solid
- ✦ Pacific Biosciences
- ✦ Oxford Nanopores
- ✦ Many more

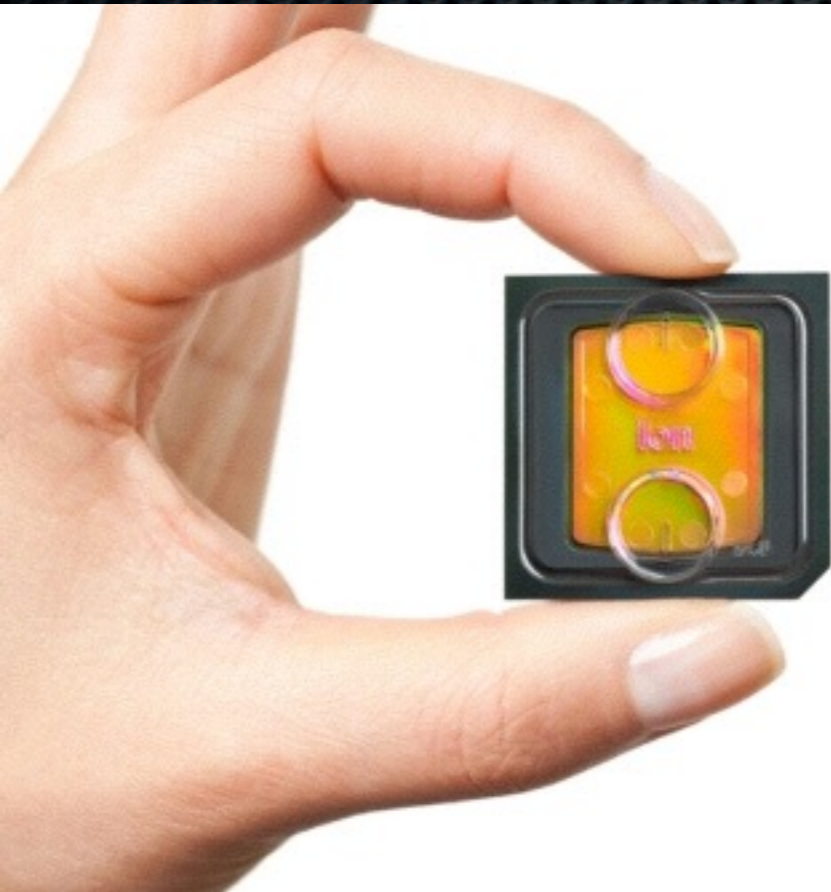
Next Generation techniques



Next Generation Sequencing

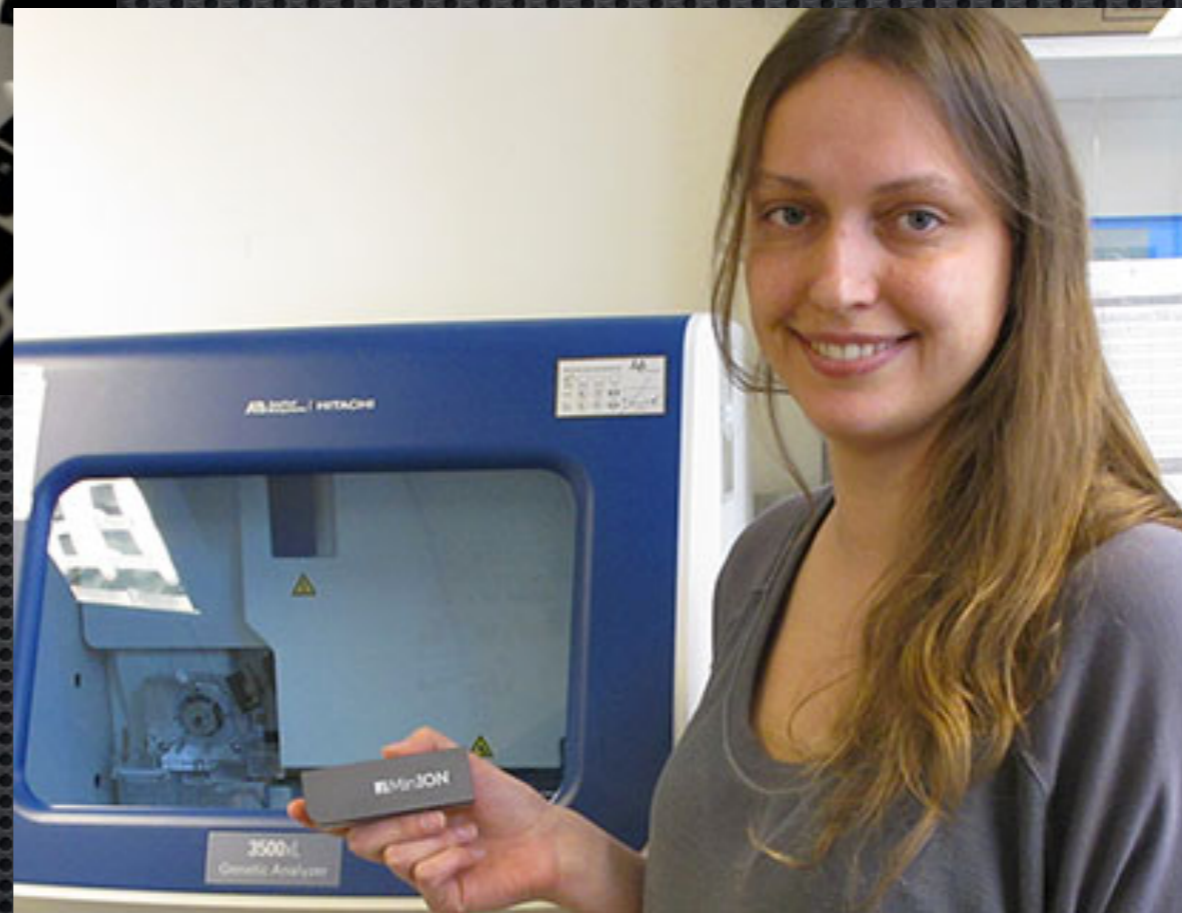


Ion Torrent: "Personal Genome Machine".



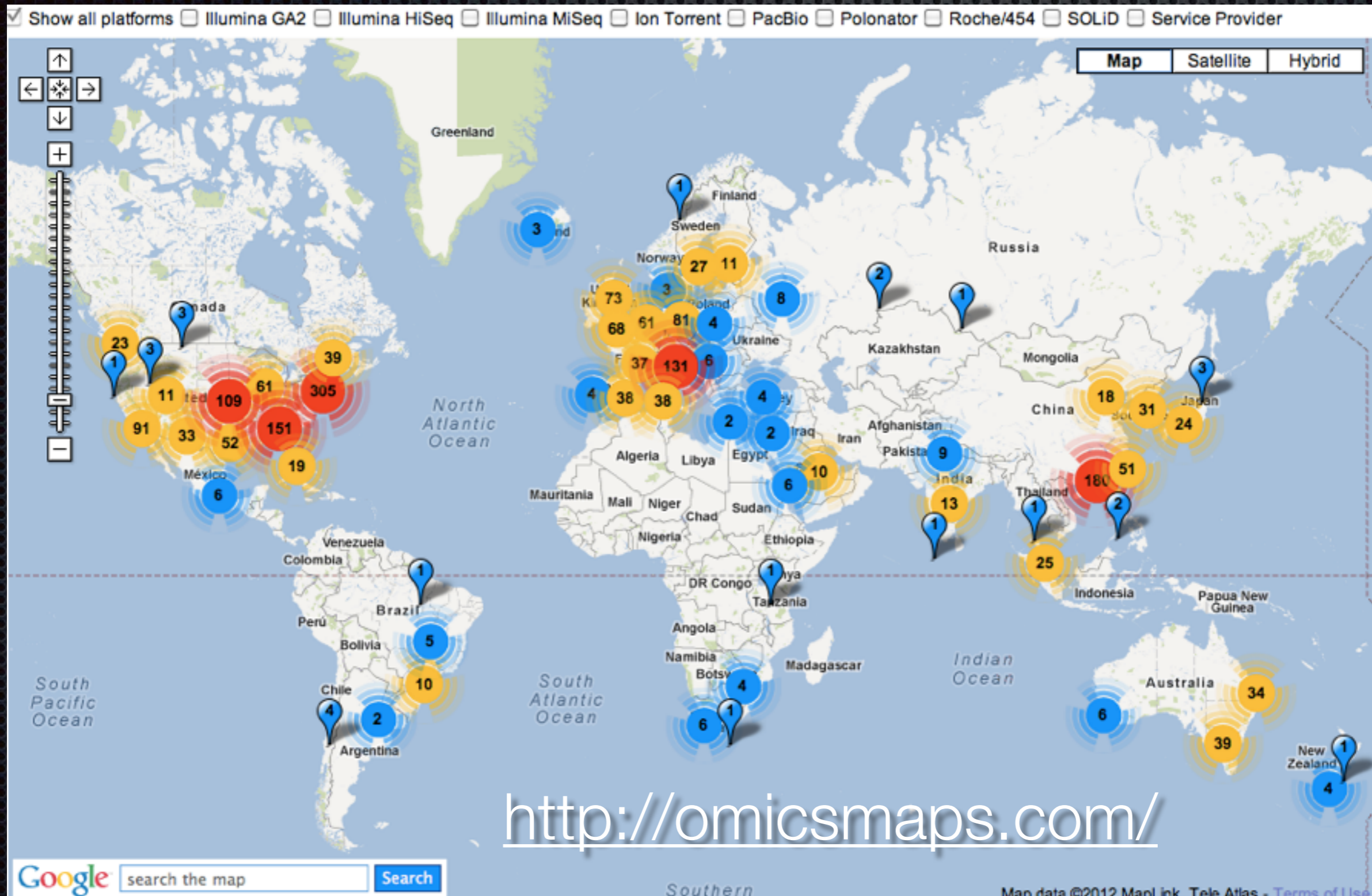
✦ LIFE TECHNOLOGIES CORPORATION

Oxford Nanopore



<http://www.nanoporetech.com/>

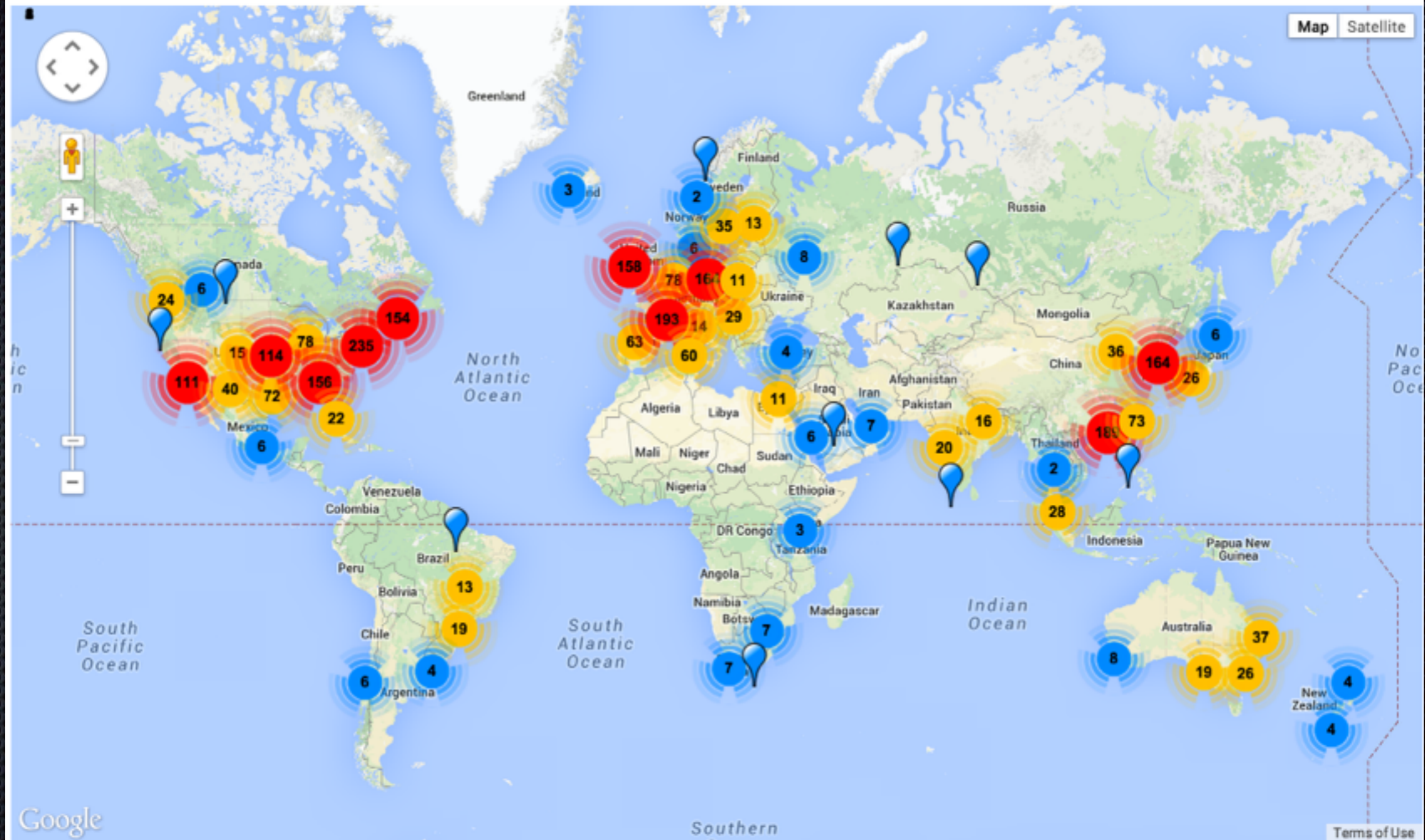
World NGS Map



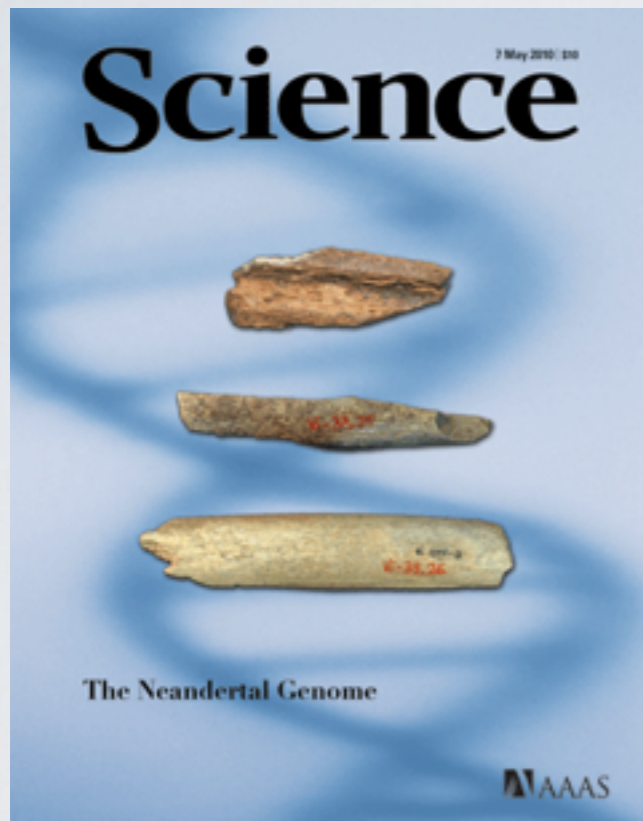
Today

Next Generation Genomics: World Map of High-throughput Sequencers

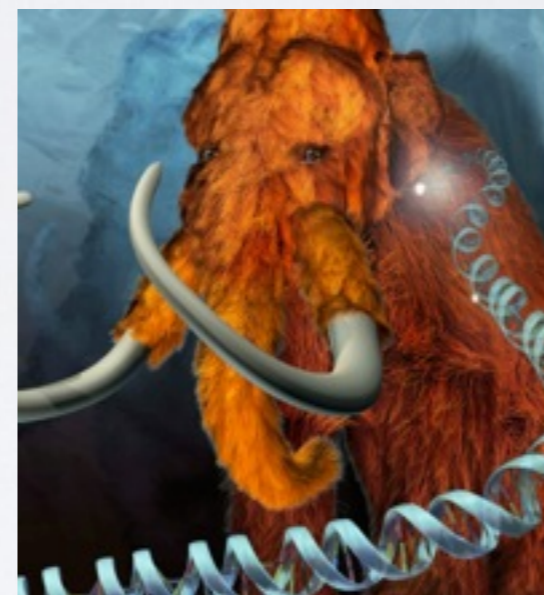
Show all platforms 454 HiSeq Illumina GA2 Ion Torrent MiSeq PacBio Polonator Proton SOLiD Service Provider



NGS SUCCESS STORIES



Tomato Genome



Mammoth Genome



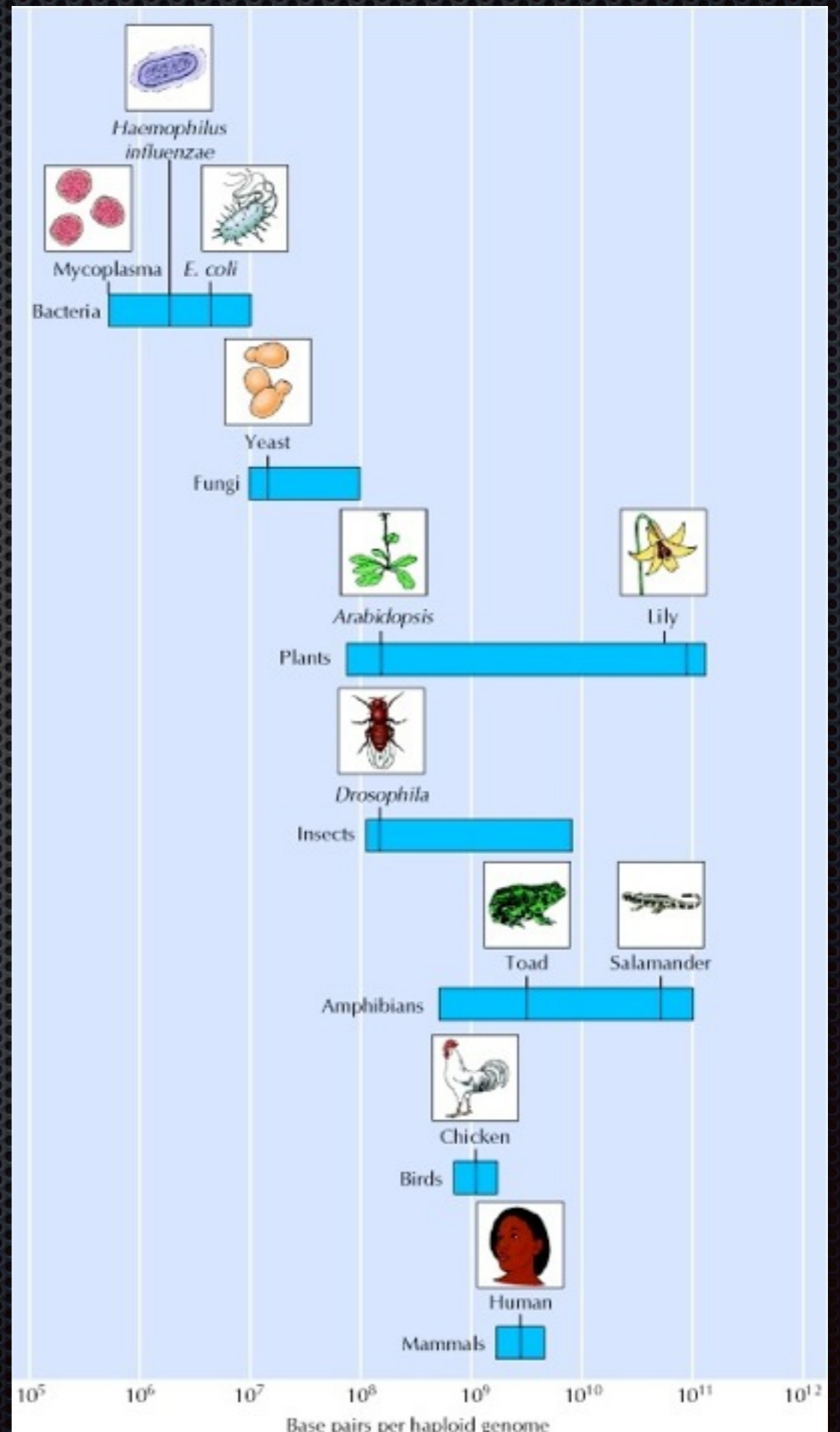
Chicken domestication



One of the Centres: BGI

- ✦ The incredible productive efficiency of BGI has moved huge projects on human and other life forms variability from imagination to actual reality.
- ✦ The Center is taking a leading role in sequencing 10,000 vertebrates through the Genome 10K project; 5,000 insects and other arthropods through the i5k initiative; and more than 1,000 birds, including some extinct ones in a separate project.
- ✦ BGI has launched the Million Project, aimed to expand the sequenced samples to 1M for human, animals, plants and bacteria

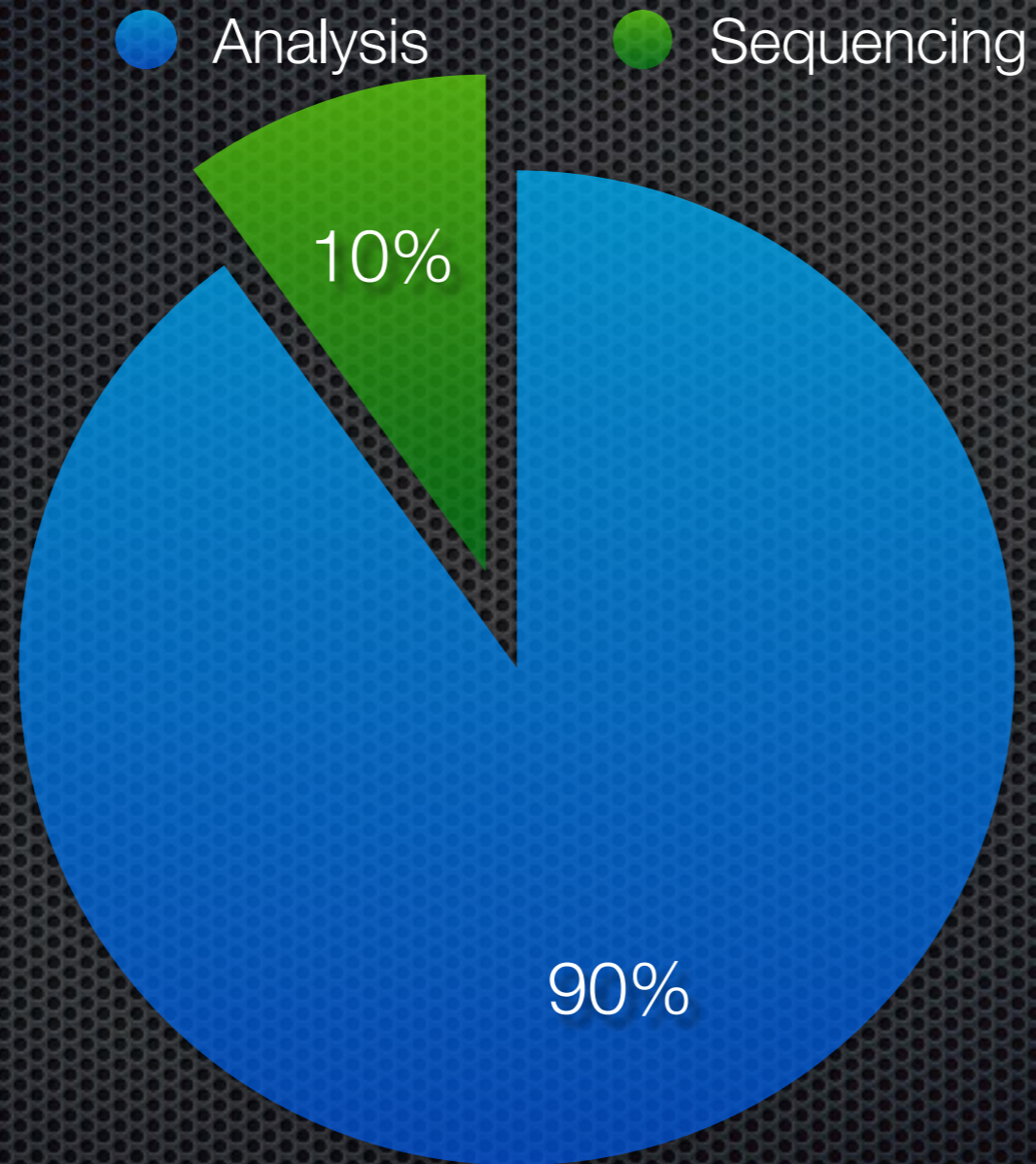
Genome sizes



Is wonderful! Or?

- ✦ Sequence without knowledge connected to it is worth: 0
- ✦ The deluge of data produced by these hordes of machines worldwide demand automatic workflows
- ✦ Complete new systems to shuffle data around
- ✦ Storage of never used amounts
- ✦ Machines with gigantic amounts of RAM
- ✦ BIOINFORMATICIANS!!

COSTS



OTHER PROBLEMS


- ✦ NOmenclature
- ✦ Publishing culture
- ✦ Moving target development
- ✦ Old ways of work and resistance to changes in culture

A brief overview of how to derive a genome or transcriptome from a single cell.

Subject terms: DNA sequencing · RNA sequencing · Whole genome amplification ·

Transcriptomics

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New challenges



today TB of data
soon PB

UPPMAX/UPPNEX



5 PB of Storage

5 PB Storage

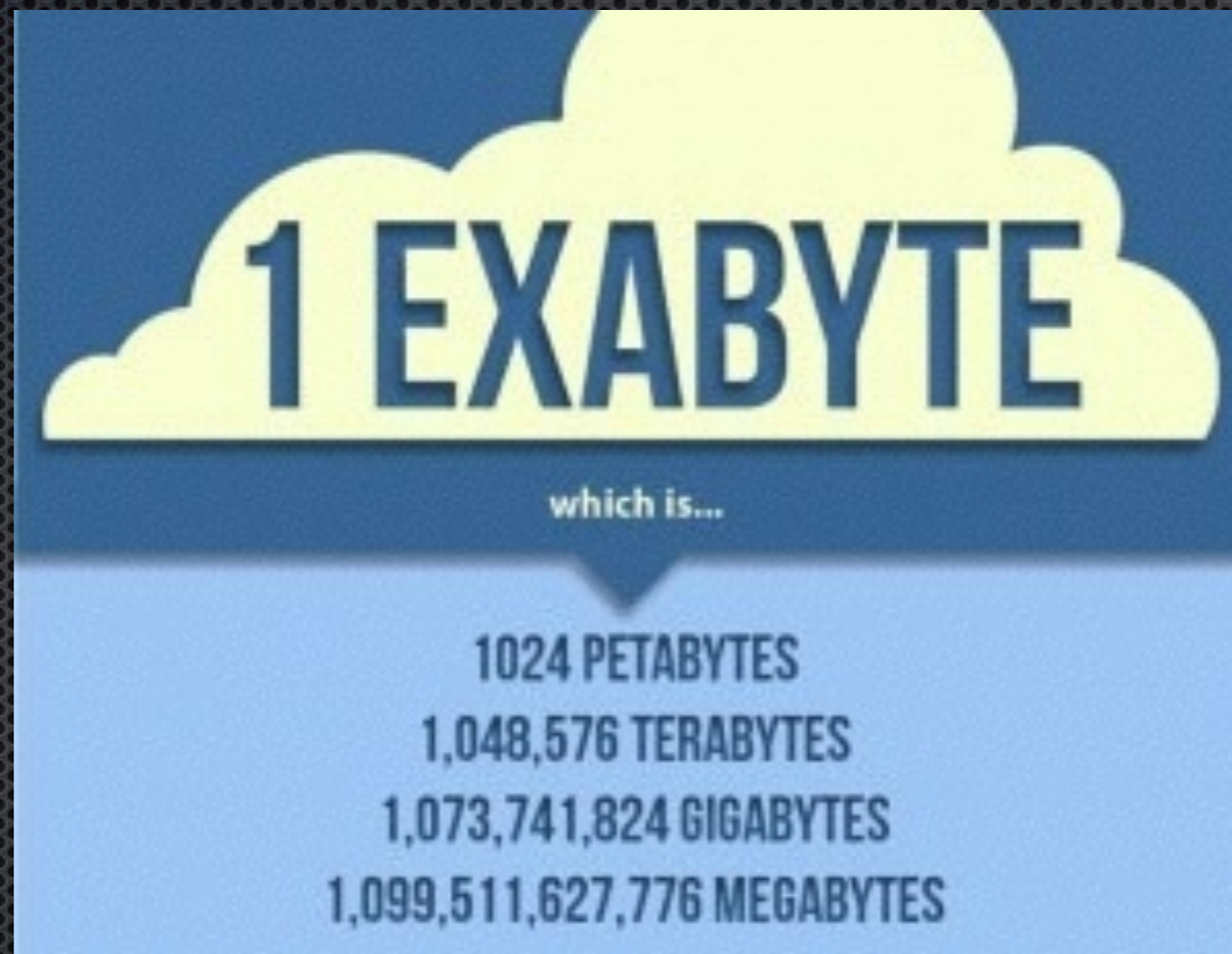
So how much is a PB?

PETABYTE

A petabyte contains enough information to fill a 45 mile high stack of CDs, the distance between SLAC and Berkeley Lab.

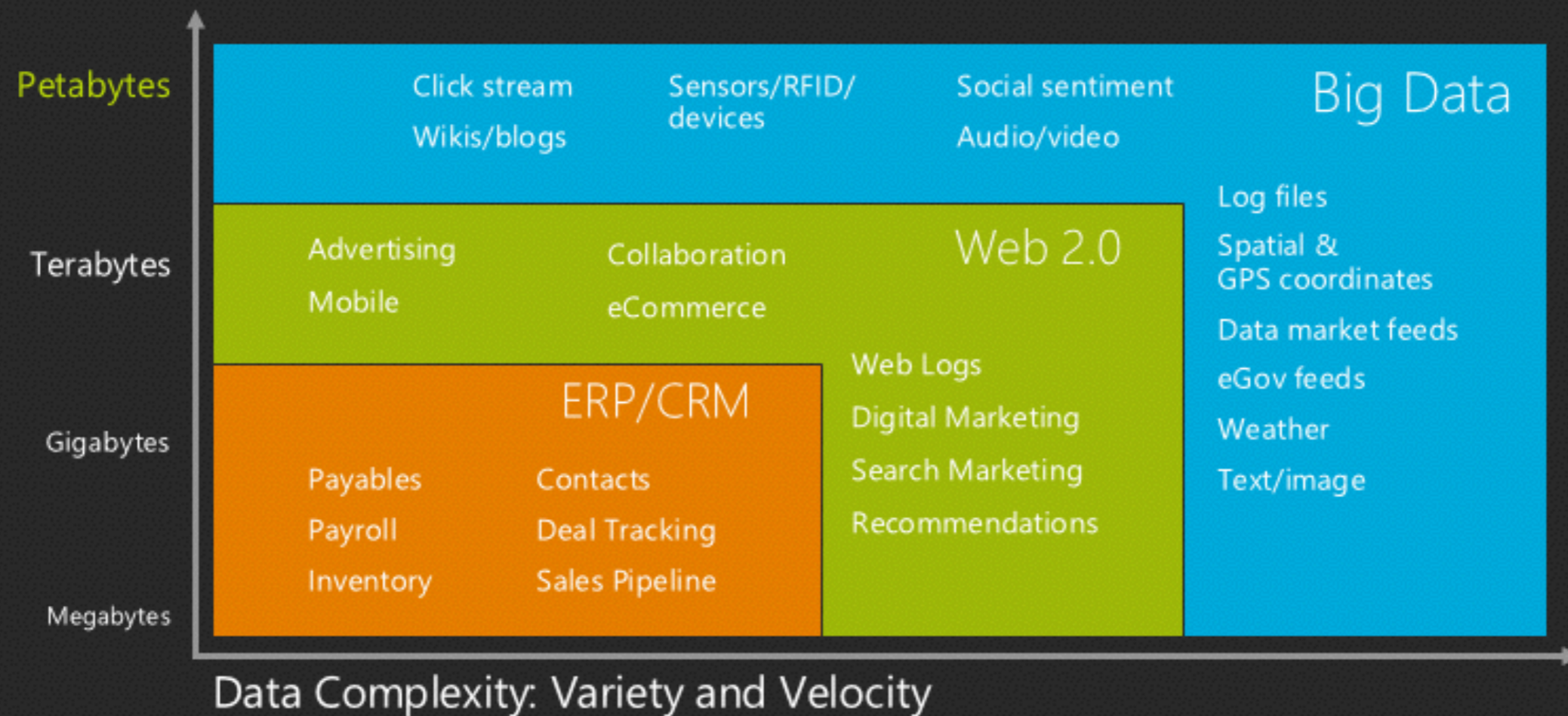


We are in the ERA of EXABYTE



BIG DATA

WHAT IS BIG DATA?



EXOME

- ✦ Whole-exome sequencing has already been used for identifying the molecular defects of single gene disorders, for elucidating some genetically heterogeneous disorders and for improving the accuracy of diagnosis of patients.
- ✦ data for whole-exome sequencing is orders of magnitude smaller than for whole-genome sequencing
- ✦ On average, whole-exome sequencing identifies 12 000 variants in coding regions, of which ~90% are found in publicly available databases. In comparison, ~5 million variants, including 144 000 new variants, are reported on average by whole-genome sequencing.

EXOME

- ✦ OMIM the catalog of Mendelian disorders, lists >3000 disorders where the molecular basis has been reported. Still, >3500 disorders are listed where the genetic cause remains unknown and has yet to be identified
- ✦ Other main fields of interest for human geneticists are complex diseases and cancer. For example, the usage of NGS has led to the identification of driver mutations for specific types of cancer. **Single cell sequencing.**

Single cell sequencing

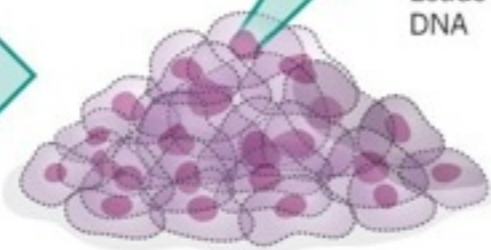
ONE GENOME FROM MANY

Sequencing the genomes of single cells is similar to sequencing those from multiple cells — but errors are more likely.

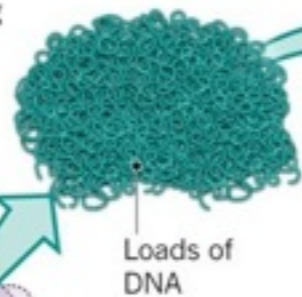
► Standard genome sequencing



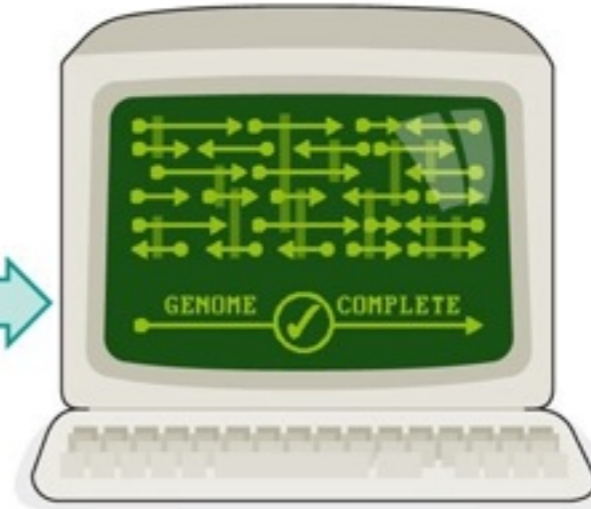
A sample containing thousands to millions of cells is isolated.



DNA is extracted from all the nuclei.

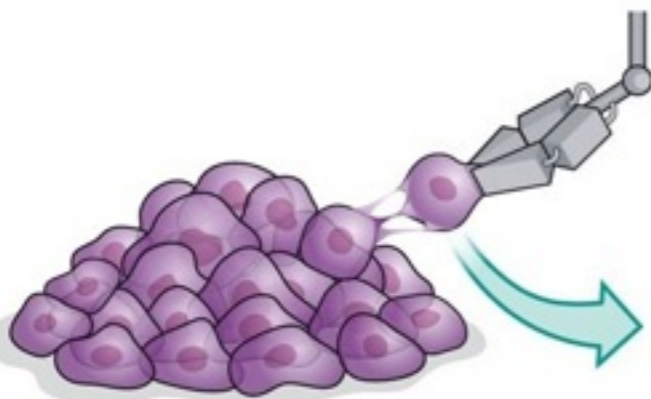


DNA is broken into fragments and then sequenced.

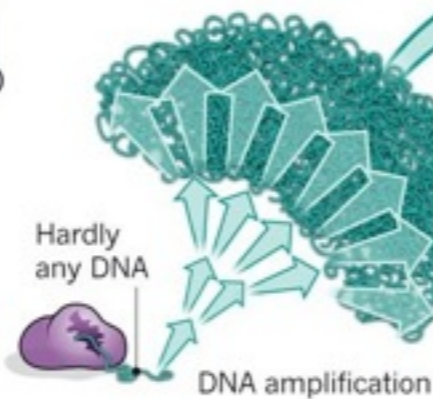


The sequences are assembled to give a common, 'consensus' sequence.

► Single-cell sequencing



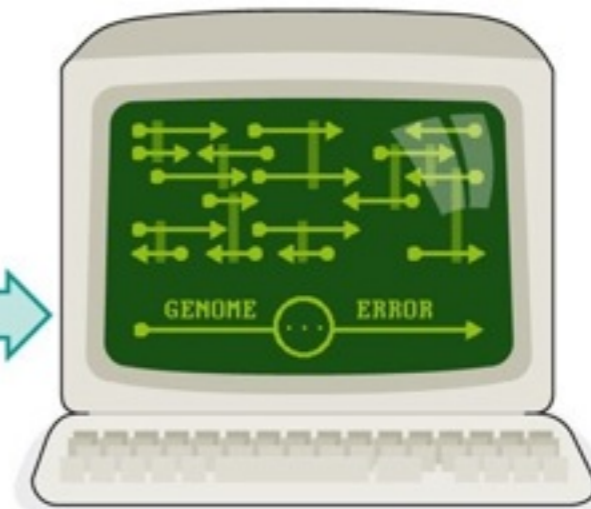
A single cell is difficult to isolate, but it can be done mechanically or with an automated cell sorter.



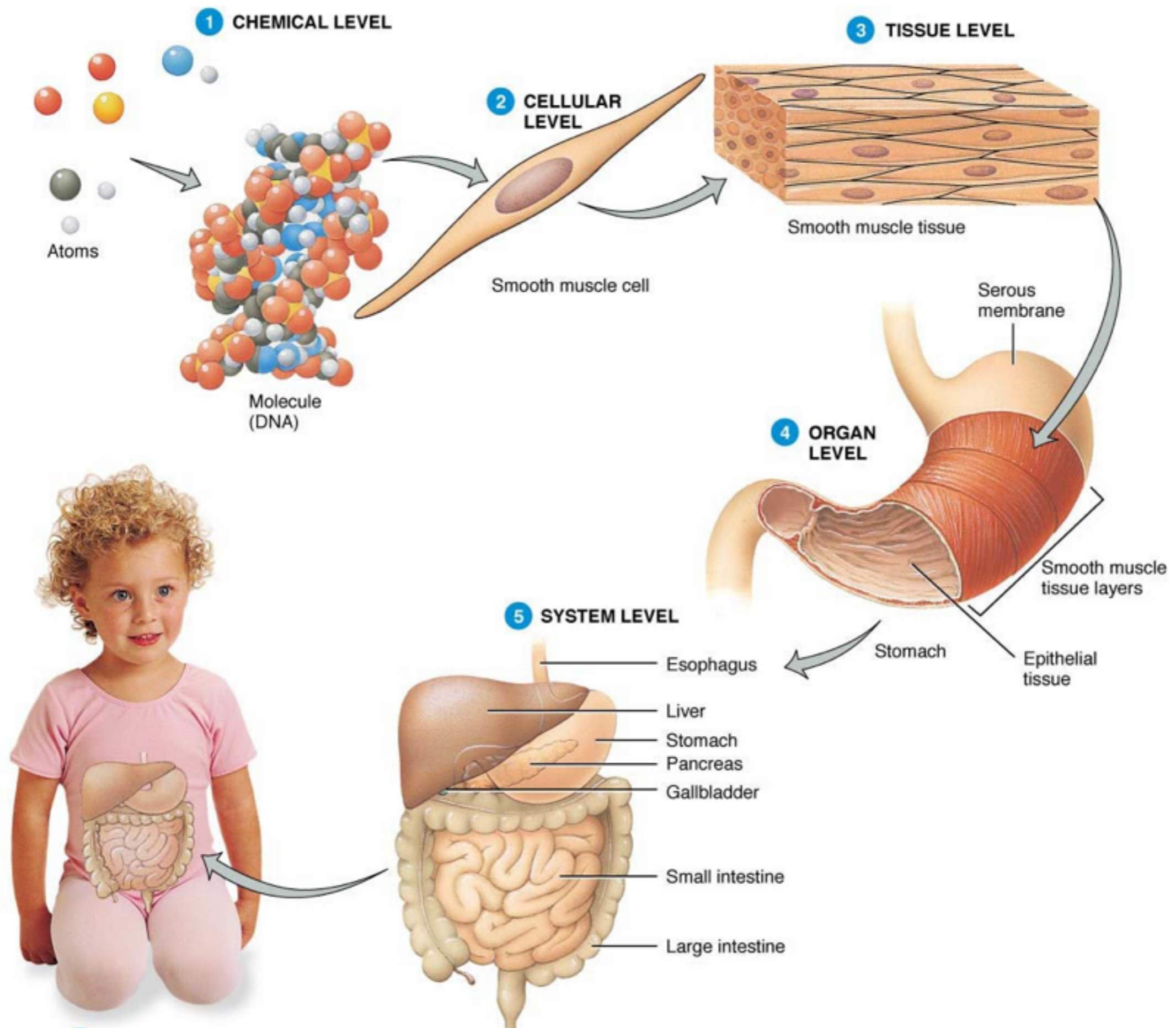
The DNA is extracted and amplified, during which errors can creep in.



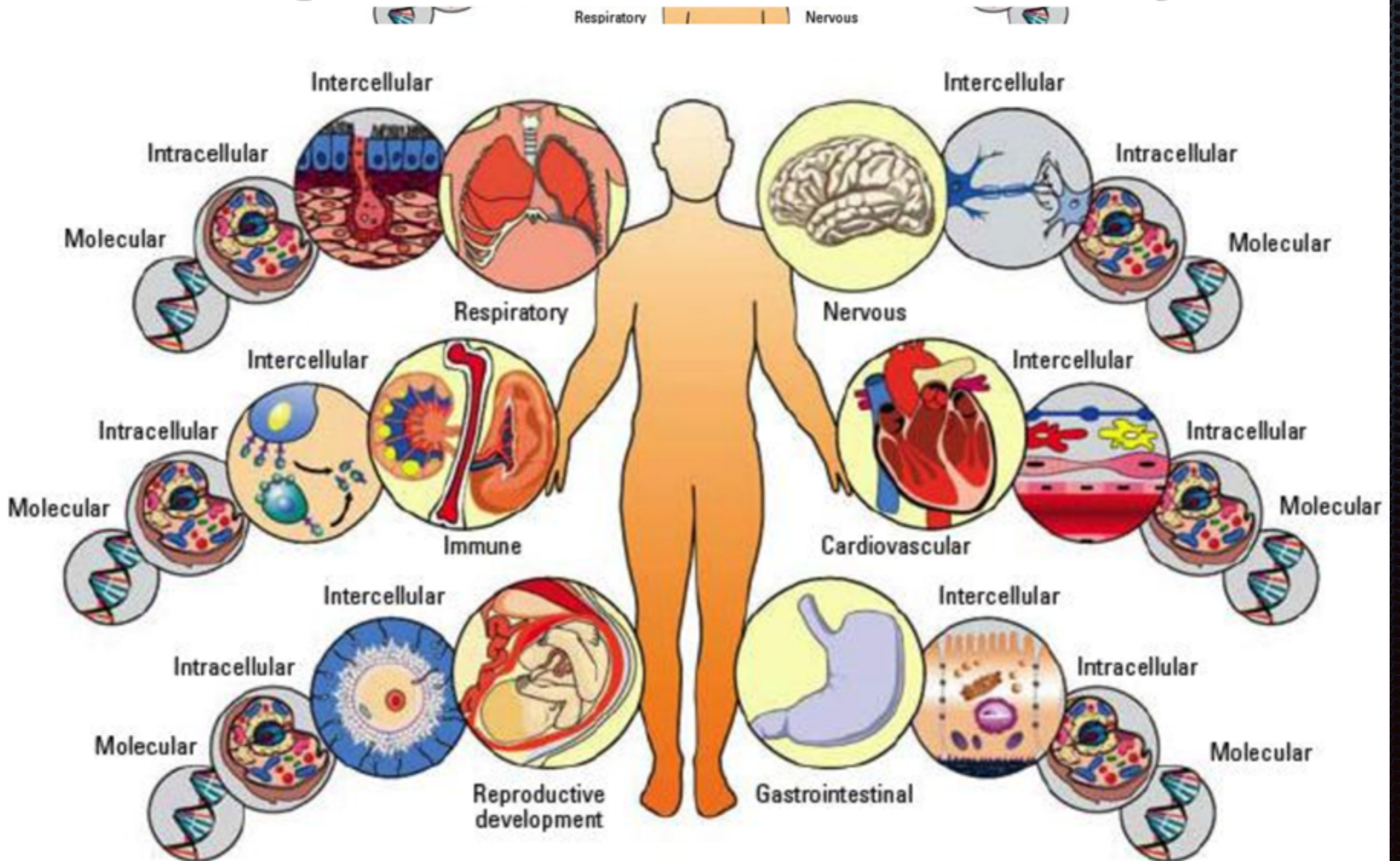
Amplified DNA is sequenced.



Errors introduced in earlier steps make sequence assembly difficult; the final sequence can have gaps.



Organization of the Body

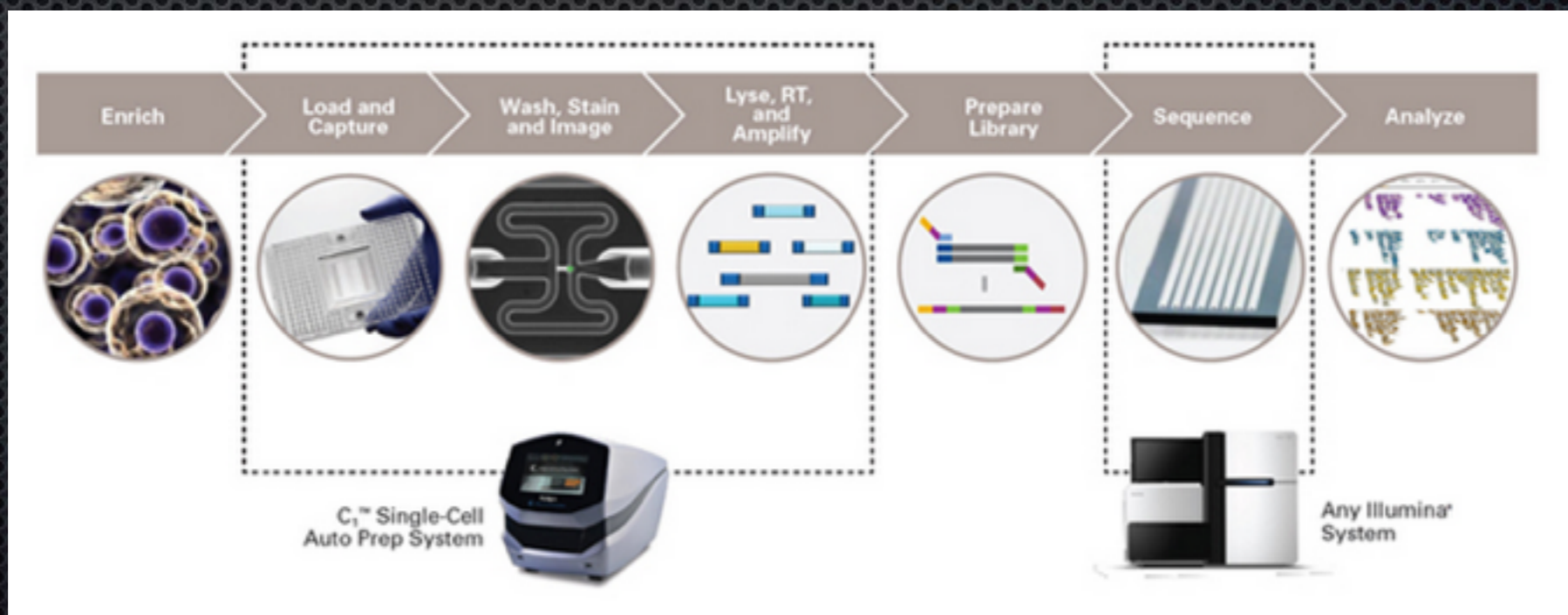


Single Cell mRNA Sequencing

REVEAL HIDDEN VARIATION

C₁TM Single-Cell DNA Sequencing

One workflow to discover genetic variants in individual cells



Epigenomics and Epigenetics

- ✦ Epigenomics deals with the chemical modifications (e.g., 5' methylation) of DNA and RNA and the impacts of such changes on levels of gene expression.
- ✦ Tumor methylation profiles could also be detected in body fluid specimens showing a promising role as non-invasive markers for cancer diagnosis towards an early detection and potentially for the surveillance of cancer patients in a near future. However, the epigenomic exploration of cancer has only just begun.

Microbiome

A microbiome is the totality of microbes, their genetic elements (genomes), and environmental interactions in a particular environment.



<http://www.secondgenome.com>

The Economist

AUGUST 18TH-24TH 2012

Economist.com

The Catholic church's unholy mess

Paul Ryan: the man with the plan

Generation Xhausted

China, victim of the Olympics?

On the origin of specie

Microbes maketh man



Epigenetics



Fat and lean

- ✦ Metabolic effects of transplanting gut microbiota from lean donors to subjects with metabolic syndrome.
A. Vrieze et al, EASD abstracts, 24 September 2012.
- ✦ The result was: Lean donor faecal infusion improves hepatic and peripheral insulin resistance as well as fasting lipid levels in obese individuals with the metabolic syndrome

New applications

- ✦ Only imagination will put the limits of what its possible to be done using Next Generation Technologies!

Human Medicine Success Stories

- ✦ Whole-genome sequencing in autism identifies hot spots for de novo germline mutation. *Cell*. 2012 Dec 21;151(7):1431-42. doi: 10.1016/j.cell.2012.11.019.
- ✦ Genes contributing to pain sensitivity in the normal population: an exome sequencing study. *PLoS Genet*. 2012 Dec;8(12)

23andMe: Linda Avey and Anne Wojcicki

The screenshot shows the 23andMe website homepage. At the top left is the 23andMe logo with the tagline "genetics just got personal." To the right are links for "log in", "claim codes", "blog", "we're hiring!", and "help". A search bar labeled "Search 23andMe" is also present. Below the header is a navigation bar with links for "home", "our service", "genetics 101", "store", and "about us". The main content area features a large headline: "23andMe Democratizes Personal Genetics. More Data, New Features, Now \$399." To the left of this headline is an image of a 23andMe kit box and several sample reports, including "Clinical Reports" with "Disease Risks" and "Traits" sections. To the right of the headline are four key features listed in blue text: "Health and Traits: What do your genes mean for your health? Information on 90+ traits and diseases, with more added monthly.", "Ancestry: Where are your ancestors from? Ancestry features trace continental origins plus parental lineage.", "Compare Your DNA: How similar are you to friends and family? Tools for side-by-side comparisons, family inheritance and global similarity.", and "23andMe Research: Be part of the discovery process! Anyone can help advance science by participating in our research program." Below these features are three buttons: "Learn More >>>", "buy \$399 USD", and "try a demo". At the bottom of the page, there are three columns: "New at 23andMe" with a brain icon and text about a new Health and Traits view; "Recent News and Press Coverage" with dates and headlines; and "Learn More" with a list of links including "How the Process Works", "Frequently Asked Questions", "Scientific Standards", "Considerations", and "Complete List of Health and Traits".

23andMe genetics just got personal.

log in | claim codes | blog | we're hiring! | help

Search 23andMe

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23andMe Democratizes Personal Genetics.

More Data, New Features, Now \$399.

Clinical Reports

Disease Risks

Protein

World Traveler

1 2 3 4 5 6

7 8 9 10

Traits

Alcohol Flush Reaction

Other Taste Preferences

Eye Color

Lactose Intolerance

Health and Traits: What do your genes mean for your health?
Information on 90+ traits and diseases, with more added monthly.

Ancestry: Where are your ancestors from?
Ancestry features trace continental origins plus parental lineage.

Compare Your DNA: How similar are you to friends and family?
Tools for side-by-side comparisons, family inheritance and global similarity.

23andMe Research: Be part of the discovery process!
Anyone can help advance science by participating in our research program.

Learn More >>> buy \$399 USD try a demo

New at 23andMe

Oct. 8, 2008: 23andMe launches new, more informative Health and Traits view, 1 new survey, and 2 new reports.

Recent News and Press Coverage

Oct. 2, 2008 - 23andMe Announces Breast Cancer Initiative

Sept. 9, 2008 - 23andMe Democratizes Personal Genetics—More Data, New Features, Dramatically Lower Cost at \$399

Learn More

- How the Process Works
- Frequently Asked Questions
- Scientific Standards
- Considerations
- Complete List of Health and Traits

www.23andme.com



welcome

health

ancestry

how it works

store

search

help

Get to know you. Health and ancestry start here.



- Reports on 240+ health conditions and traits
- Discover your lineage, find relatives and more
- Get updates on your DNA as science advances

order now

\$99


Watch a life-changing story.

Take a more active role in managing your health

Knowing how your genes may impact your health can help you plan for the future and personalize your healthcare with your doctor.

Order Now »

23andMe estimates your genetic chances of getting
Type 2 Diabetes



AS LOW AS **8 %** AS HIGH AS **52 %**

23andMe will tell you:
Your genetic risk
What you can do

*This result based on our calculations for males with European ancestry



Plan for the future
Find out if your child will be at risk for 44 inherited conditions and learn about steps you can take.
[about carrier status](#)



Be on the lookout now
Knowing your health risks will help you and your doctor figure out health areas to keep an eye on.
[about disease risks](#)



Plan with your doctor
Personalize your healthcare by knowing in advance how you will respond to certain medications like Warfarin.
[about drug response](#)

Future

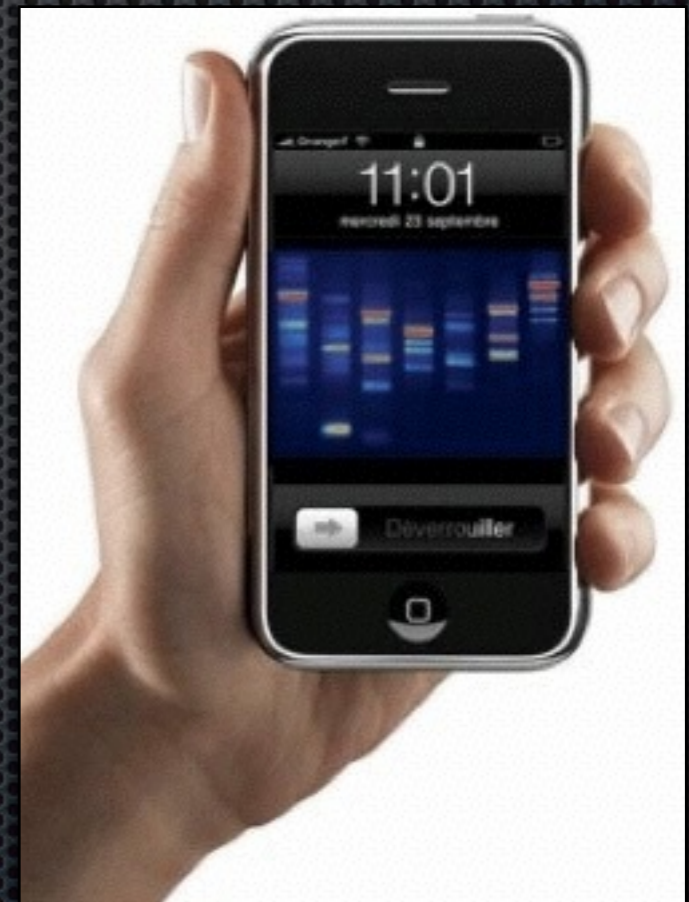


- ❖ Cell-free fetus DNA sequencing in the era of NGS: New opportunities for pre-natal diagnosis
- ❖ The main advantages in detecting free fetal DNA from maternal blood are the quick and early response and the total absence of risk for the fetus itself. In fact fetal DNA could be detected as early as the 6th week of pregnancy and the test simply requires a sample of mother's blood.



Share your genome sequence using your smartphone

- A smartphone app that can store your DNA – and perhaps one day allow your partner, your relatives or your doctor to scan it



Top 6 Deadly Diseases





- ✦ Trachea, Bronchus, Lung Cancers
- ✦ Malaria
- ✦ TBC
- ✦ Diarrheal
- ✦ Perinatal Conditions
- ✦ Chronic Obstructive Pulmonary Disease

High technology everywhere!



Field data collection


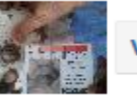


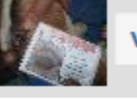


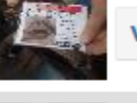


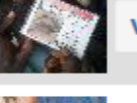



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Form USDA Goat Sampling v2 **Filter** none
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	pics rear	pics face_w_teeth	pics famacha	pics other	measures chest_girth	measures height	measures length	measures chest_bones_width	mea pin_bor
			View	71.5	65.0	64.0	13.5		
			View	67.0	63.0	63.0	13.0		
			View	73.0	67.0	57.0	14.0		
			View	75.0	68.0	65.0	15.5		

Submissions per page

Filters Applied

Display Metadata

Connectomics



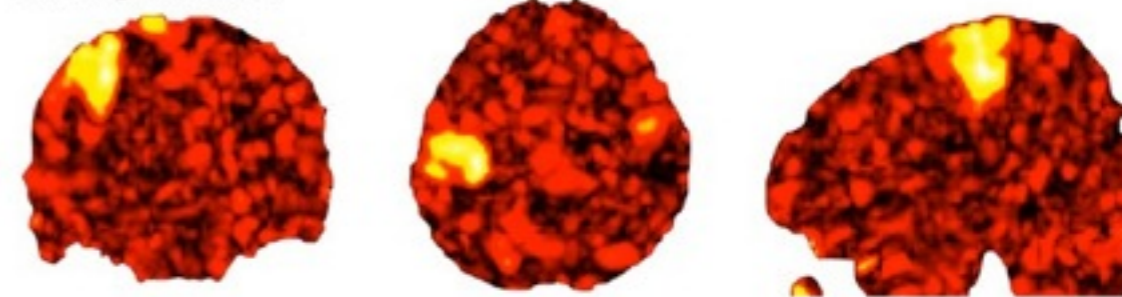
Connectomics



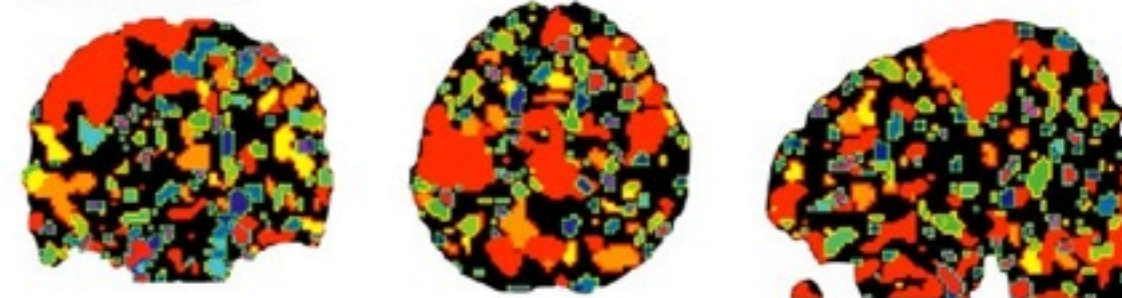
Student's *t*-test overlay



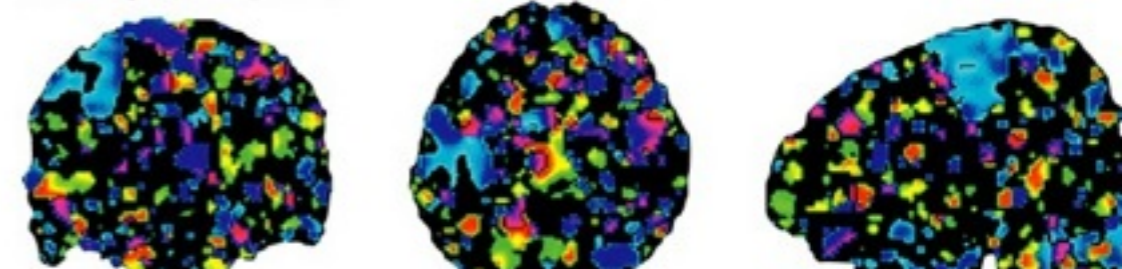
Fisher's *g* max statistic



Frequency of *g* max



Phase of *g* max components



USA:



- ✦ HGP: \$3.8 billion over 13 years.
- ✦ Map Brain project: \$3 billion

EU: €100M/year for ten years



HBP

The Human Brain Project



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THE PROJECT

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PARTNERS

FAQ

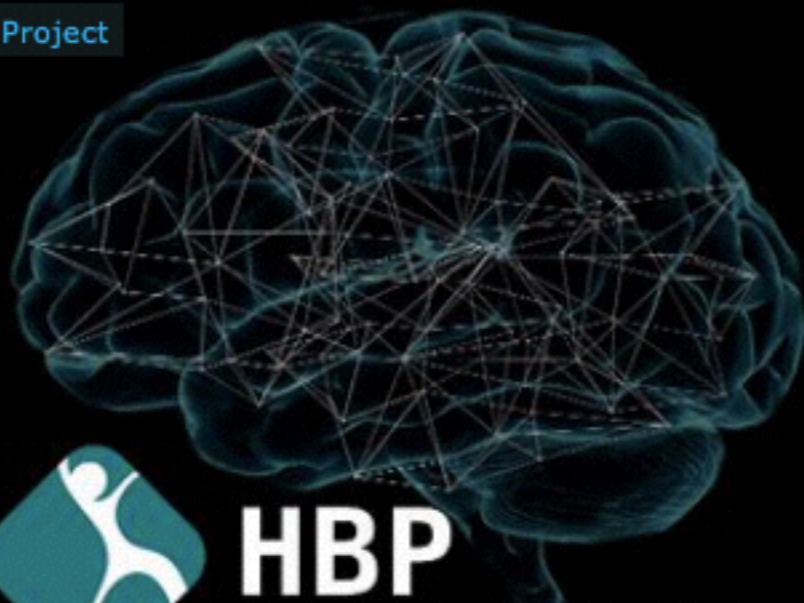
A EUROPEAN FLAGSHIP

The Human Brain Project has been selected as a European Flagship!



HBP-videooverview

from Human Brain Project



HBP

The Human Brain Project

EUROPEAN PARTNERS/



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THANKS!!