INTRODUCTION **BIOINFORMATICS** & **BIOLOGICAL** DATABASES

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nature

Andra Waagmeester (BW January 10th, 2013)

BLOCKCOORDINATOR

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ORGANISATION

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Practical information

- Lectures will mostly be in **Dutch** Slides and practical exercises are in **English**
- All the practical sessions, should be signed off.
- You are required to study the literature provided for each practical session, <u>before</u> the session starts.

For the first session, the literature can be studied in the session

- The Bioinformatics trajectory (BMW2003) in year 2 is present in periods 1, 3, 4 and 5. Per block a bioinformatics exam will be given. This exam is separate from the block exam. In the end you will get **one grade** for the Bioinformatics trajectory
- First exam Bioninformatics trajectory
 - Tuesday April 4rd, 13.00-16:00 <u>Open</u> book exam.

Course Material

ELEUM:

- The *slides* of the lecture will become available after the lecture.
- Per practical session *literature references* are provided.
- The exercises are available before the start of each practical session.
- The *answers* to the questions are available a week after each practical session.

Course Material

ELEUM:

- The slides of the lecture will become available after the lecture.
- Per practical session *literature references* are provided. We strongly recommend you to read these before the session.
- The exercises are available before the start of each practical session.
- The *answers* to the questions are available a week after each practical session.

BOOKS (available at the "studielandschap")

- Understanding bioinformatics M Zvelebil and J.O. Baum
- Bioinformatics:
 Sequence and genome analysis
 David W. Mount
- Bioinformatics and Functional Genomics J. Pevsner
- Learning Perl
 Randal L. Schwartz and Tom Phoenix



Subjects of bioinformatics track in BW2.3

- 1. Introduction Bioinformatics and Biological Databases
- 2. Protein Structures

Next block 2.4 the programme will continue:

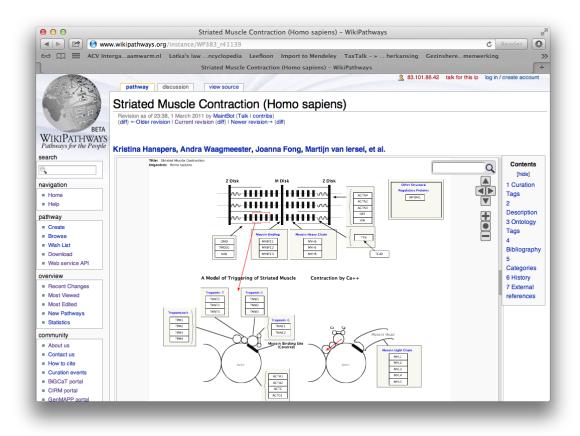
Gene Expression data

Pathway analysis – Network analysis

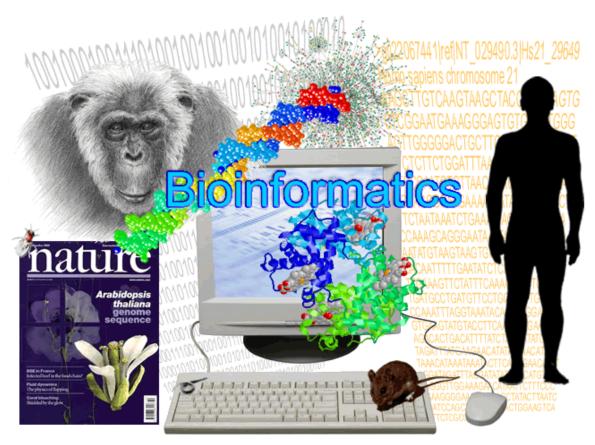
Introduction Programming:
Matlab
2 sessions in

Introduction to Bioinformatics

Pathways

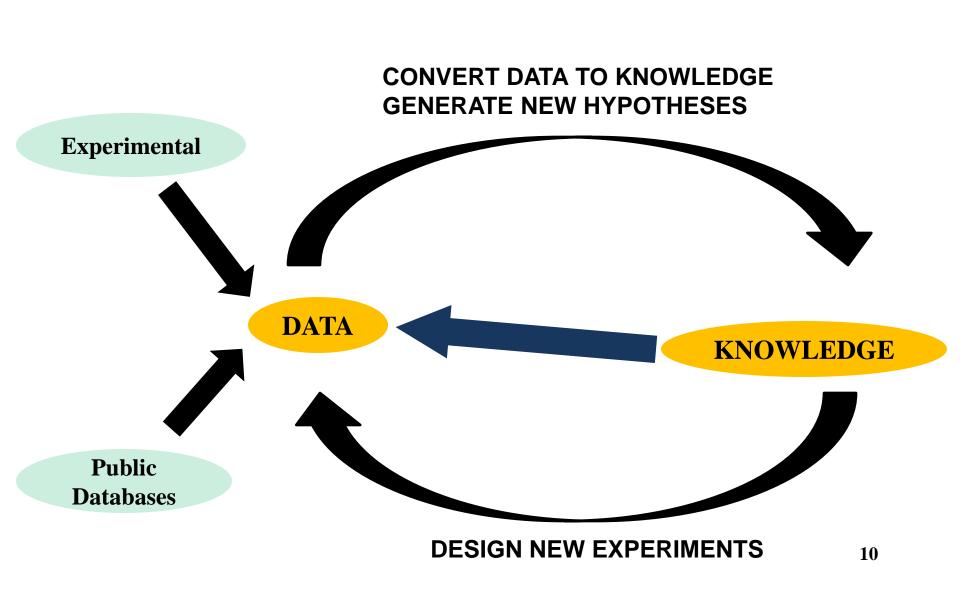


What is Bioinformatics?

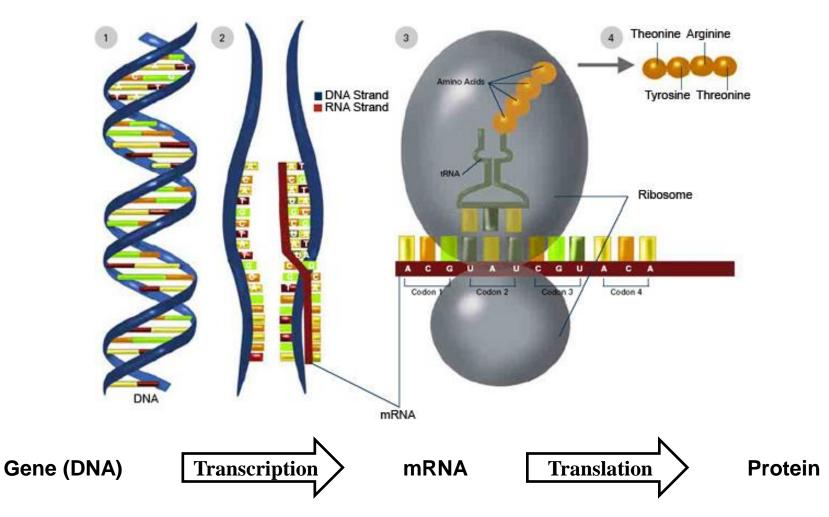


Bioinformatics uses **"informatics" techniques** (from applied math, computer science, statistics, etc.) to **understand** and **organize** biological information, like genes, proteins and molecules on a **large-scale.** 9

Why Bioinformatics?

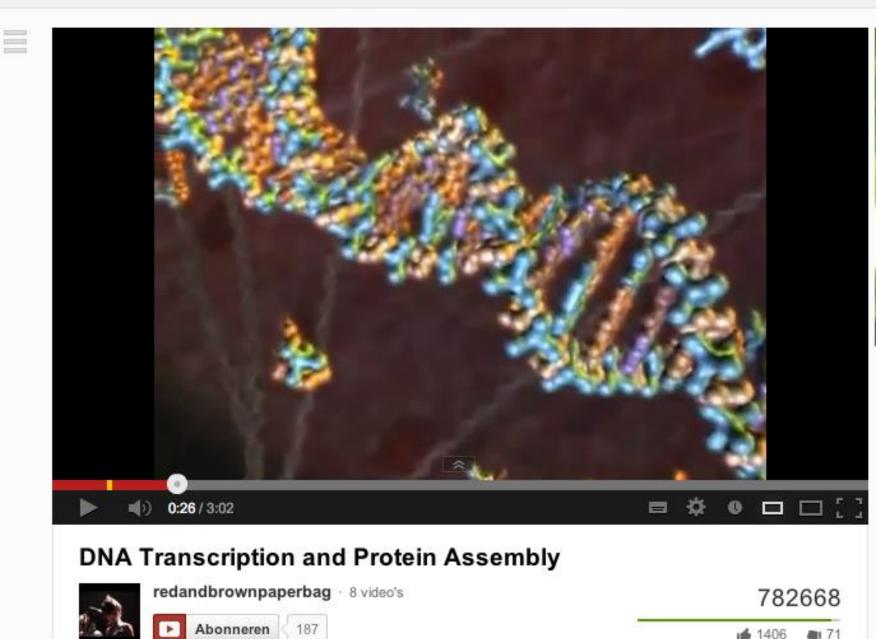


Central dogma of Molecular Biology



Cells express **different** subset of the genes in different tissues and under different conditions

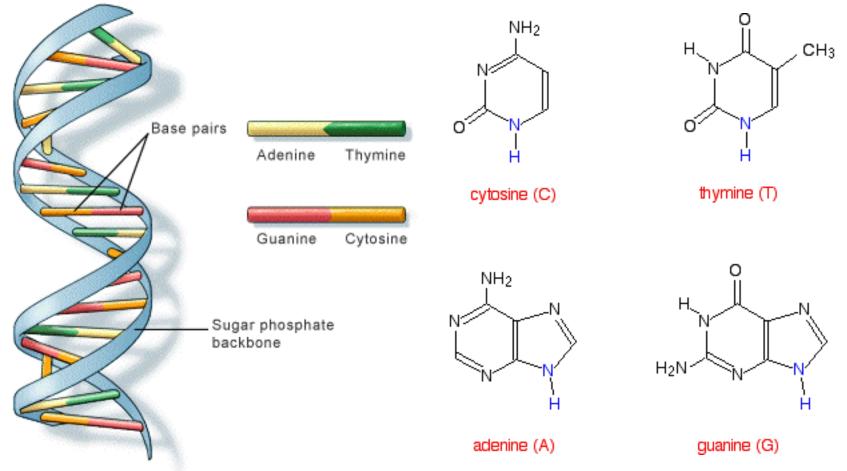




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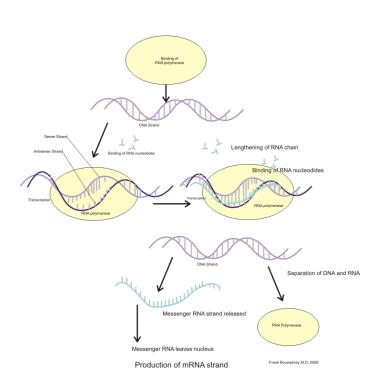
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DNA



U.S. National Library of Medicine

mRNA



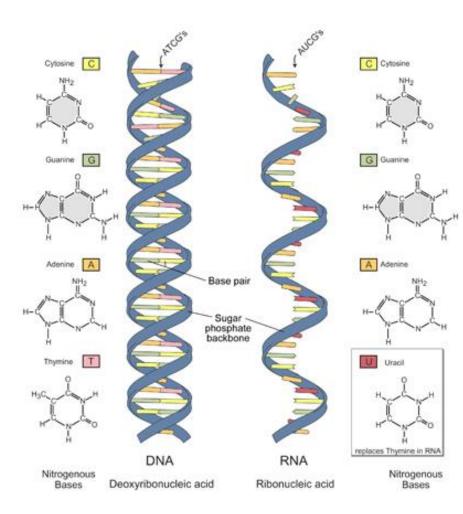
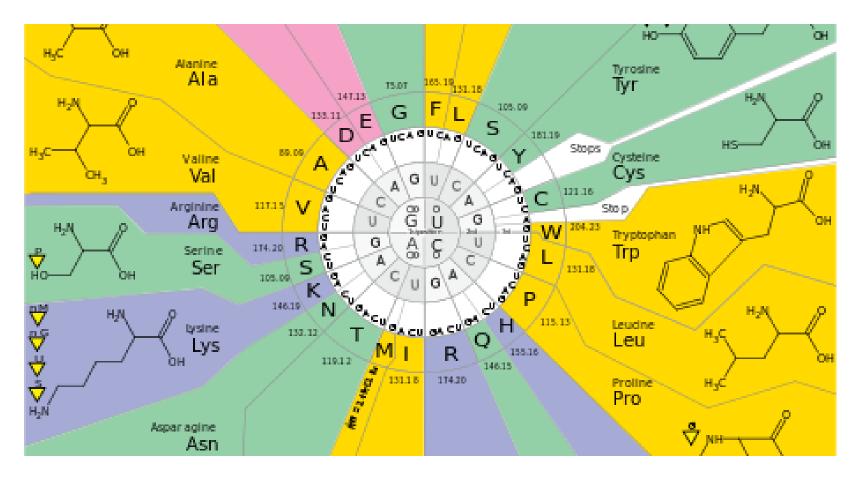


Image adapted from: National Human Genome Research Institute.

Proteins



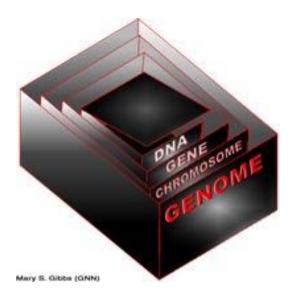
Genome Sequences / the Human Genome Project

AGTCCGCGAATACAGGCTCGGT



A genome is the collection of DNA that comprises an organism.

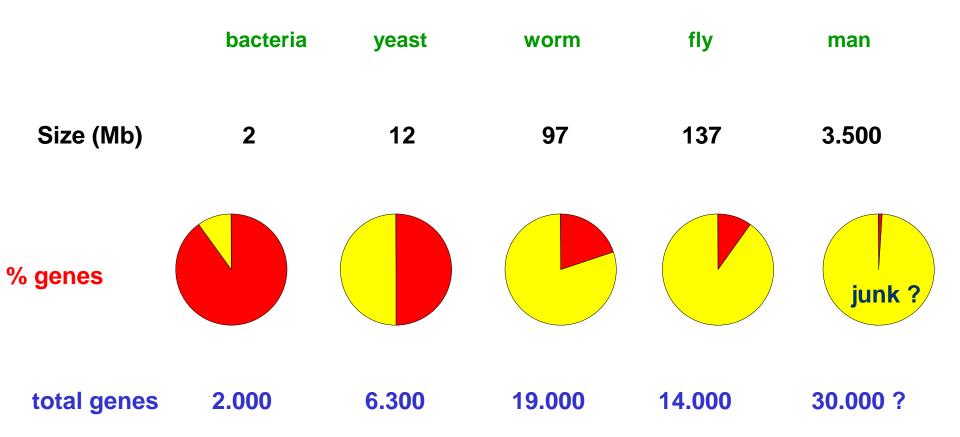
Today we have assembled the sequence of hundreds of genomes.



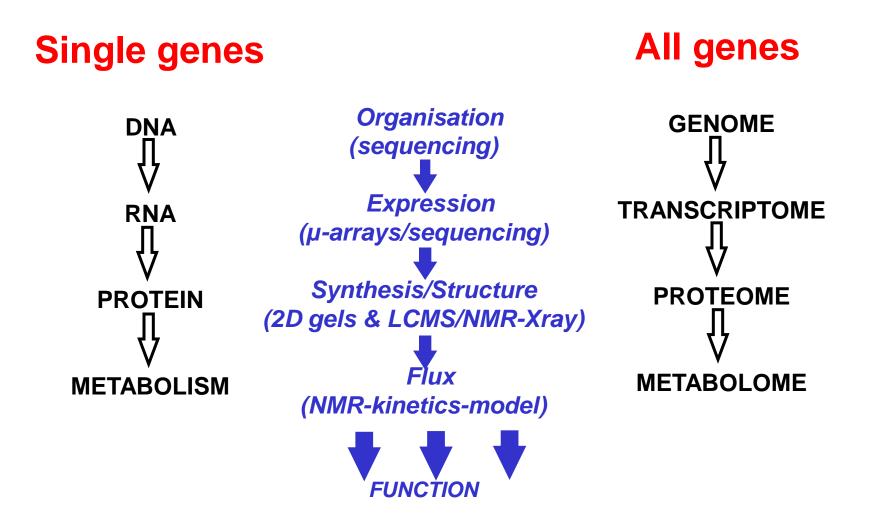
The genome is divided into chromosomes, chromosomes contain genes, and genes are made of DNA.

Each one of earth's organism has its own distinctive genome (except identical twins).

Genome content



Functional genomics



The vertebrate genomes available in Ensembl

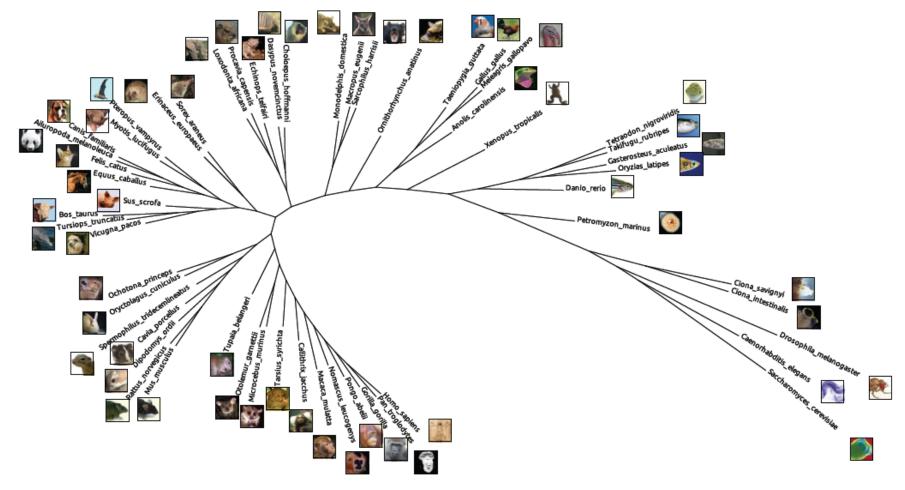


Image obtained using Dendroscope (D.H. Huson et al. "Dendroscope- An interactive viewer for large phylogenetic trees", BMC Bioinformatics 8:460, 2007)

Human Genome project

Introduction video

http://www.youtube.com/watch?v=N4i6lYfYQzY&list=PLF0701633C91835BF&index=1)

- Strategies
- Conclusions

International Human Genome Sequencing Consortium, Finishing the euchromatic sequence of the human genome. Nature 431, 931-945 (21 October 2004).

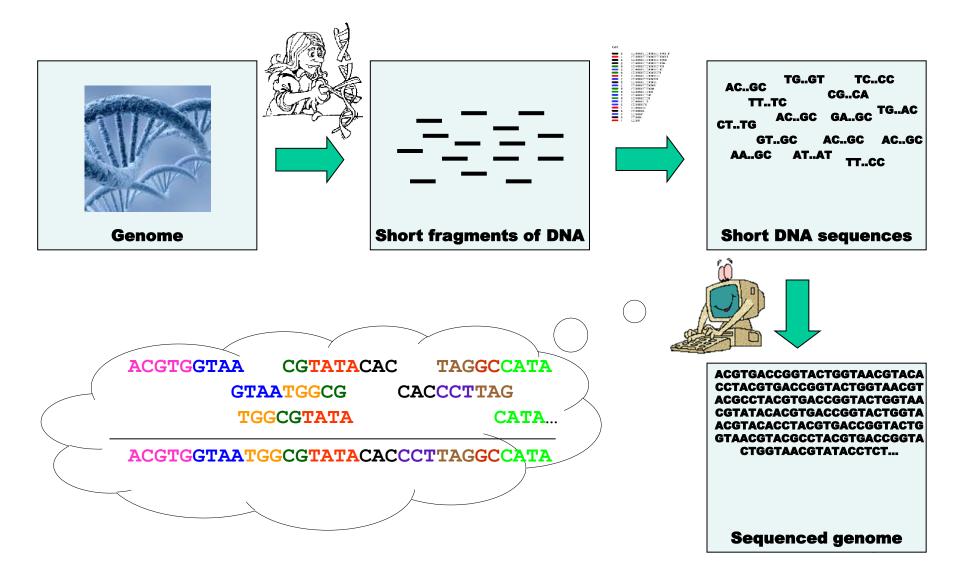
Overview of genome analysis

There are two main strategies for sequencing genomes

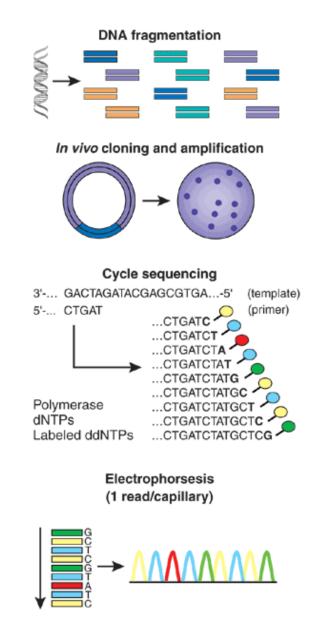
An approach used to decode an organism's genome by shredding it into smaller fragments of DNA which can be sequenced individually. The sequences of these fragments are then ordered, based on overlaps in the genetic code, and finally reassembled into the complete sequence.

The 'whole genome shotgun' (WGS) method is applied to the entire genome all at once, while the 'hierarchical shotgun' method is applied to large, overlapping DNA fragments of known location in the genome.

Genome sequencing

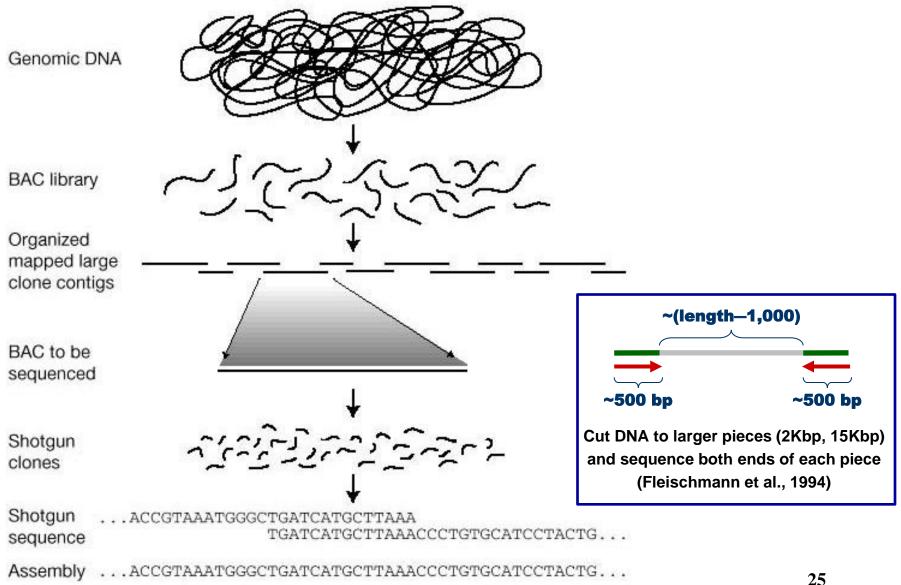


Workflow of Sanger sequencing



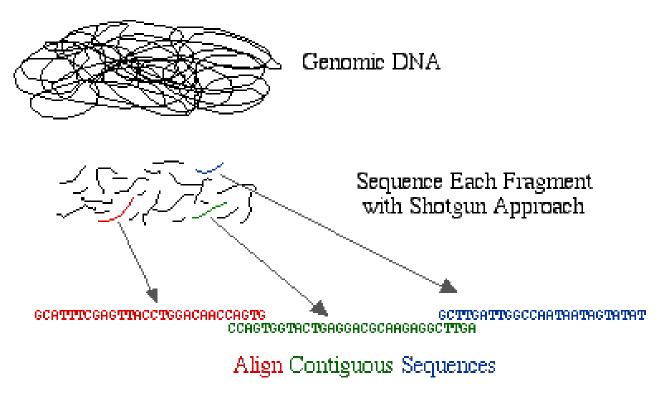
24 Jay Shendure & Hanlee Ji, Nature Biotechnology 26, 1135 - 1145 (2008)

Hierarchical shotgun sequencing



Source: IHGSC (2001)

Whole genome shotgun sequencing



GCATTTCGAGTTACCTGGACAACCAGTGGTACTGAGGACGCAAGAGGCCTTGATTGGCCAATAATAGTATAT

Generate Finished Sequence

26 Source: IHGSC (2001)

When has a genome been fully sequenced?

A typical goal is to obtain five to ten-fold coverage.

Finished sequence: a clone insert is contiguously sequenced with high quality standard of error rate 0.01%. There are usually no gaps in the sequence.

Draft sequence: clone sequences may contain several regions separated by gaps. The true order and orientation of the pieces may not be known.

Main conclusions of human genome project (1)

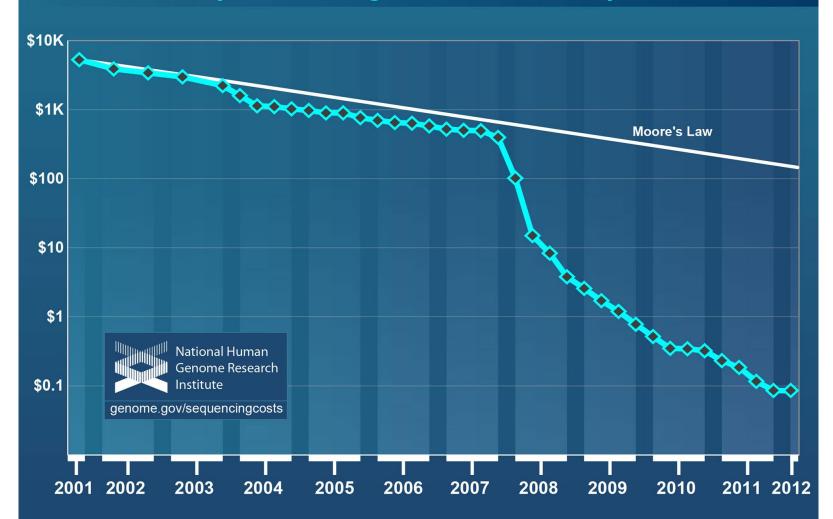
- 1. We have about the same number of genes as fish and plants, and not that many more genes than worms and flies 20,000-30,000
- 2. The human proteome is far more complex than the set of proteins encoded by invertebrate genomes alternative splicing
- 3. Hundreds of human genes were acquired from bacteria by lateral gene transfer
- 4. 98% of the genome does not code for genes and >50% of the genome consists of repetitive DNA
- 5. Segmental duplication is a frequent occurrence in the human genome

Main conclusions of human genome project (2)

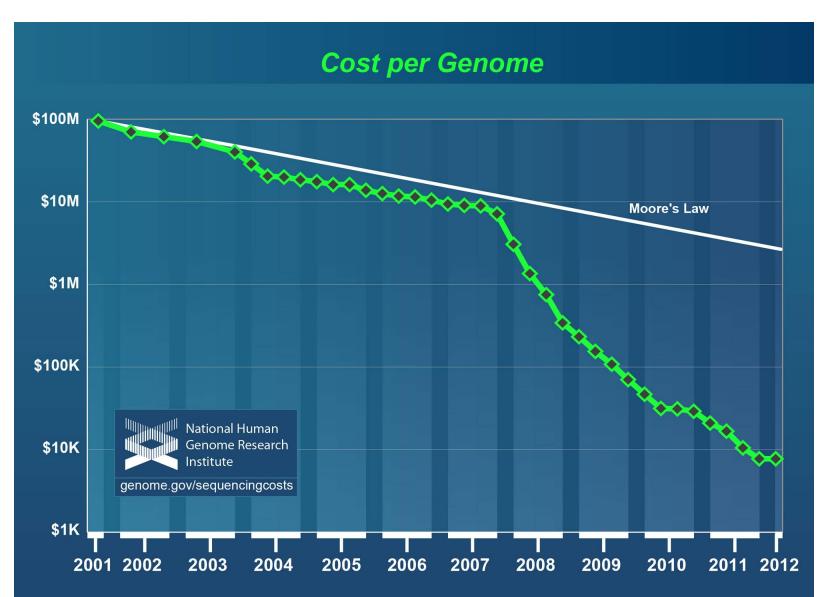
- 6. There are 300,000 Alu repeats in the human genome
 - These are about 300 base pairs and contain an Alul restriction enzyme site.
 - They occupy 3% of the genome and may confer some benefit
- 7. The mutation rate is about twice as high in male meiosis than female meiosis; most mutation probably occurs in males
- 8. 1.5 2 million single base pair changes or single nucleotide polymorphisms (SNPs) were originally identified.
 - Currently, dbSNP at NCBI over 10 million human SNPs
 - Half of these have been validated
 - A SNP occurs every 100 to 300 base pairs
 - Fewer than 1% of SNPs alter protein sequence
- 9. Noncoding RNAs are also important (for example miRNAs)

Sequencing the Human Genome

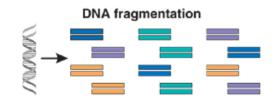
Cost per Raw Megabase of DNA Sequence



Sequencing the Human Genome



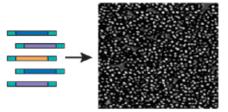
Nowadays: Work flow second-generation sequencing



In vitro adaptor ligation



Generation of polony array



Cyclic array sequencing (>10⁶ reads/array) Cycle 1 Cycle 2 Cycle 3 Cycle 2 Cycle 3 Cycle 2 Cycle 3 Cycle 2 Cycle 3 Cycle 3 Cycle 2 Cycle 3 Cycle 3 Cycle 2 Cycle 3 Cycle 3 Cycle 4 Cycle 2 Cycle 3 Cycle 5 C

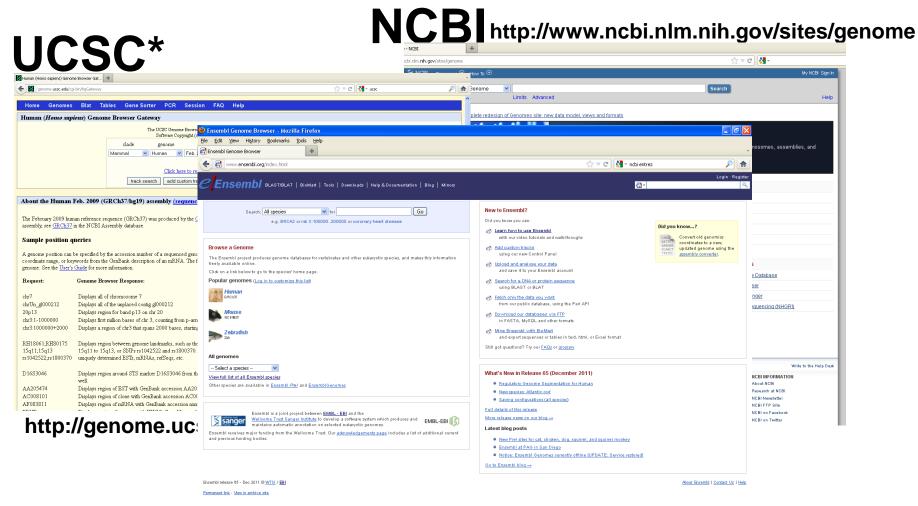
32 Jay Shendure & Hanlee Ji, Nature Biotechnology 26, 1135 - 1145 (2008)

Gateways to the genome sequences

Genome browsers: gateways to the genome sequences

- Over the last few decades a gigantic amount of information on DNA sequences, gene locations, gene transcripts, protein functions and so on has been gathered
- Now we will discuss several websites that provide all this information collection, and that you will use in the afternoon session
- They all contain essentially the same information, but have a differente interface, look-and-feel, viewing options

Genome Browsers



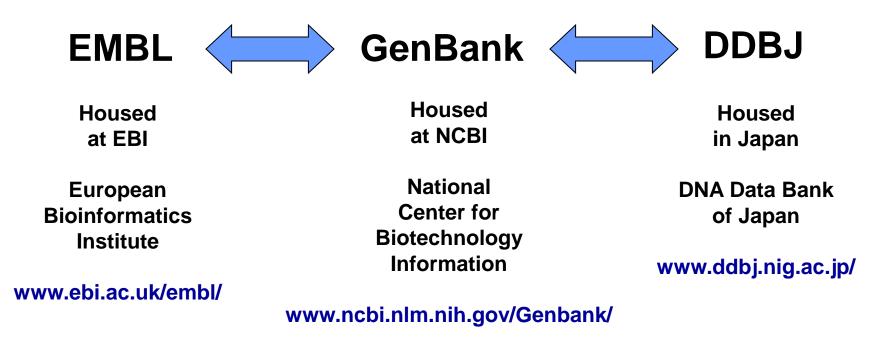


http://www.ensembl.org/

* We will use the UCSC browser later during the course

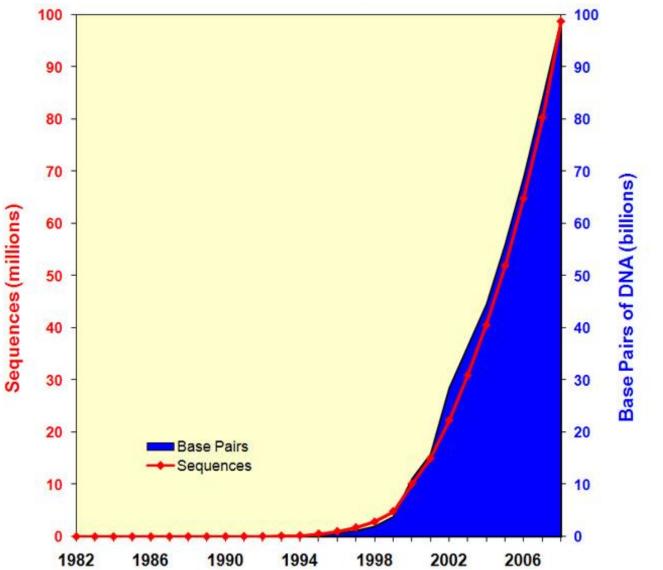
Nucleotide databases

The underlying raw DNA sequences are identical



Hundreds of thousands of species are represented

Growth of GenBank (1982-2008)



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NCBI nucleotide databases

GenBank

- Individual submissions
- Bulk submissions (Genome centers)
 - High throughput sequencing (DNA)
 - Expressed Sequence Tags (mRNA)
- RefSeq
 - Curated subset of GenBank
 - "Reference" sequence
 - Single sequence per locus / molecule

Protein sequence databases

- NCBI
 - RefSeq and Protein
- EBI
 - Swiss-Prot and TrEMBL \rightarrow UniProt
- Translated from nucleotide sequence
- Curated
- Combined

Accession numbers (Identifiers)

Label to unambiguously identify a sequence

Examples (all for retinol-binding protein, RBP4):

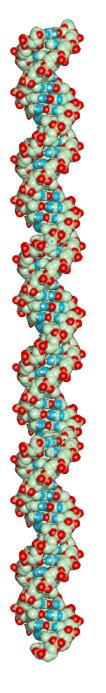
DNA	X02775 NT_030059 Rs7079946	GenBank genomic DNA sequence Genomic contig dbSNP (single nucleotide polymorphism)
RNA	N91759.1 NM_006744	An expressed sequence tag (1 of 170) RefSeq DNA sequence (from a transcript)
protein	NP_007635 AAC02945 Q28369 1KT7	RefSeq protein GenBank protein UniProt protein Protein Data Bank structure record

From Sequence to Genes: where are the genes?

- Gene prediction
 - Extrinsic
 - Search for genes based on observed mRNA / Protein sequences
 - UniGene
 - Ab initio
 - Predict genes based on genomic sequence alone
 - Promoter sequence
 - Poly(A) tail binding sites, CG content (higher in genes), splicing sites

UniGene

- Predict genes based on ESTs
- EST:
 - DNA sequence corresponding to mRNA from expressed gene
 - ~500 base pairs long
 - Sequenced from a cDNA library
- Cluster ESTs from many cDNA libraries to predict distinct genes



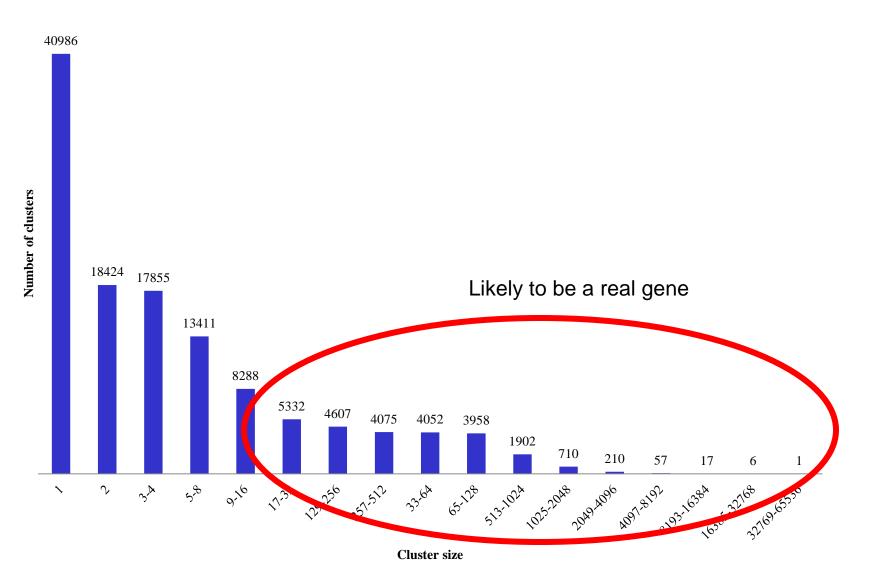
EST clusters

This is a gene with 1 EST associated;

the cluster size is 1

This is a gene with 10 ESTs associated; the cluster size is 10

UniGene clusters



44

Ensembl website (1)

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Zebrafish	Mine Ensembl with BioMart	
200 CH	and export sequences or tables in text, html, or Excel format	
	Still got questions? Try our <u>FAQs</u> or <u>glossary</u>	
All genomes		
Select a species 💌	What's New in Release 65 (December 2011)	
View full list of all Ensemblispecies	Regulatory Genome Segmentation for Human	
Other species are available in Ensembl Pre/ and EnsemblGenomes	New species: Atlantic cod	
	Saving configurations (all species)	
Ensembl is a joint project between EMBL - EBI and the	Full details of this release	
Wellcome Trust Sanger Institute to develop a software system which produces and maintains automatic annotation on selected eukaryotic genomes.	More release news on our blog	
Ensembl receives major funding from the Wellcome Trust. Our <u>acknowledgements page</u> includes a list of additional current	Latest blog posts	
and previous funding bodies.	New Prel sites for cat, chicken, dog, squirrel, and squirrel monkey	
	Ensembl at PAG in San Diego	
	Notice: Ensembl Genomes currently offline [UPDATE: Service restored]	

<u>Go to Ensembl blog \rightarrow </u>

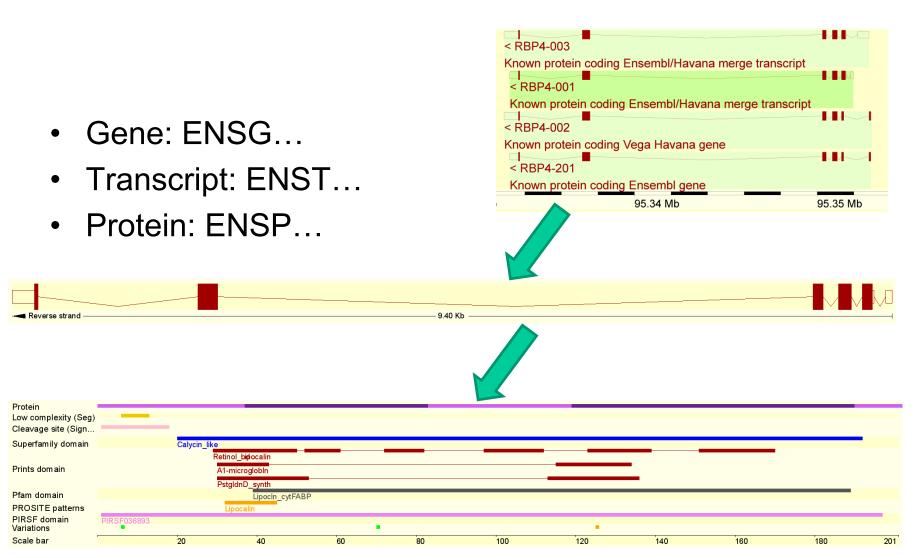


Ensembl website (2)

Ensembl genome browser 65: Homo :	apiens +				-
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 Transcript summary Supporting evidence (14) 		Hansenpt. RBF -			
E Sequence	Description retinol binding protein 4, plasma [S	Source:HGNC Symbol;Acc:9922]			
Exons (6)	Location Chromosome 10: 95,351,444-95,38	<u>61,420</u> reverse strand.			
- cDNA - Protein	Gene	ENSG00000138207 - This gene has	6 transcripts		
□ □· External References					
E 📙 – General identifiers (41)	Show/hide columns		Filter		
│	Name Transcript ID 💠 Length (bp) 💠	Protein ID 🕴 Length (aa) 🍦	Biotype 🔶 CCDS 🔶		
- Ontology graph (38)		ENSP00000360519 201	Protein coding CCDS31249		
│	RBP4-002 ENST00000371469 1016 E	ENSP00000360524 199	Protein coding -		
Population comparison	RBP4-003 ENST00000371467 1314 E	ENSP00000360522 201	Protein coding <u>CCDS31249</u>		
🗆 Comparison image	RBP4-201 ENST00000371463 838 E	ENSP00000360518 199	Protein coding -		
E ⊢ Protein Information	RBP4-004 ENST00000471333 399	No protein product -	Processed transcript -		
– Domains & features (16)	RBP4-005 ENST00000471469 360	No protein product -	Processed transcript -		
Variations (18)	Transcript and Gene level displays				
E ⊨ External Data E ⊨ Personal annotation	Views in Ensembl are separated into gene based viev	ve and transcript based views accordi	ing to which level the information is m	ore appropriately associated with. This view is a t	transcrit
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	Alternative transcripts This transcript corresponds	to the following database identific	ers:		
	Havana transcript: <u>OTTH</u>	UMT0000049431 (version 1) [view al	l locations]		
	Ensembl release 65 - Dec 2011 © WTSI / EBI			About Ensembl Contact Us	l Heln

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Ensembl identifiers



NCBI website (1)

NCBI Home Welcome to NCBI Resource List (A2) All Resources Publed All Resources Data & Software PubMed Central Data & Software PubMed Central PubMed Central DNA & RNA Domains & Structures PubMed Central Genes & Expression 0 foots: Analyze data using NCBI software PubMed Central Domains & Structures 0 foots: Analyze data using NCBI software PubMed Central Pownloads: Get NCBI data or software Genomes Genome Domains: Submit data to GenBank or other NCBI databases SNP Gene Protein SubChamissions: Submit data to GenBank or other NCBI databases Protein Proteins NCBI YouTube channel PubChem Sequence Analysis Learn how to get the most out of NCBI tools and databases with video tutorials on the NCBI YouTube Channel. OCS and databases with video tutorials on the NCBI YouTube Channel. Training & Tutorials Col Stand databases with video tutorials on the NCBI YouTube Channel. NCBI Discovery Workshop: A Practical Hands-On Course NCBI Discovery Workshop: A Practical Hands-On Course Yatan 2012 Yatan 2012	www.ncbi.nlm.nih.gov			☆ ▼ C	🚼 🛪 ncbi	
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NCBI Education

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NCBI website (2)

www.ncbi.nlm. nih.gov /sites/gquery		🏫 マ C 🛛 🚼 マ ncbi entrez					
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EARCH SITE MAP PubMed	All Databases	Human	Genome	GenBank	Map Viewer	BLAST	
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Protein: sequence database				data for clone resources			
Genome: whole genome seque	nces		UniSTS: markers a	nd mapping data		8	
Structure: three-dimensional m	nacromolecular structures	0	PopSet: population	n study data sets		0	
Taxonomy: organisms in GenBa	ank	0	GEO Profiles: expression and molecular abundance profiles		0		
SNP: short genetic variations		Ø	GEO DataSets: exp	perimental sets of GEO data		Ø	
😡 db Var: Genomic structural varia	tion	0	Epigenomics: Epige	enetic maps and data sets		0	
Gene: gene-centered information	on	0	💋 🛛 PubChem BioAssa	y: bioactivity screens of chemica	al substances	Ø	
() SRA: Sequence Read Archive		0	PubChem Compou	nd: unique small molecule chen	nical structures	0	
BioSystems: Pathways and systems:	tems of interacting molecules	@ (PubChem Substan	ce: deposited chemical substan	ice records	e	
HomoloGene: eukaryotic homol	logy groups	0	Protein Clusters: a	collection of related protein se	quences	0	
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NCBI identifiers

- RefSeq:
 - Chromosome: NC_
 - mRNA: NM_
 - Protein: NP_
- Genbank:
 - Many types of IDs
- Entrez gene ID:
 - Number

- OMIM ID:
 - Number
- Pubmed ID:
 Number
- UniGene ID:
 - Abbreviation of species.number (e.g. Hs.50223)