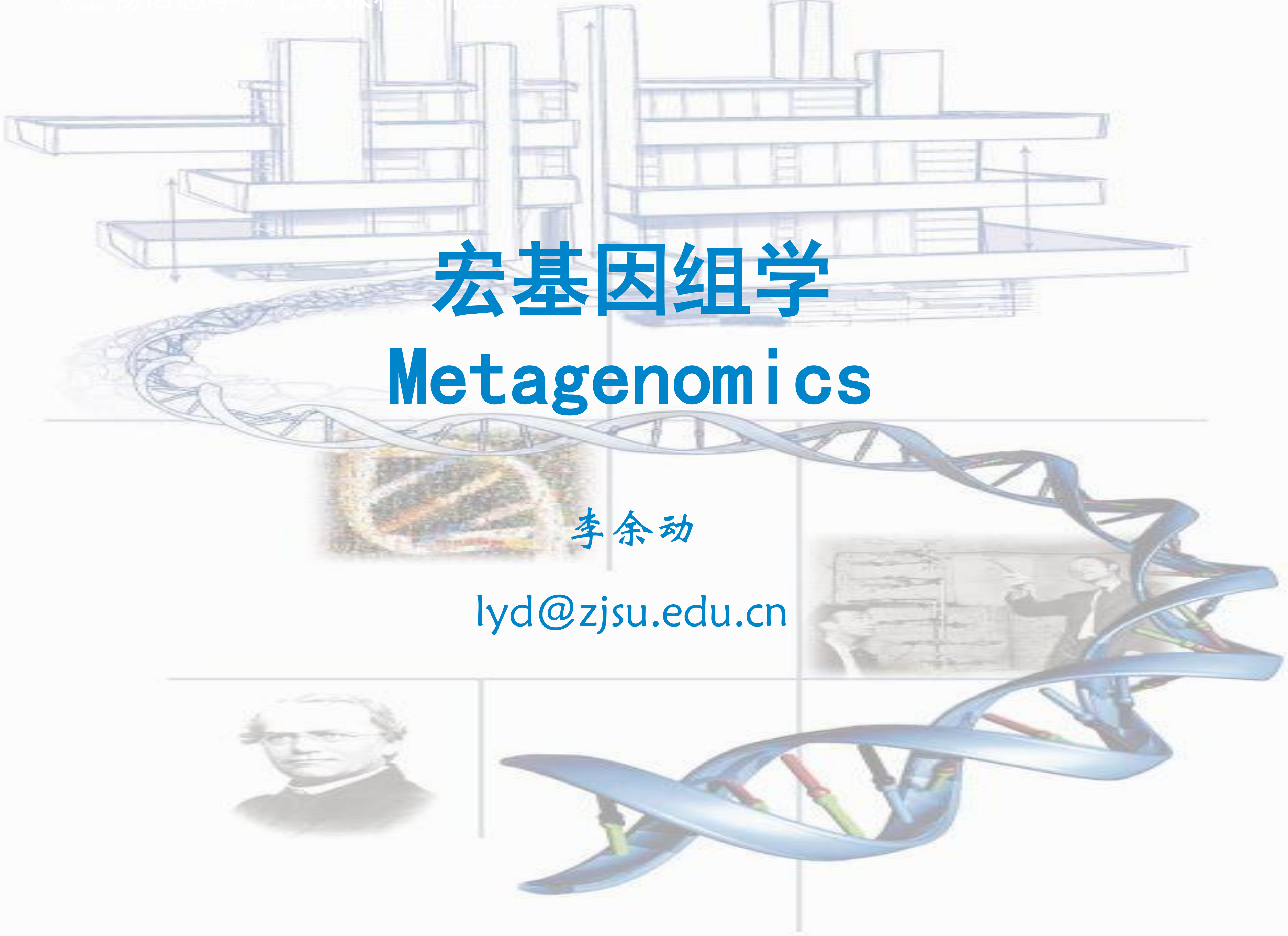


宏基因组学
Metagenomics

李余动

lyd@zjsu.edu.cn

Our world is full of microbes (微生物无处不在)

human

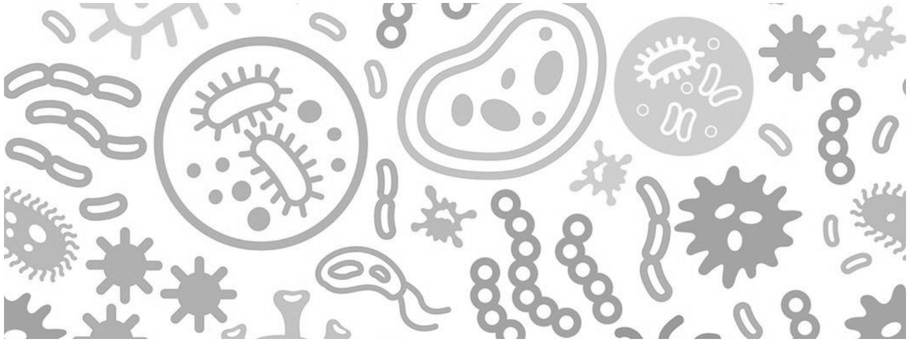
soil

(~$10^{9\text{-}10}$个微生物/1克土)

ocean

hot springs

Upon a closer look…



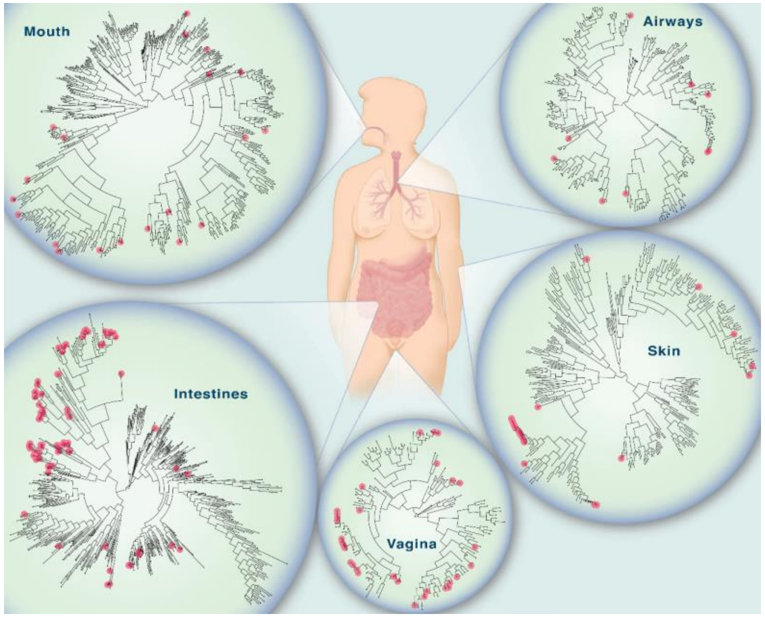
Microbes

- bacteria
- fungi
- viruses
- archaea
- protists (原生生物)
- ……

微生物(microbes)是指“一切肉眼看不见或看不清的微小生物的总称”。

Human Microbiome (人体微生物组)

- 在人体内及体表生活着大量的微生物，这些微生物群及其遗传信息的总和被称为人体微生物组，主要分布于肠道、皮肤、口腔、呼吸道、泌尿生殖道。
- 现代人生活方式的改变，导致肠道微生物消失，这影响身体的健康状态，是人类在21世纪面临的巨大挑战。



(Lee & Mazmanian, 2010, Science)

添加干酪乳杆活性菌
源自养乐多专利菌种

微生物组研究的核心：测序 + 数据分析

贵

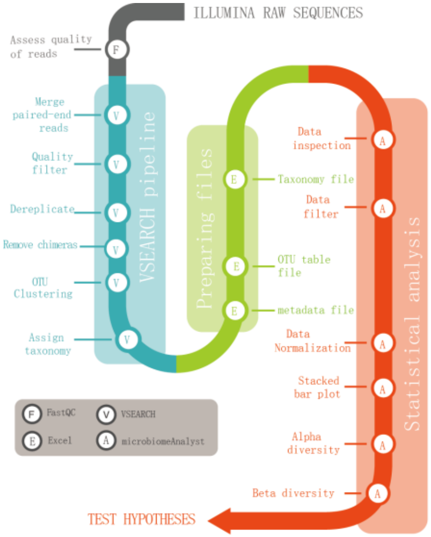
- 设备与试剂贵，不易获得

繁

- 操作步骤多，易出错

难

- 数据量大，分析难度大



微生物组仿真实验提供学生模拟实验操作机会，使学生直观感受微生物组研究过程。

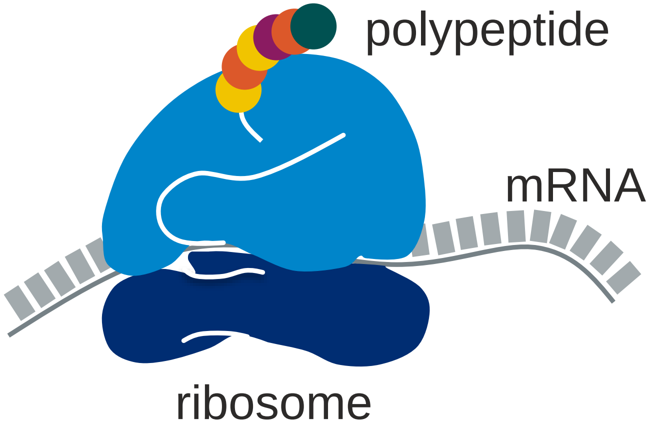
How do we identify members of microbiomes（如何鉴定微生物）？

How can we study it by DNA sequencing（测序）？

The 16S ribosomal RNA as a microbial fingerprint

- Fingerprints are *both* **universally present** on all people *and* **unique**

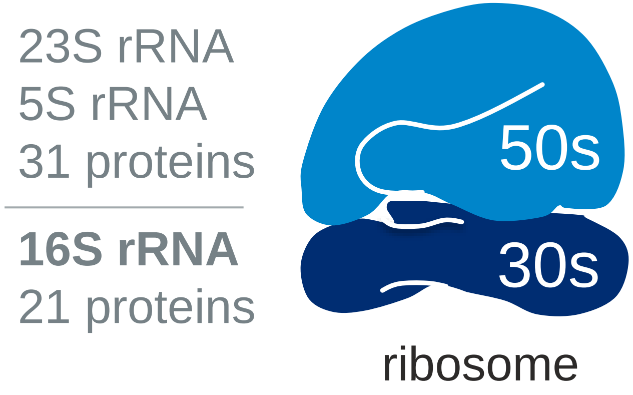
- The **ribosome** (核糖体) is essential for survival across all kingdoms of life and is thus **highly conserved**



核糖体是蛋白质翻译的场所

16S rRNA：细菌的“分子化石”

- Specifically, the **16S rRNA** component of the ribosome is highly **conserved** among bacteria/archaea, yet contains **hypervariable** regions.



23S rRNA
5S rRNA
31 proteins

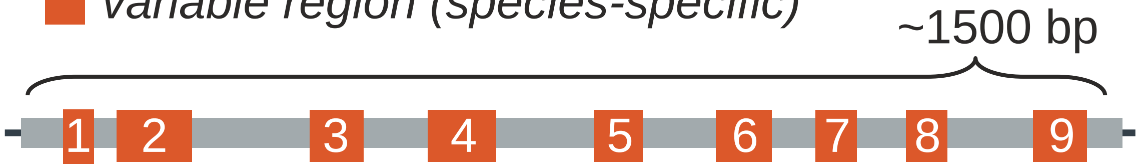
16S rRNA
21 proteins

16S rRNA在细菌/古菌的进化过程中高度保守，又有高度变异的区域。

→ **16S rRNA contains 9 variable regions**

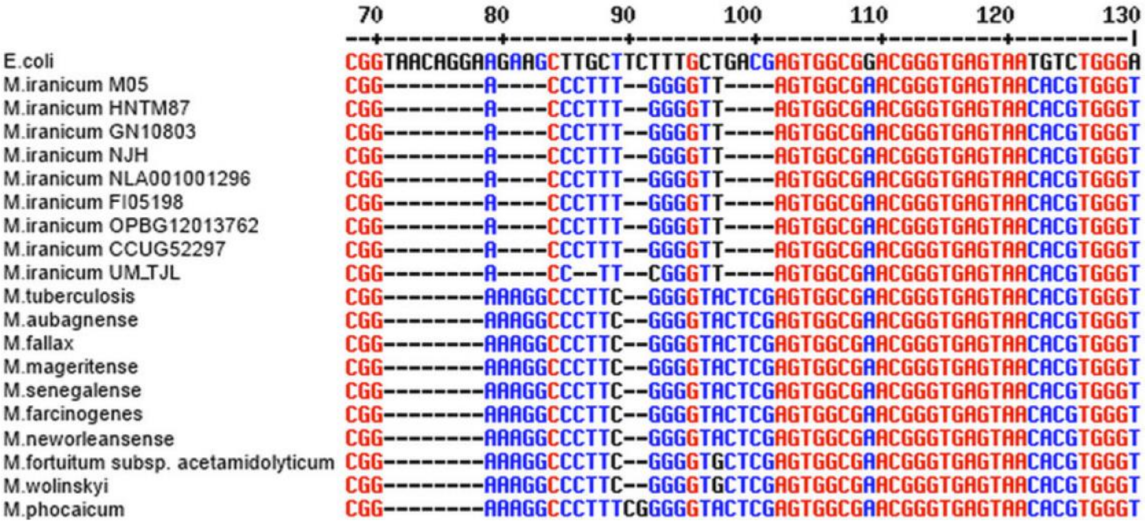
■ *conserved region (non-specific)*

🟧 *variable region (species-specific)*



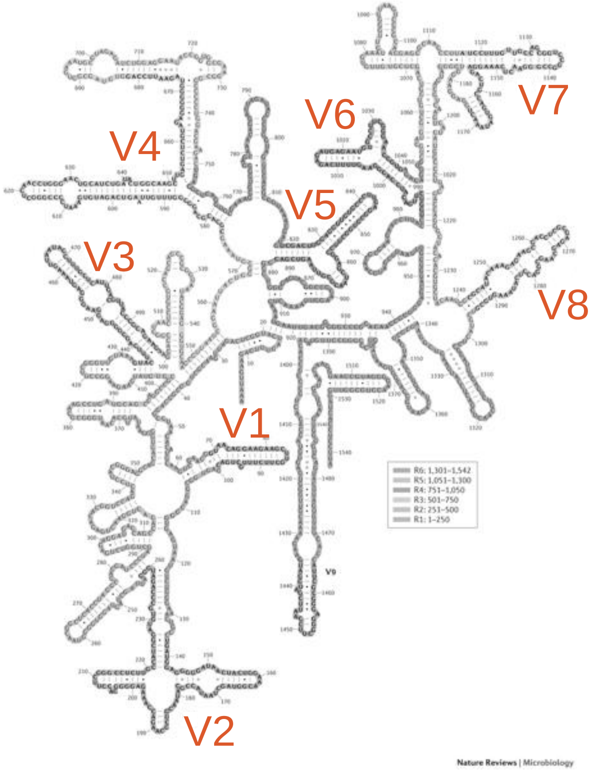
Hypervariable regions (可变区) can be used for species identification

→ **More distantly related species exhibit more divergent 16S rRNA sequences**



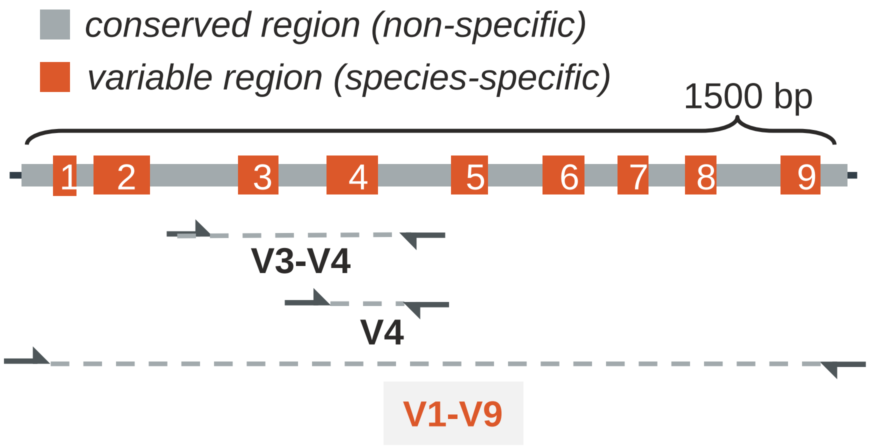
```
                                     70        80        90       100       110       120       130
                                    --+---------+---------+---------+---------+---------+---------|
E.coli                              CGGTAACAGGAAGAAGCTTGCTTCTTTGCTGACGAGTGGCGGACGGGTGAGTAATGTCTGGGA
M.iranicum M05                      CGG--------A----CCCTTT--GGGGTT----AGTGGCGAACGGGTGAGTAACACGTGGGT
M.iranicum HNTM87                   CGG--------A----CCCTTT--GGGGTT----AGTGGCGAACGGGTGAGTAACACGTGGGT
M.iranicum GN10803                  CGG--------A----CCCTTT--GGGGTT----AGTGGCGAACGGGTGAGTAACACGTGGGT
M.iranicum NJH                      CGG--------A----CCCTTT--GGGGTT----AGTGGCGAACGGGTGAGTAACACGTGGGT
M.iranicum NLA001001296             CGG--------A----CCCTTT--GGGGTT----AGTGGCGAACGGGTGAGTAACACGTGGGT
M.iranicum FI05198                  CGG--------A----CCCTTT--GGGGTT----AGTGGCGAACGGGTGAGTAACACGTGGGT
M.iranicum OPBG12013762             CGG--------A----CCCTTT--GGGGTT----AGTGGCGAACGGGTGAGTAACACGTGGGT
M.iranicum CCUG52297                CGG--------A----CCCTTT--GGGGTT----AGTGGCGAACGGGTGAGTAACACGTGGGT
M.iranicum UM_TJL                   CGG--------A----CC--TT--CGGGTT----AGTGGCGAACGGGTGAGTAACACGTGGGT
M.tuberculosis                      CGG--------AAAGGCCCTTC--GGGGTACTCGAGTGGCGAACGGGTGAGTAACACGTGGGT
M.aubagnense                        CGG--------AAAGGCCCTTC--GGGGTACTCGAGTGGCGAACGGGTGAGTAACACGTGGGT
M.fallax                            CGG--------AAAGGCCCTTC--GGGGTACTCGAGTGGCGAACGGGTGAGTAACACGTGGGT
M.mageritense                       CGG--------AAAGGCCCTTC--GGGGTACTCGAGTGGCGAACGGGTGAGTAACACGTGGGT
M.senegalense                       CGG--------AAAGGCCCTTC--GGGGTACTCGAGTGGCGAACGGGTGAGTAACACGTGGGT
M.farcinogenes                      CGG--------AAAGGCCCTTC--GGGGTACTCGAGTGGCGAACGGGTGAGTAACACGTGGGT
M.neworleansense                    CGG--------AAAGGCCCTTC--GGGGTACTCGAGTGGCGAACGGGTGAGTAACACGTGGGT
M.fortuitum subsp. acetamidolyticum CGG--------AAAGGCCCTTC--GGGGTGCTCGAGTGGCGAACGGGTGAGTAACACGTGGGT
M.wolinskyi                         CGG--------AAAGGCCCTTC--GGGGTGCTCGAGTGGCGAACGGGTGAGTAACACGTGGGT
M.phocaicum                         CGG--------AAAGGCCCTTTCGGGGGTACTCGAGTGGCGAACGGGTGAGTAACACGTGGGT
```

16S rRNA secondary structure



Nature Reviews | Microbiology

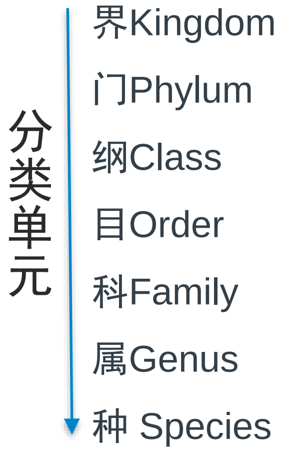
Highly conserved region (高度保守区): easy to target across all bacteria



conserved region (non-specific)
variable region (species-specific)
1500 bp
1
2
3
4
5
6
7
8
9
V3-V4
V4
V1-V9

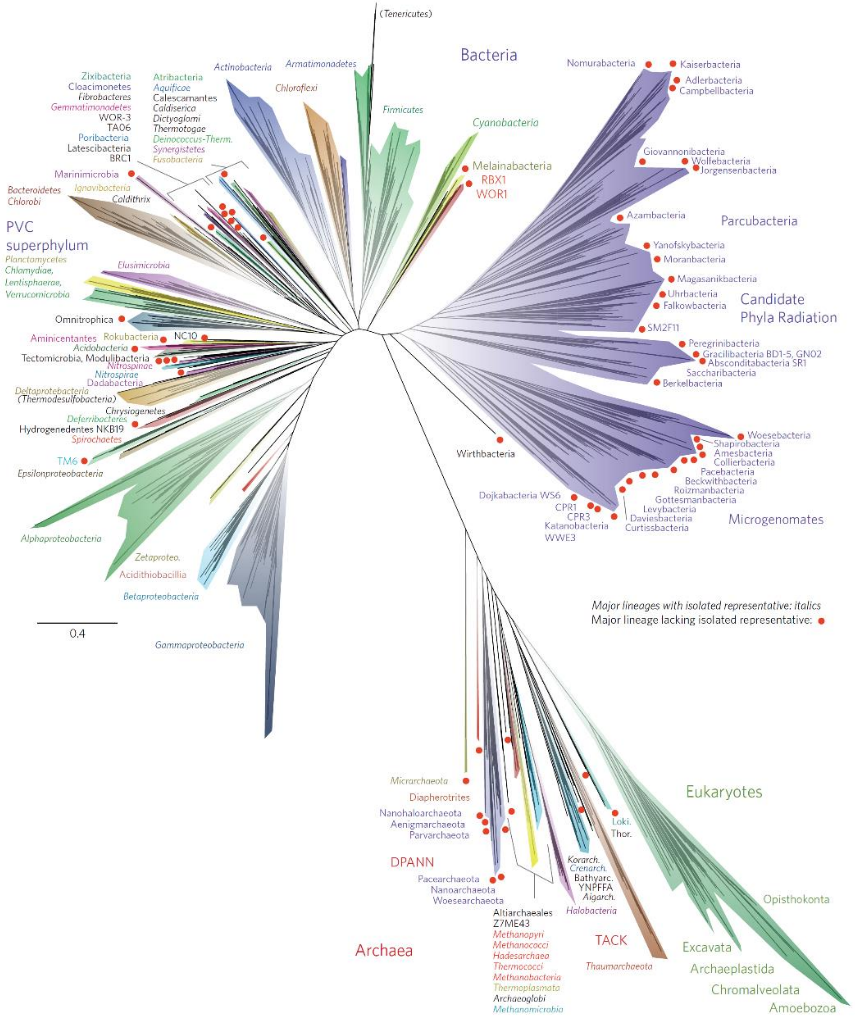
Length	Resolution
~465 bp	Genus, species
~255 bp	Family, genus
~1500 bp	Species, strain

在16S保守区域设计**PCR引物**，可扩增不同的可变区，其中V3/V4区其特异性好，数据库信息全，适合短读段的二代测序。



Tree of Life: 16S rRNA gene

生命三域：细菌、古菌与真核生物（Woese and Fox, 1977）



(Hug *et al.*, 2016, Nature Microbiology)

Carl Woese (1928-2012)

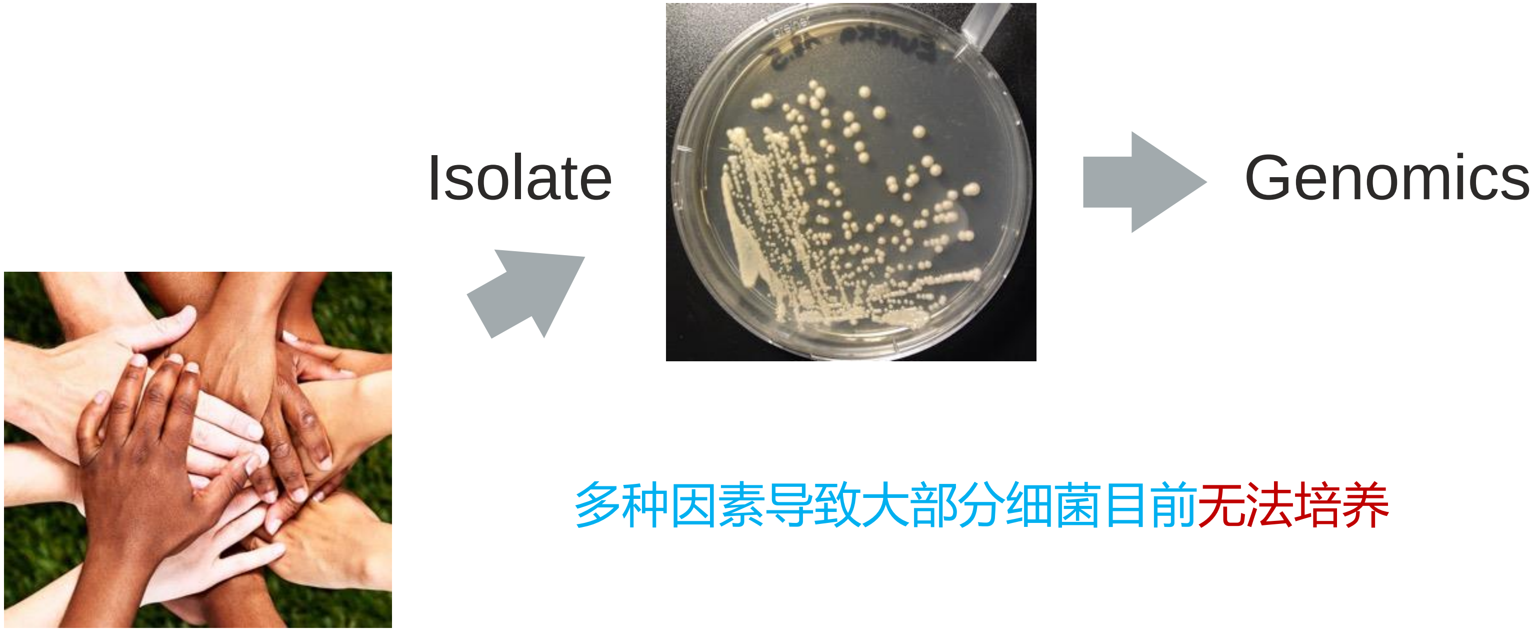
16S rRNA基因为什么作为分子标记?

- 在生物体间普遍分布，序列有高度保守性
- 又有可变区，在不同生物中有一定变化，而且有稳定的突变速率。
- 分子大小适中(1500bp)，可进行测序分析
- 在细胞中含量高(rRNA基因拷贝数多)，易分离纯化

How to analyze microbial communities(微生物群落)?

How can we better understand our microbiomes?

Microbial genomics suffers from lack of cultivation approaches(纯培养方法)

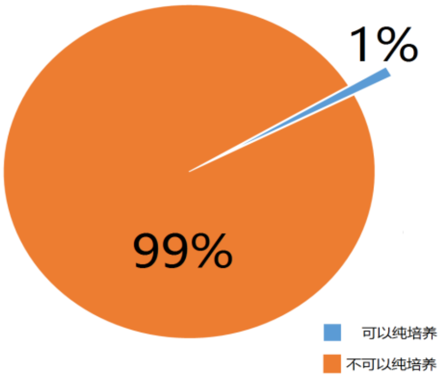


Isolate

Genomics

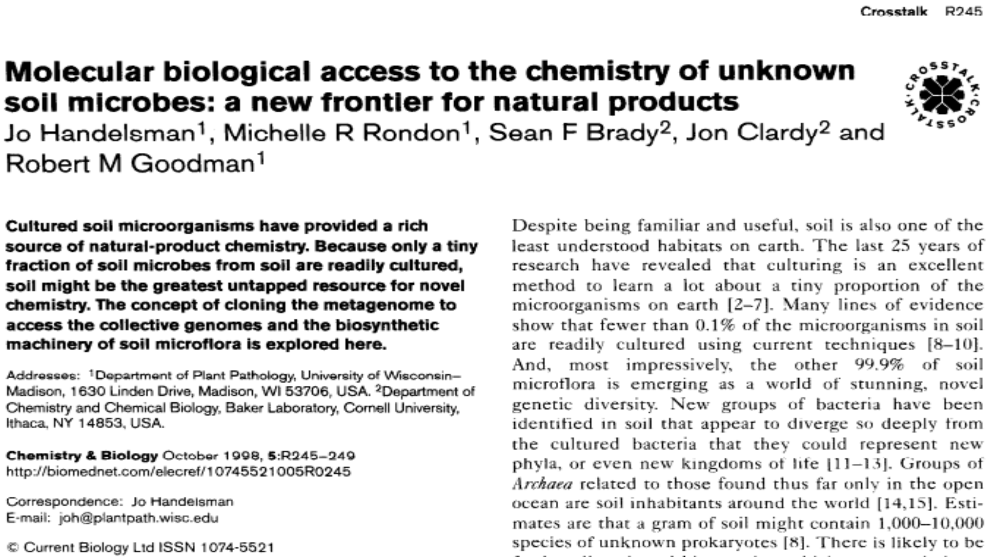
多种因素导致大部分细菌目前无法培养

“The estimate that fewer than 1% of the prokaryotes in most environments can be cultivated in isolation has produced a quandary: what is the significance of the field of modern microbial genomics if it is limited to culturable organisms?”
Schloss et al, Genome Biology, 2005



宏基因组学(Metagenomics)

- 宏基因组学（Metagenomics）又称环境微生物基因组学，是指不经过微生物培养阶段，采用直接提取环境中总DNA的方法，对微生物基因总和进行研究的一门新学科。
  - ○ Metagenomics is the study of genetic materials recovered directly from environmental samples. The broad field may also be referred to as environmental genomics, ecogenomics or community genomics.
- 宏基因组(Metagenome) 是由 Handelsman等1998年提出的新名词，其定义为“the genomes of the total microbiota found in nature”，即生境中全部微生物基因组的总和。
  - ○ **包含可培养的和不可培养的微生物**，目前主要指环境样品中的细菌和真菌。



Crosstalk R245

Molecular biological access to the chemistry of unknown soil microbes: a new frontier for natural products

Jo Handelsman[1], Michelle R Rondon[1], Sean F Brady[2], Jon Clardy[2] and Robert M Goodman[1]

Cultured soil microorganisms have provided a rich source of natural-product chemistry. Because only a tiny fraction of soil microbes from soil are readily cultured, soil might be the greatest untapped resource for novel chemistry. The concept of cloning the metagenome to access the collective genomes and the biosynthetic machinery of soil microflora is explored here.

Addresses: [1]Department of Plant Pathology, University of Wisconsin–Madison, 1630 Linden Drive, Madison, WI 53706, USA. [2]Department of Chemistry and Chemical Biology, Baker Laboratory, Cornell University, Ithaca, NY 14853, USA.

Chemistry & Biology October 1998, **5**:R245–249
http://biomednet.com/elecref/10745521005R0245

Correspondence: Jo Handelsman
E-mail: joh@plantpath.wisc.edu

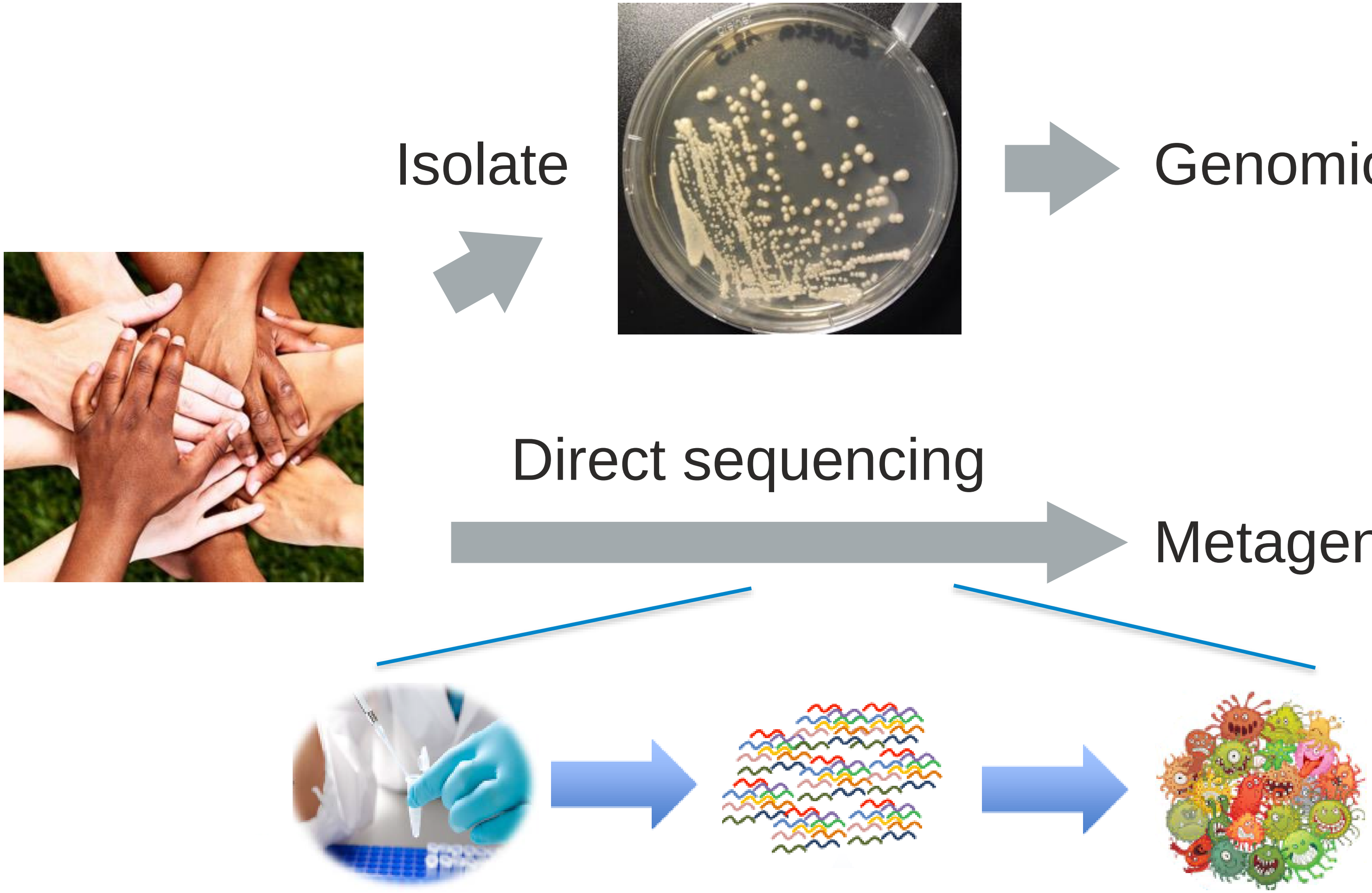
© Current Biology Ltd ISSN 1074-5521

Despite being familiar and useful, soil is also one of the least understood habitats on earth. The last 25 years of research have revealed that culturing is an excellent method to learn a lot about a tiny proportion of the microorganisms on earth [2–7]. Many lines of evidence show that fewer than 0.1% of the microorganisms in soil are readily cultured using current techniques [8–10]. And, most impressively, the other 99.9% of soil microflora is emerging as a world of stunning, novel genetic diversity. New groups of bacteria have been identified in soil that appear to diverge so deeply from the cultured bacteria that they could represent new phyla, or even new kingdoms of life [11–13]. Groups of *Archaea* related to those found thus far only in the open ocean are soil inhabitants around the world [14,15]. Estimates are that a gram of soil might contain 1,000–10,000 species of unknown prokaryotes [8]. There is likely to be

Jo Handelsman

Chemistry & Biology. 1998,5(10):R245-9

Metagenomics has revolutionized microbiome studies



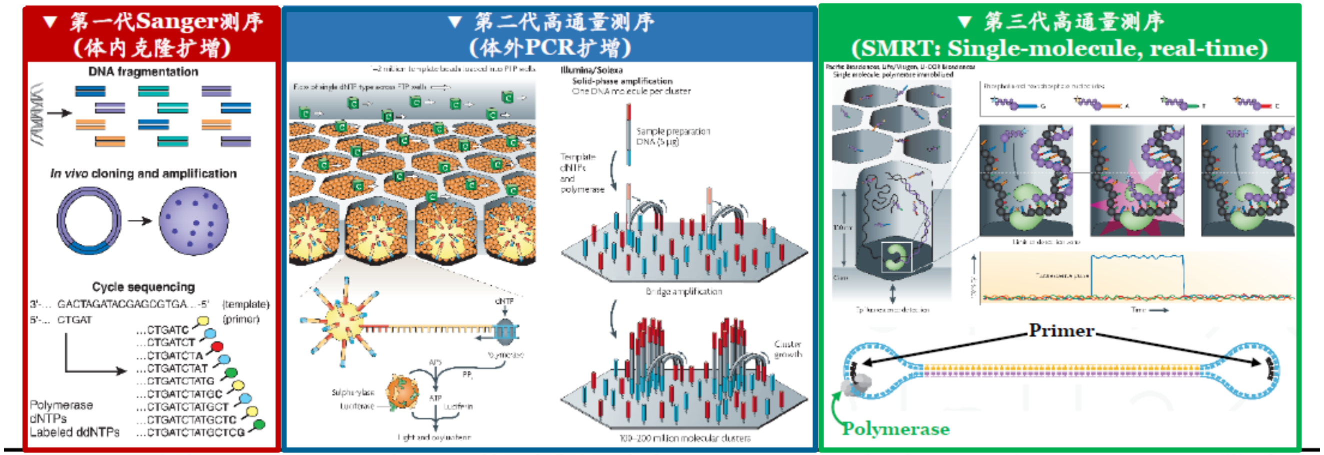
Isolate

Genomics

Direct sequencing

Metagenomics

下一代测序技术催生了宏基因组学
Next Generation Sequencing (NGS) technology



Method	Generation	Read length (bp)	Single pass error rate	No. of reads per run	Time per run	Cost per million bases
Sanger ABI 3730xl	1st	600–1000	0.001%	96	0.5–3 h	$500
454 (Roche) GS FLX+	2nd	700	1%	1×10^6	23 h	$8.57
Illumina HiSeq 2500 (High Output)	2nd	2 × 125	0.1%	8×10^9 (paired)	7–60 h	$0.03
Illumina HiSeq 2500 (Rapid Run)	2nd	2 × 250	0.1%	1.2×10^9 (paired)	1–6 days	$0.04
Ion Torrent	2nd	200	1%	8.2×10^7	2–4 h	$0.1
SOLiD 5500xl	2nd	2 × 60	5%	8×10^8	6 days	$0.11
PacBio RS II: P6-C4	3rd	Avg. 10–15 k	13%	$3.5–7.5 \times 10^4$	0.5–4 h	$0.40–0.80
Oxford Nanopore MinION	3rd	Avg. 2–5 k	38%	$1.1–4.7 \times 10^4$	50 h	$6.44–17.90

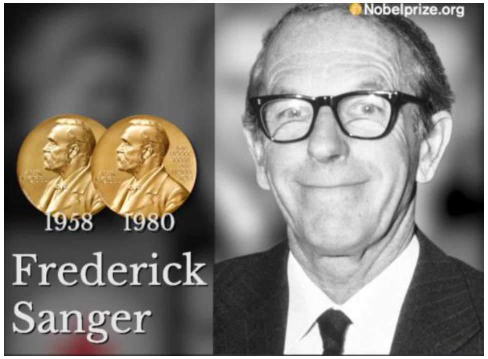
高通量测序

(Shendure & Ji, 2008, Nature Biotechnology; Metzker, 2010, Nature Reviews Genetics; Rhoads & Au, 2015, Genomics Proteomics Bioinformatics)

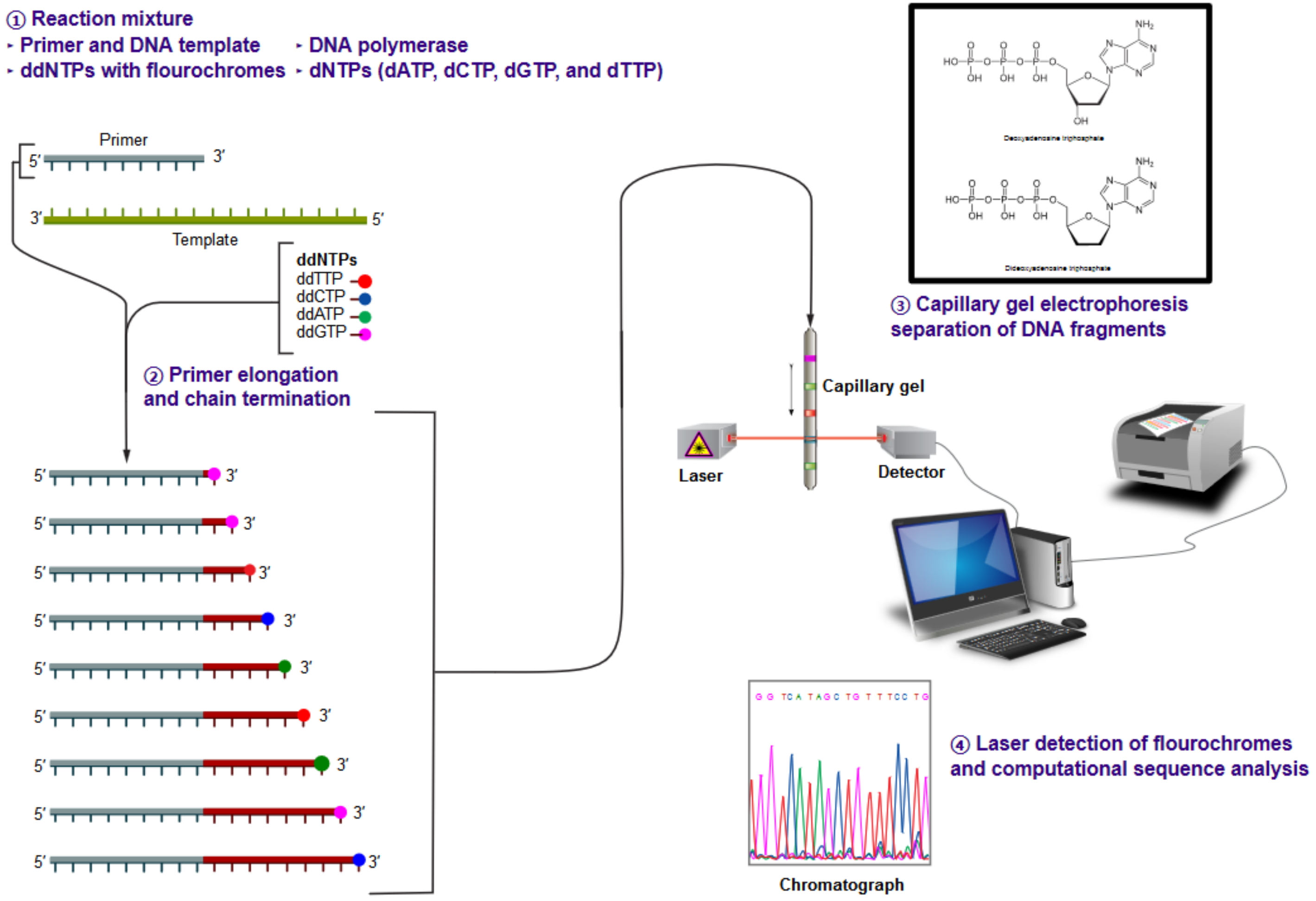
Next generation sequencer determines the bases of every DNA molecule.

Principles of Sanger Sequencing

（第一代测序）



双脱氧核苷酸链终止法



① Reaction mixture

- Primer and DNA template
- DNA polymerase
- ddNTPs with flourochromes
- dNTPs (dATP, dCTP, dGTP, and dTTP)

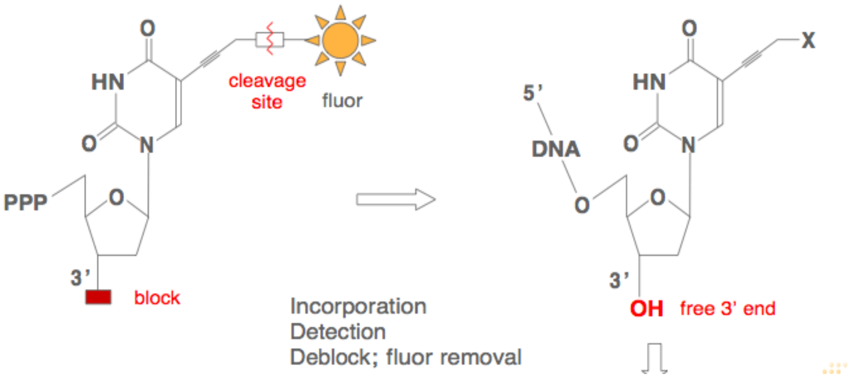
② Primer elongation and chain termination

③ Capillary gel electrophoresis separation of DNA fragments

④ Laser detection of flourochromes and computational sequence analysis

Principles of Illumina Sequencing (第二代测序)

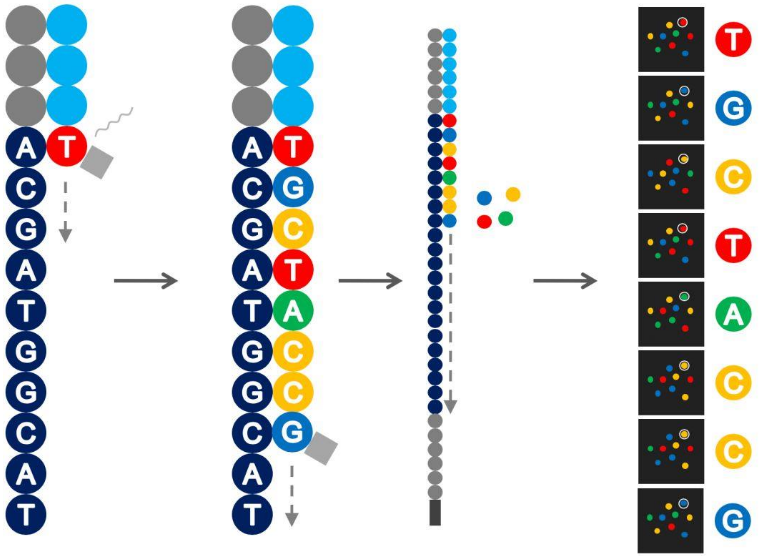
- All 4 labelled nucleotides in 1 reaction
- Higher accuracy



Incorporation
Detection
Deblock; fluor removal

3'-blocked reversible terminator (可逆屏蔽终结子)

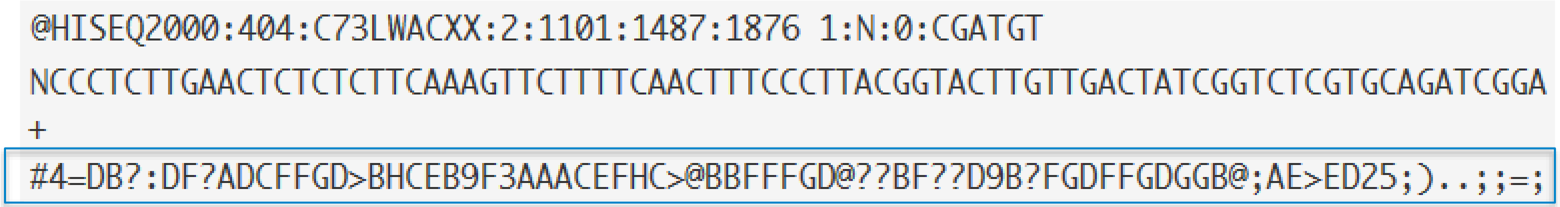
Sequencing by Synthesis
边合成边测序



通过给不同的dNTP加上不同的荧光基团，再与固定到测序芯片上的DNA片段进行合成反应，通过观测发出的荧光信号颜色，判断这一步合成的核苷酸类型。

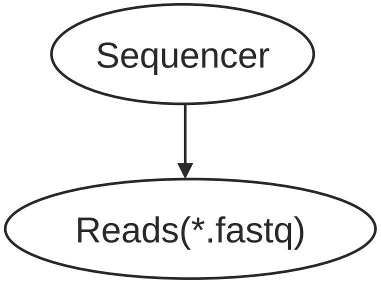
测序数据

- Read (读段)：A short DNA fragment which is read out by sequencer.
  - ○ DNA sequence
  - ○ Quality information(质量值以ASCII码表示，一般要Q>30)



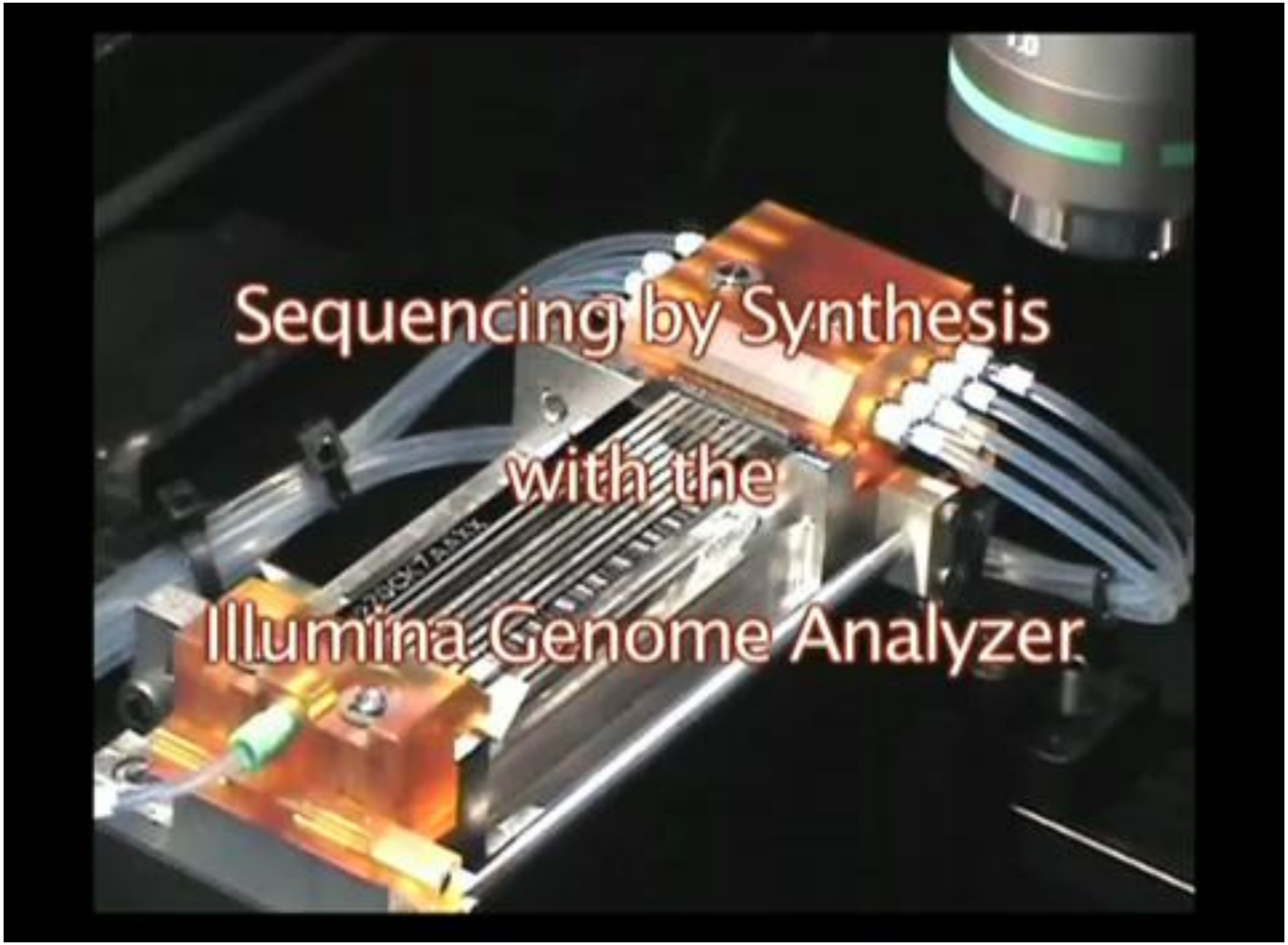
```
@HISEQ2000:404:C73LWACXX:2:1101:1487:1876 1:N:0:CGATGT
NCCCTCTTGAACTCTCTCTTCAAAGTTCTTTTCAACTTTCCCTTACGGTACTTGTTGACTATCGGTCTCGTGCAGATCGGA
+
#4=DB?:DF?ADCFFGD>BHCEB9F3AAACEFHC>@BBFFFGD@??BF??D9B?FGDFFGDGGB@;AE>ED25;)..;;=;
```

- 高通量测序的序列数据一般存储在FASTQ格式文件，文件后缀一般为“.fastq”, “.fq”等

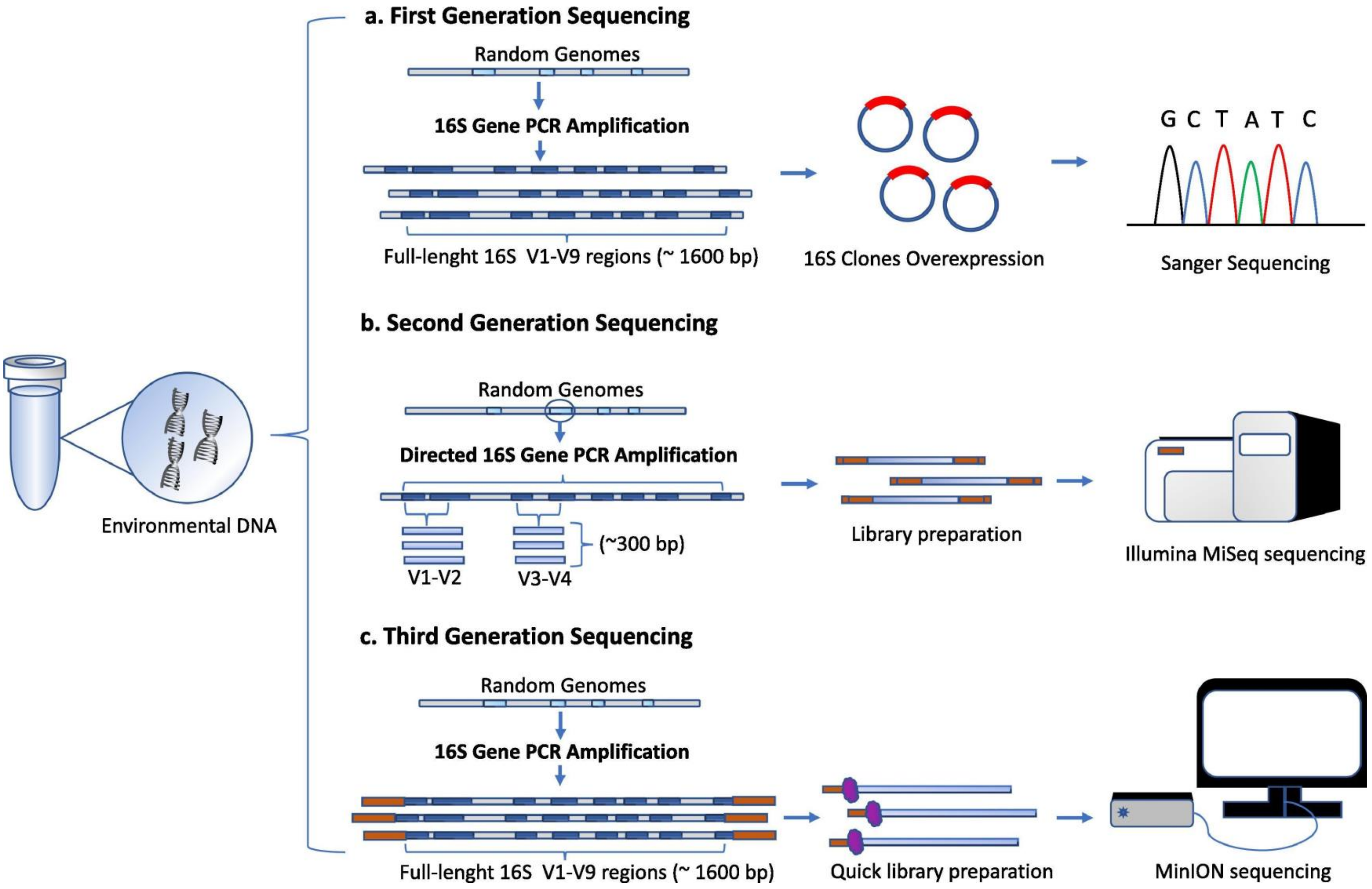

Sequencer

Reads(*.fastq)

第二代测序视频

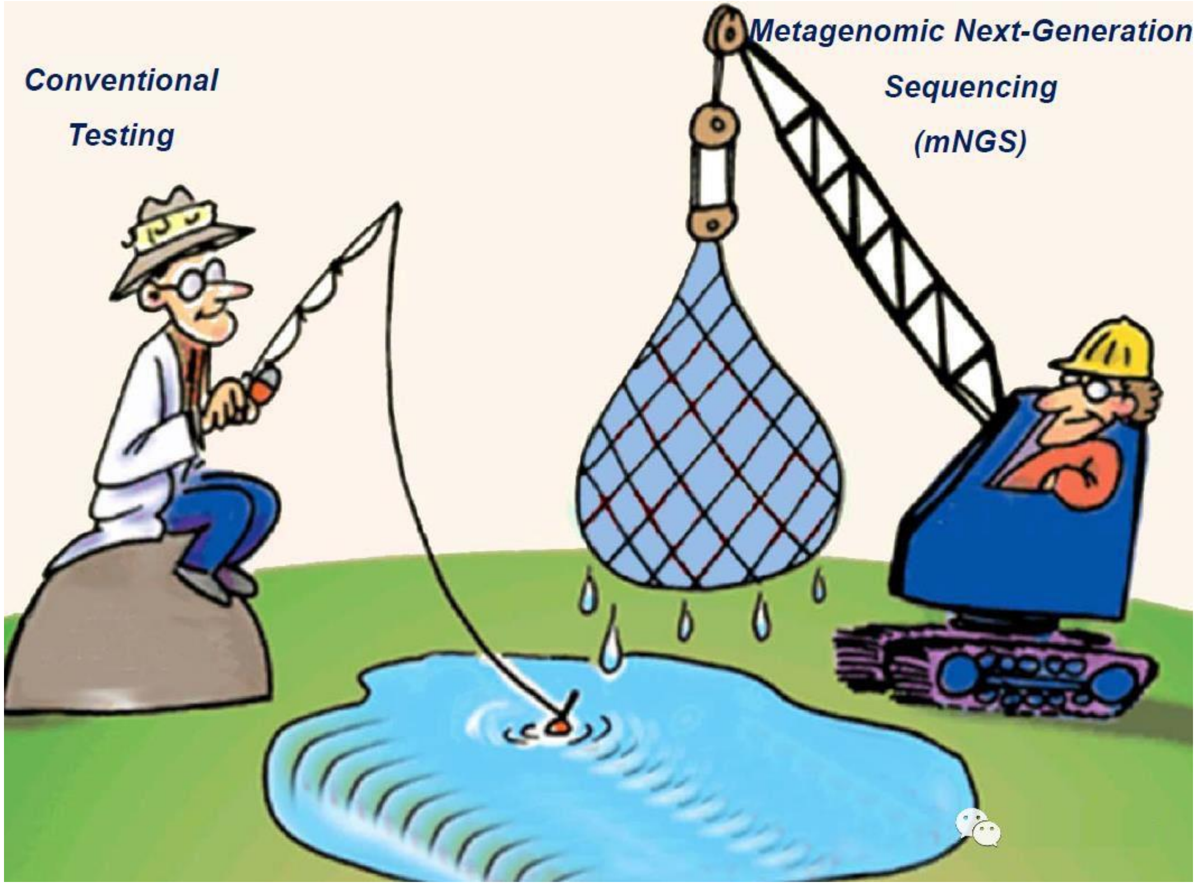

Sequencing by Synthesis
with the
Illumina Genome Analyzer

Three metagenomic strategies for each sequencing technology generation


a. First Generation Sequencing
Random Genomes
16S Gene PCR Amplification
Full-lenght 16S V1-V9 regions (~ 1600 bp)
16S Clones Overexpression
G C T A T C
Sanger Sequencing
b. Second Generation Sequencing
Environmental DNA
Random Genomes
Directed 16S Gene PCR Amplification
(~300 bp)
V1-V2
V3-V4
Library preparation
Illumina MiSeq sequencing
c. Third Generation Sequencing
Random Genomes
16S Gene PCR Amplification
Full-lenght 16S V1-V9 regions (~ 1600 bp)
Quick library preparation
MinION sequencing

Santos et al., Computational and Structural Biotechnology Journal 2020,18, 296–305.

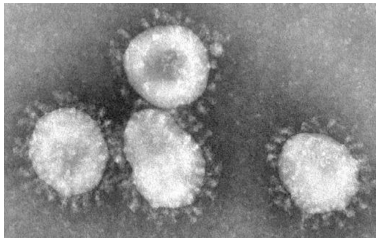
宏基因组下一代测序技术(mNGS)

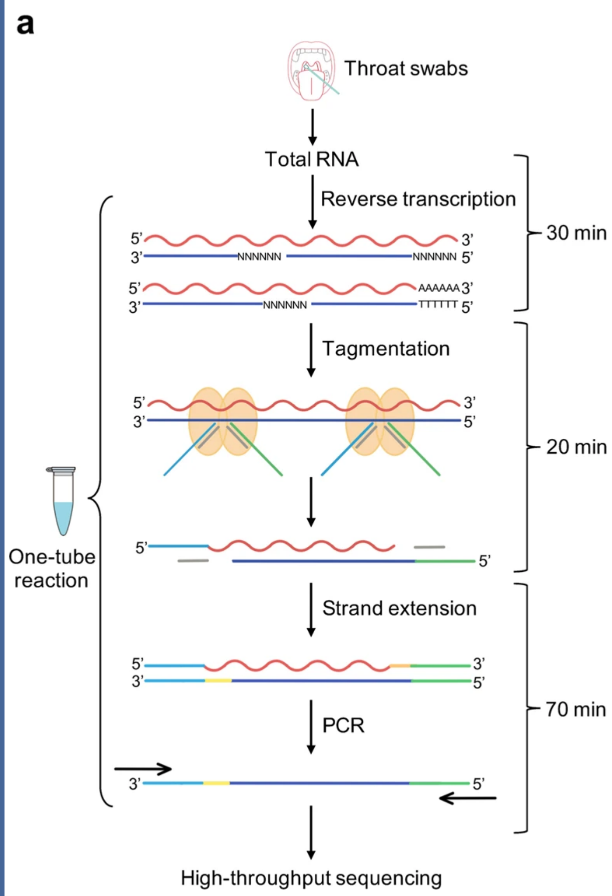


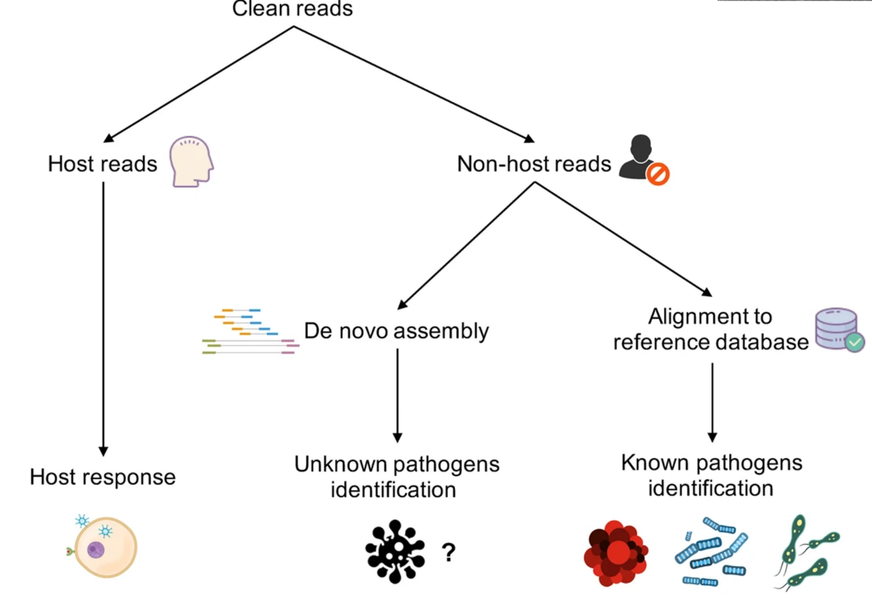
Metagenomic strategy for detecting SARS-CoV-2

传染病病原体鉴定时间：

2003年非典病毒(半年) → 2019年新冠病毒(一周)





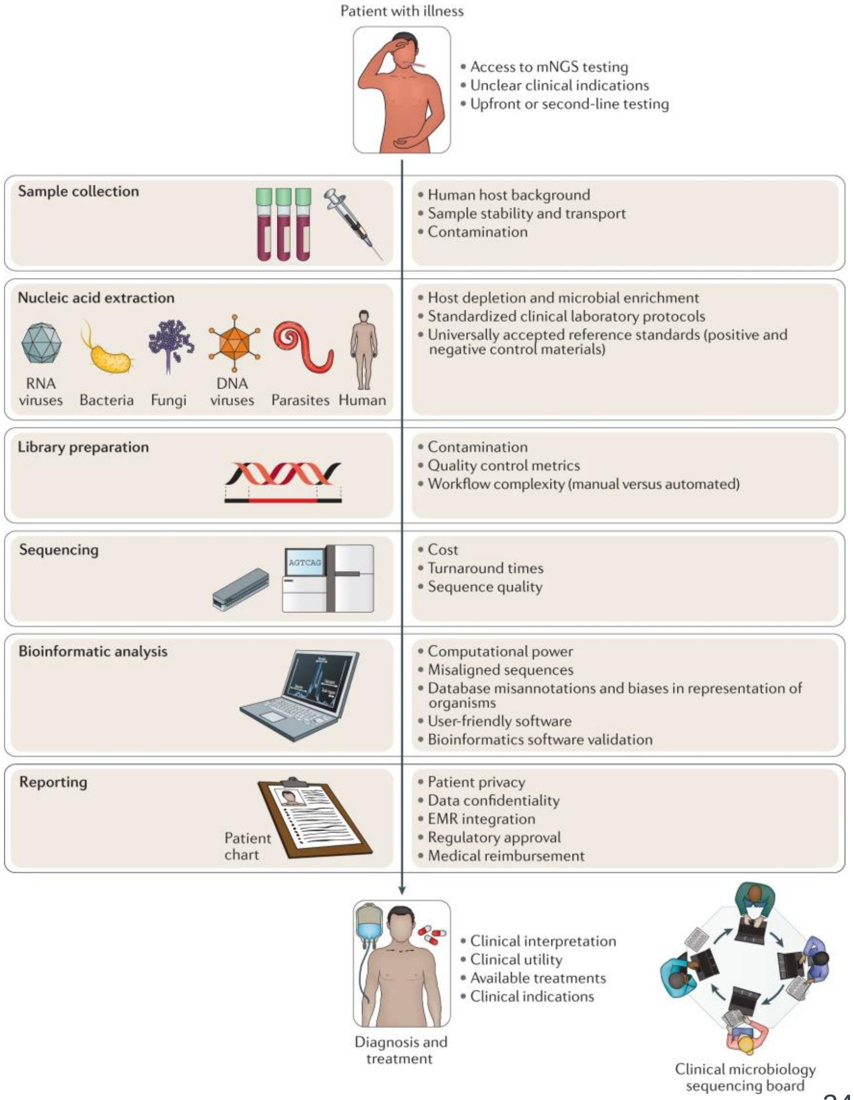


Santos et al., Computational and Structural Biotechnology Journal 2020,18, 296–305.

Clinical metagenomics

- Clinical metagenomic next-generation sequencing (mNGS), the comprehensive analysis of microbial and host genetic material (DNA and RNA) in samples from patients, is rapidly moving from research to clinical laboratories.

- The capacity to detect all potential apthogens – bacteria, viruses, fungi and parasites – in a sample and simultaneously interrogate host responses has great potential utility in the diagnosis of infectious disease.



Patient with illness

- Access to mNGS testing
- Unclear clinical indications
- Upfront or second-line testing

Sample collection

- Human host background
- Sample stability and transport
- Contamination

Nucleic acid extraction

- Host depletion and microbial enrichment
- Standardized clinical laboratory protocols
- Universally accepted reference standards (positive and negative control materials)

Library preparation

- Contamination
- Quality control metrics
- Workflow complexity (manual versus automated)

Sequencing

- Cost
- Turnaround times
- Sequence quality

Bioinformatic analysis

- Computational power
- Misaligned sequences
- Database misannotations and biases in representation of organisms
- User-friendly software
- Bioinformatics software validation

Reporting

- Patient privacy
- Data confidentiality
- EMR integration
- Regulatory approval
- Medical reimbursement

Diagnosis and treatment

- Clinical interpretation
- Clinical utility
- Available treatments
- Clinical indications

Clinical microbiology sequencing board

Clinical microgenomics, nature reviews genetics 2019

How does metagenomic sequencing work (宏基因组测序流程)?

What is a typical experimental & computational workflow?

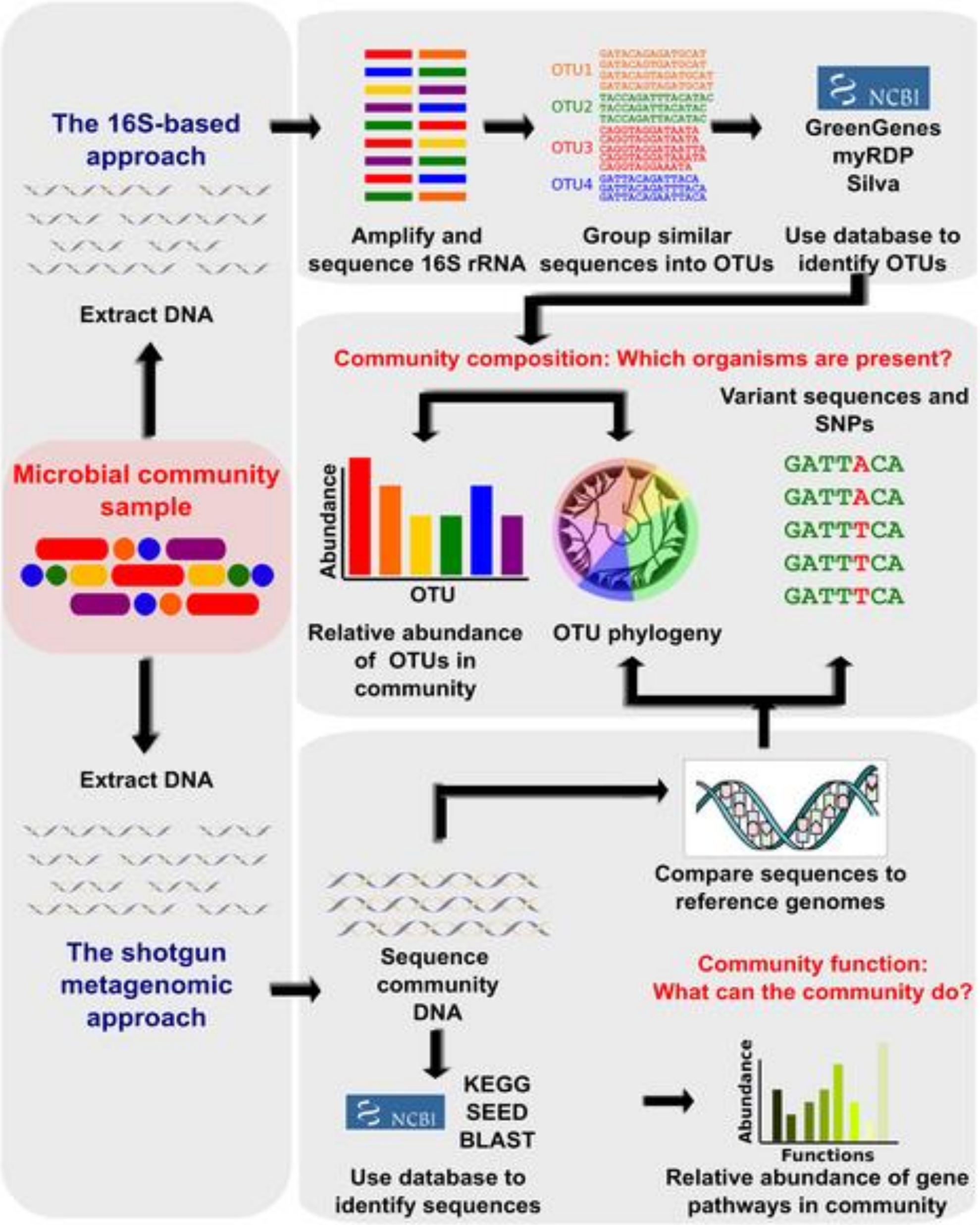
宏基因组学两种测序策略

扩增子测序

- 对不同生物中保守的标记基因（16S/18S/ITS），进行PCR扩增，再测序分析

宏基因组测序

- 直接提取样品的全部基因组DNA，进行测序分析


The 16S-based approach
Amplify and sequence 16S rRNA
OTU1
OTU2
OTU3
OTU4
Group similar sequences into OTUs
NCBI
GreenGenes
myRDP
Silva
Use database to identify OTUs
Extract DNA
Microbial community sample
Community composition: Which organisms are present?
Variant sequences and SNPs
GATTACA
GATTACA
GATTTCA
GATTTCA
GATTTCA
Abundance
OTU
Relative abundance of OTUs in community
OTU phylogeny
Extract DNA
The shotgun metagenomic approach
Compare sequences to reference genomes
Sequence community DNA
Community function: What can the community do?
NCBI
KEGG
SEED
BLAST
Use database to identify sequences
Abundance
Functions
Relative abundance of gene pathways in community

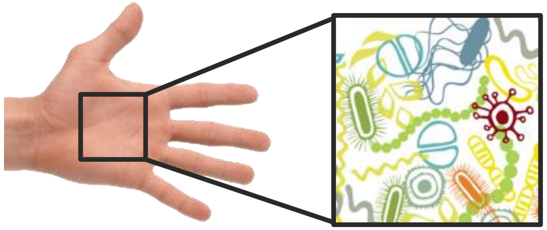
Morgan XC, Huttenhower C (2012) Chapter 12: Human Microbiome Analysis. PLoS Comput Biol 8(12): e1002808.

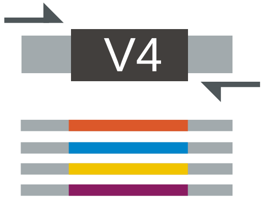
A workflow for 16S amplicon sequencing

1 Collect sample (*e.g.* skin swab)

2 Total DNA extraction

3 Amplify variable region of 16S rRNA gene

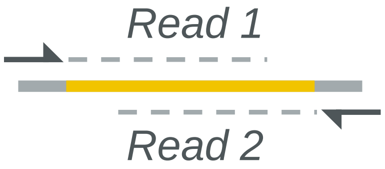


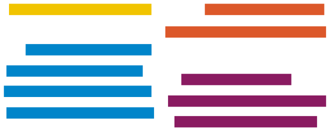


4 Sequence 16S rRNA genes

5 Processing, QC, and assembly of reads

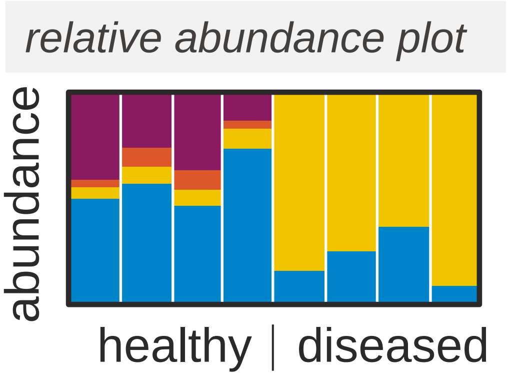
6 Generate a representative sequence for each species and assign taxonomy

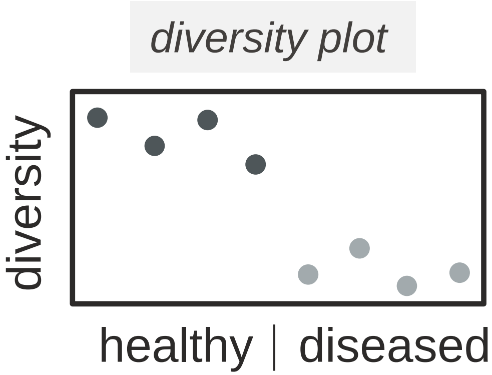




7

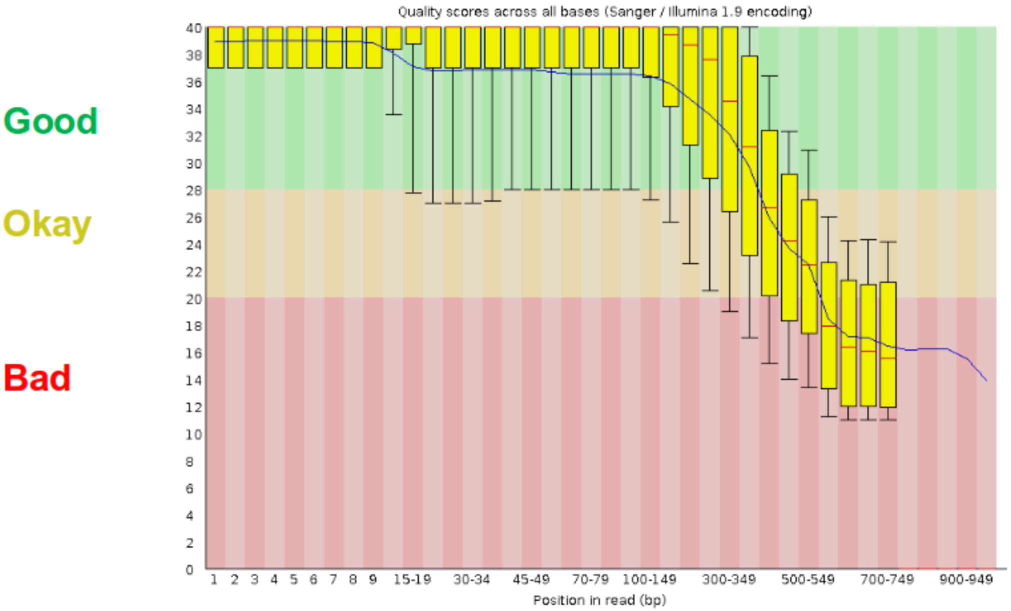
Who? How much?





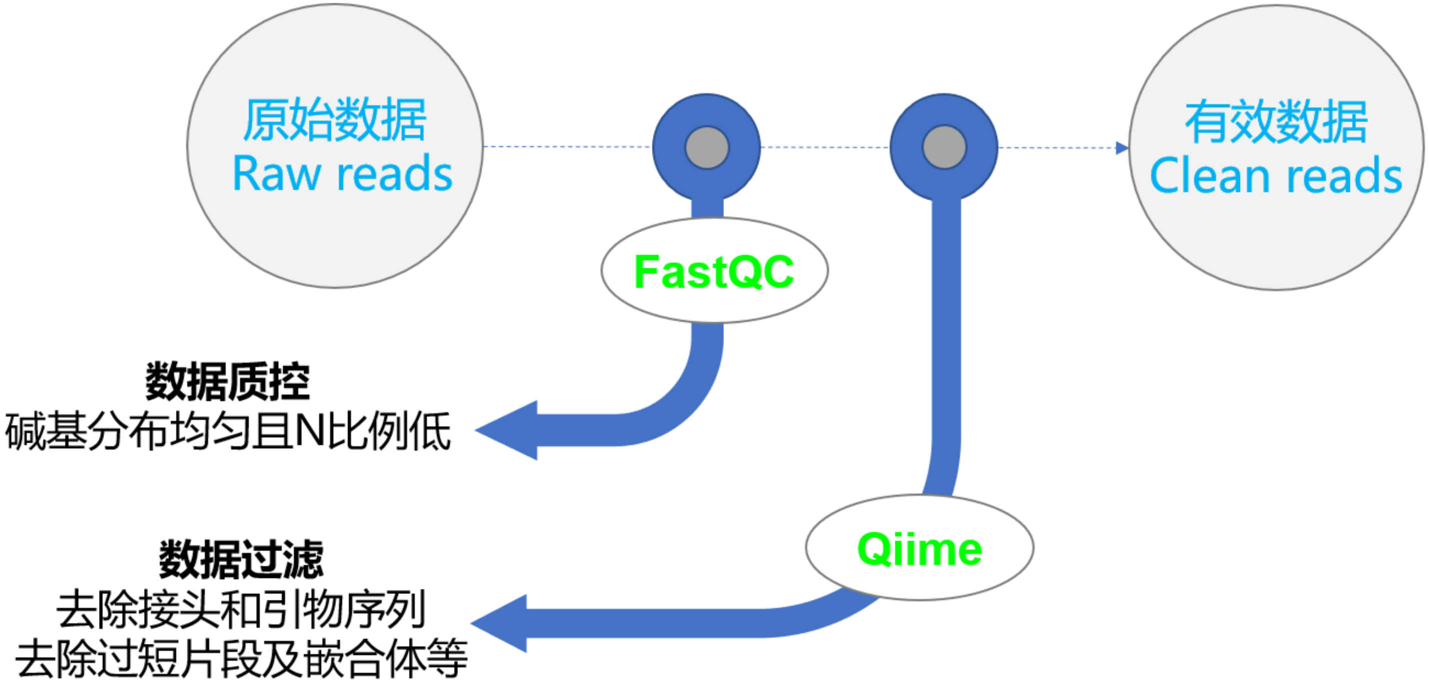
Sequence processing (测序数据处理)

5 Processing and QC of reads (测序数据质控)



Reads quality analyzed by FastQC

Sequence processing(测序数据处理)



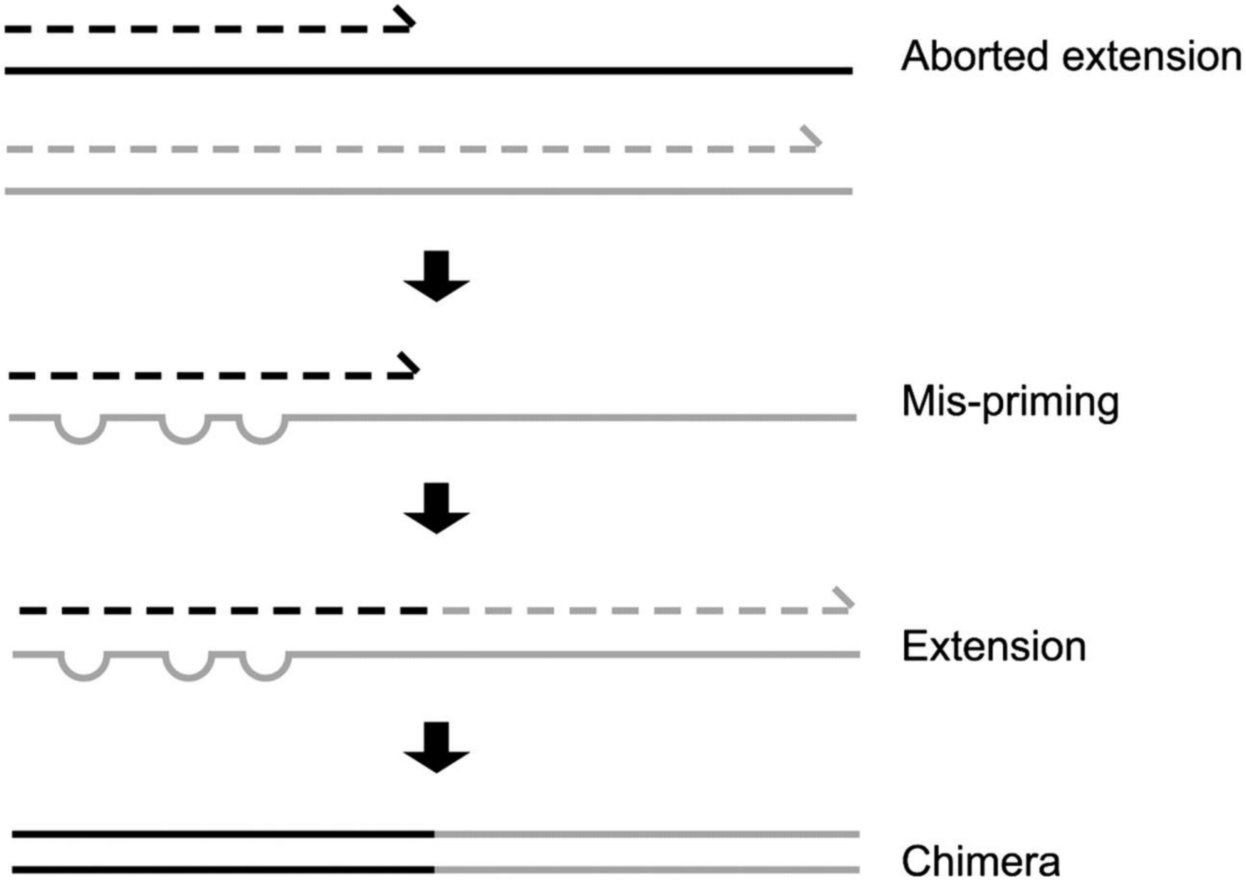
数据质控
碱基分布均匀且N比例低

数据过滤
去除接头和引物序列
去除过短片段及嵌合体等

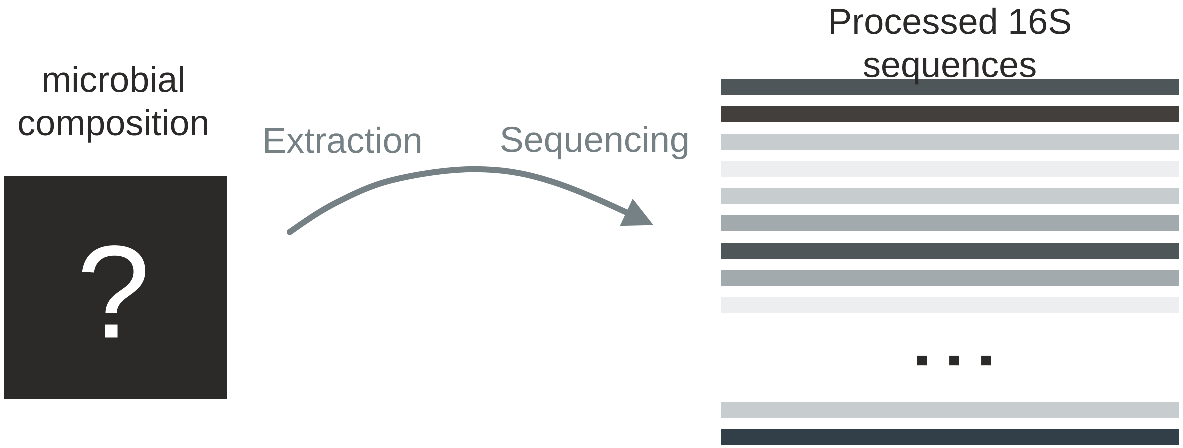
There are a lot of ways to filter and trim your data：
(i)low quality bases (Q< 20)
(ii)Remove chimeras (嵌合体)

Chimera Removal(去除嵌合体):

During PCR multiple sequences can combine to form a hybrid.



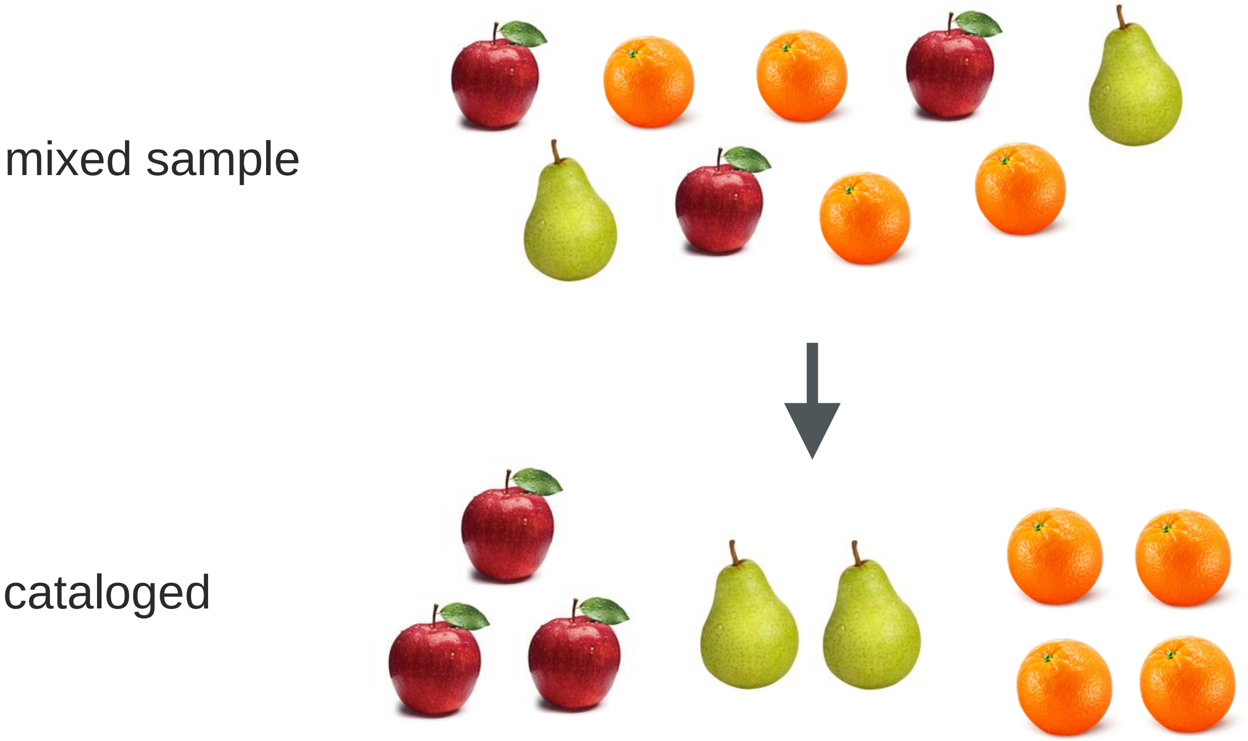
Where do we want to go next?



The challenge of metagenomics is that the sample is mixed!

→Which 16S sequence came from which bacterium?

It is trivial to catalog identical objects

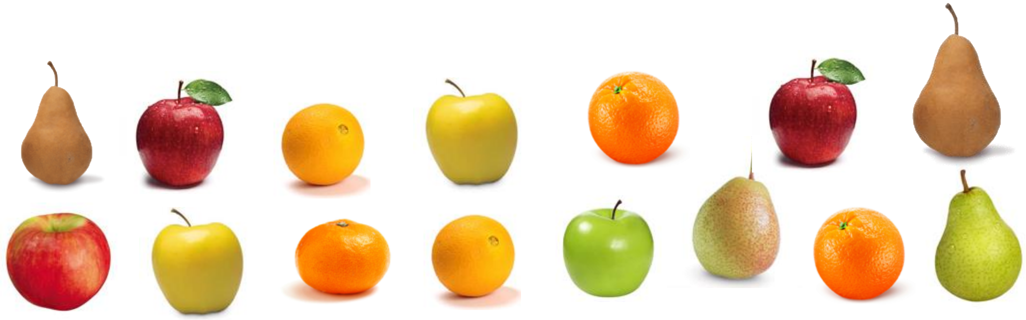


mixed sample

cataloged

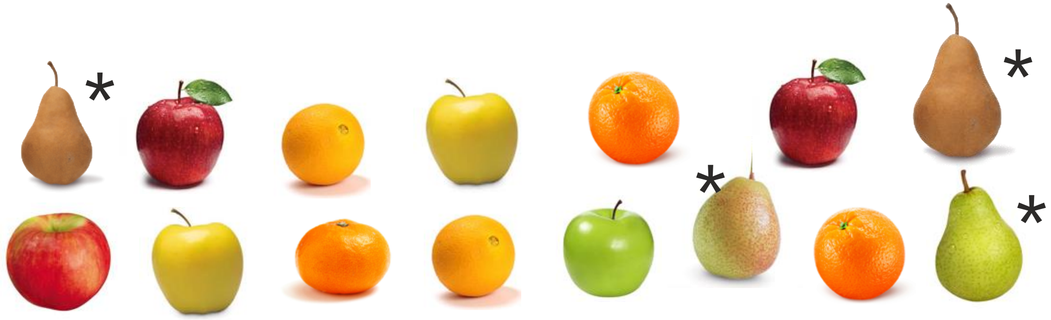
Cataloging variable objects is hard

mixed
sample



Cataloging variable objects is hard

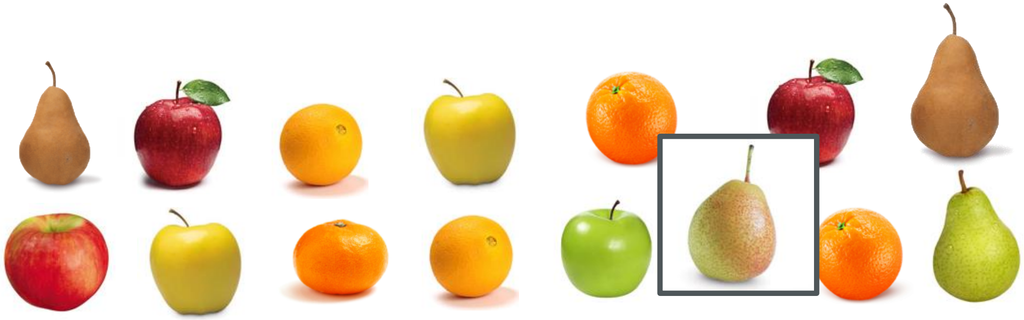
mixed
sample



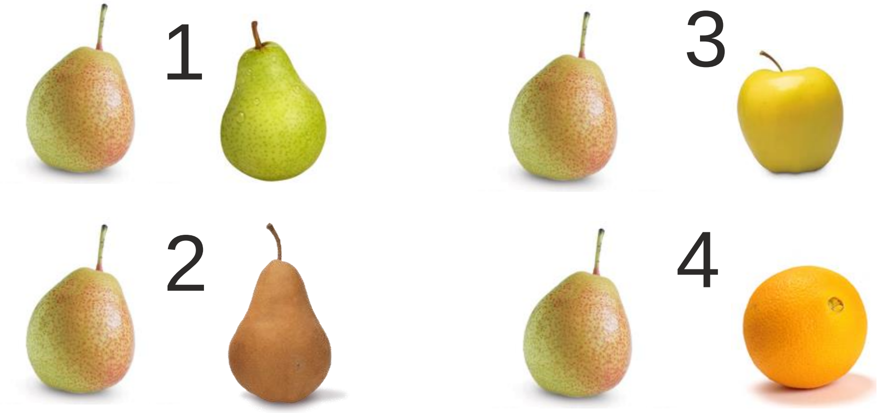
假设我们都不认识这些水果，如何把标星的梨挑选出来?

We catalog variable objects by iterative pairwise comparison(两两比较)

mixed sample

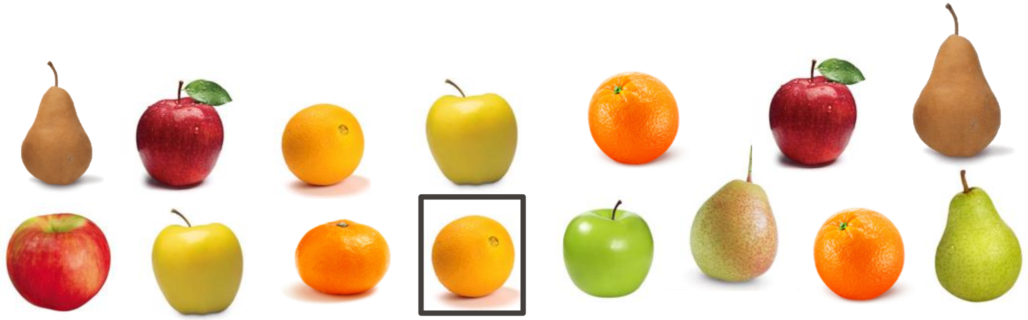


ranked pairwise comparisons

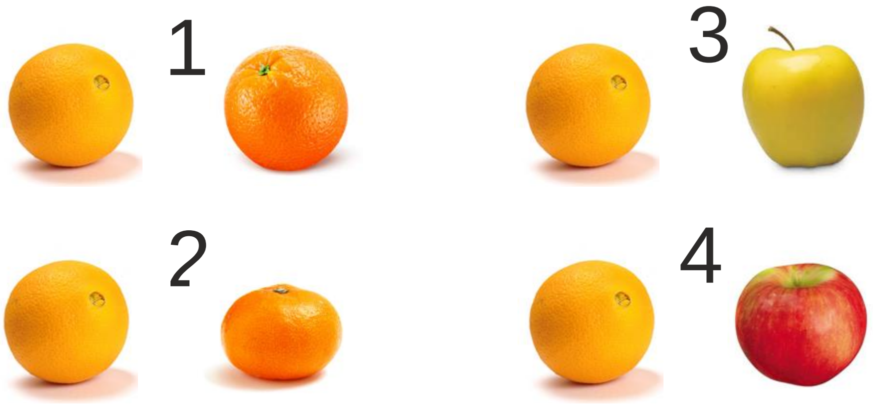


We catalog variable objects by iterative pairwise comparison

mixed sample

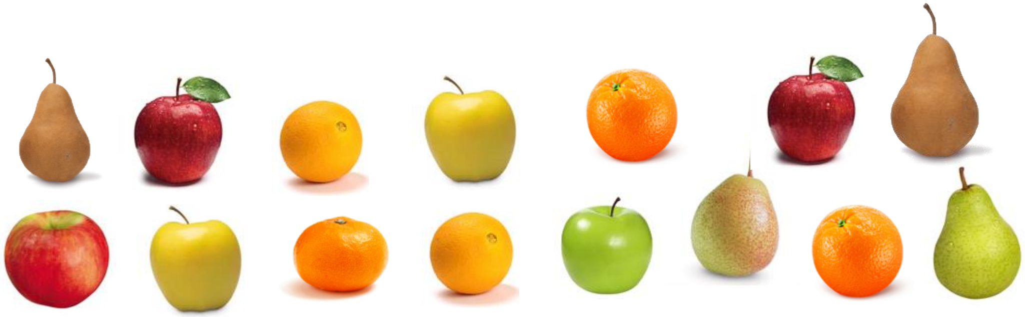


ranked pairwise comparisons

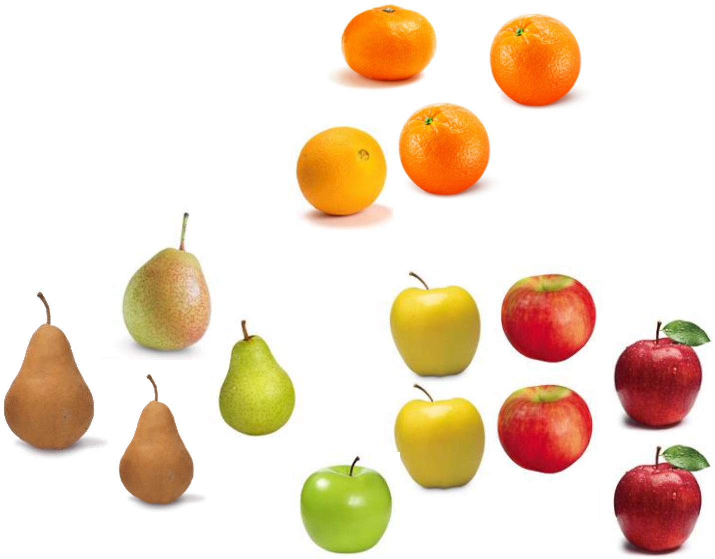


Clusters arise of similar items

mixed
sample

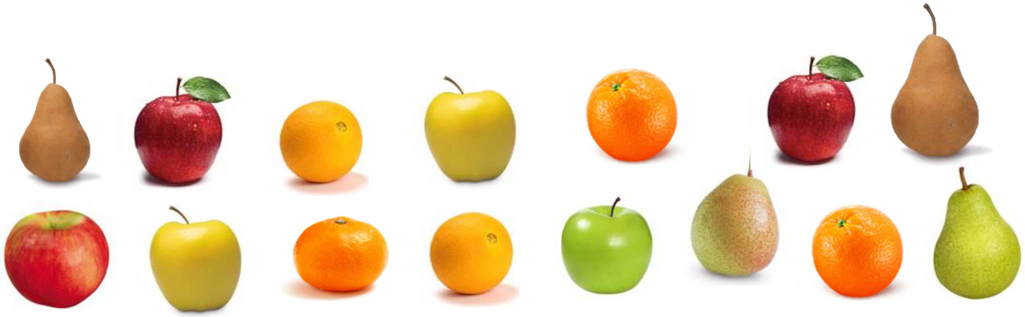


ranked
pairwise
comparisons

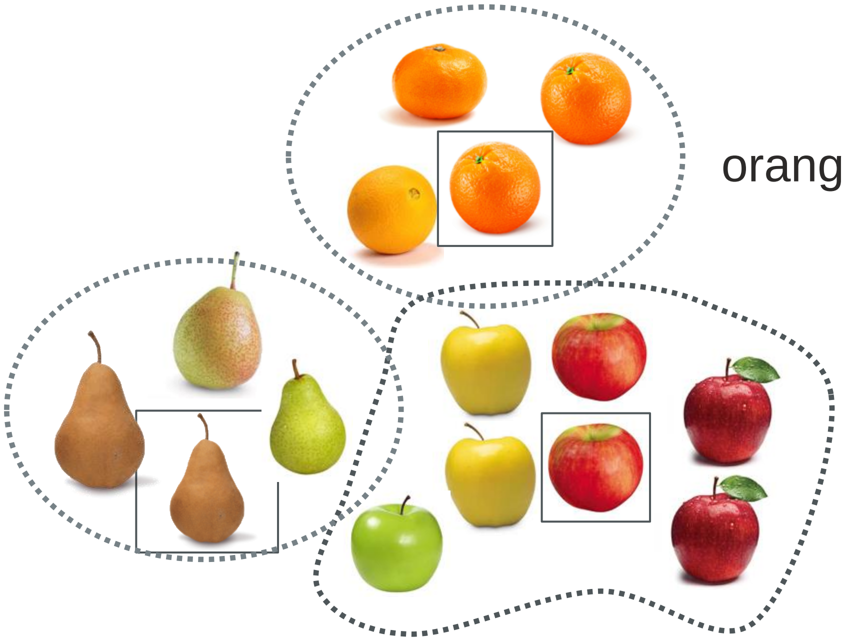


Similar fruits cluster together

mixed sample



pairwise comparison distances



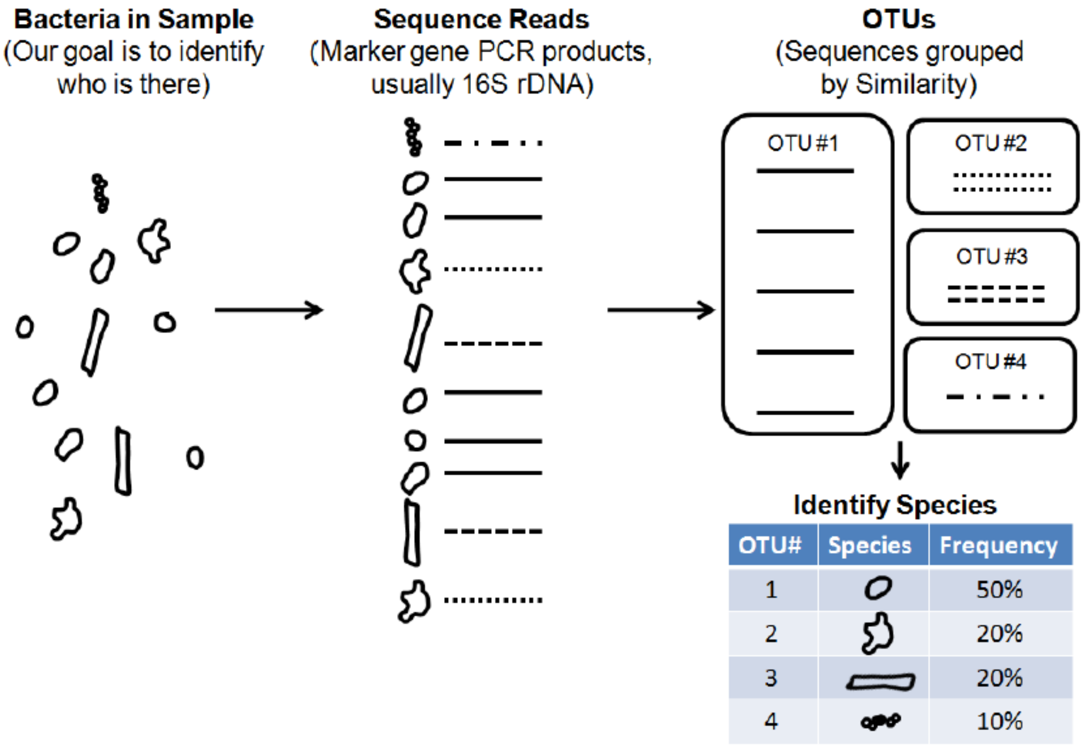
orange

pear

apple

16S rRNA 基因序列的分类鉴定

1. Identify unique sequences
2. Use pairwise comparison to cluster into operational taxonomic units (OTUs)
3. Count how many sequences match each OTU


Bacteria in Sample
(Our goal is to identify who is there)
Sequence Reads
(Marker gene PCR products, usually 16S rDNA)
OTUs
(Sequences grouped by Similarity)
OTU #1
OTU #2
OTU #3
OTU #4
Identify Species

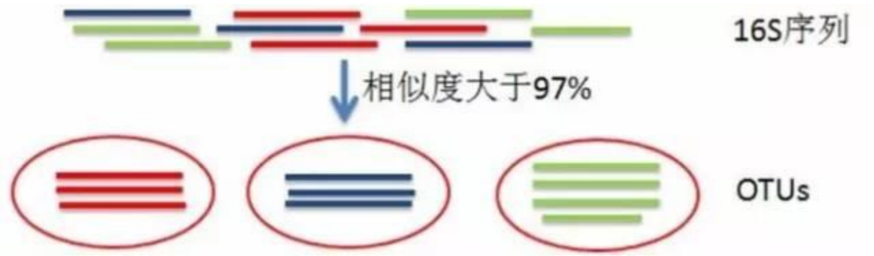
OTU#	Species	Frequency
1		50%
2		20%
3		20%
4		10%

- OTU:可操作分类单元
  - 为便于进行分类分析，人为给某一个分类单元（品系、种、属等）设置的同一标志（Marker）
- OTU also refers to clusters of organisms, grouped by DNA sequence similarity of a specific taxonomic marker gene.
  - 原核生物16S rDNA
  - 真核生物18S rDNA/ITS

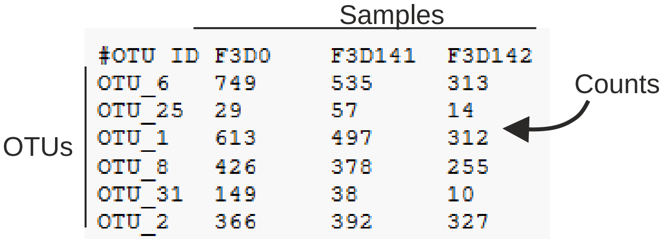
Sequences to OTUs to OTU abundance(丰度)

16S序列相似性达到97%以上的菌株为同一个种(Species)

OTU sequences are representative sequences chosen for each OTUs and are <97% similar compared to any other OTU sequences
~ 97% similarity = species



To generate a relative abundance table, count the number of 16S sequences matching each OTU sequence


Samples
OTUs
Counts

#OTU ID	F3D0	F3D141	F3D142
OTU_6	749	535	313
OTU_25	29	57	14
OTU_1	613	497	312
OTU_8	426	378	255
OTU_31	149	38	10
OTU_2	366	392	327

OTU丰度表：菌群组成

OTU taxonomy: 微生物分类注释

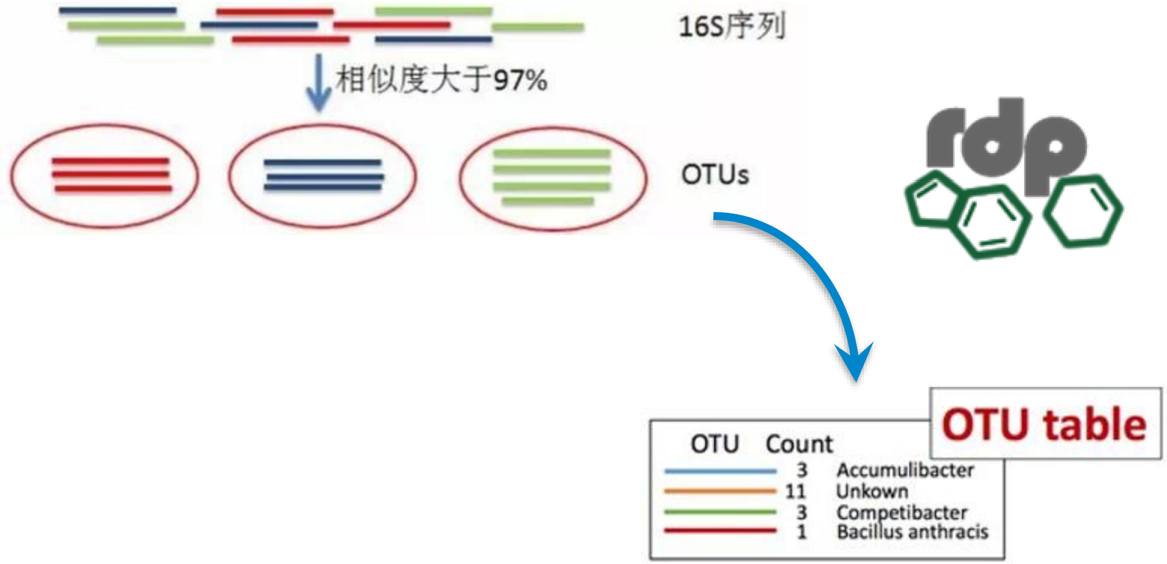
- OTU聚类后，挑选出每个OTU中的代表序列，与RDP、SILVA或GreenGenes等数据库进行比对，进行物种注释。

- Berkeley lab
- August 2013
- 202,421 entries

green genes
16S rRNA gene database and workbench compatible with ARB
greengenes.lbl.gov

- Max Planck Institute
- July 2015
- 172,418 entries

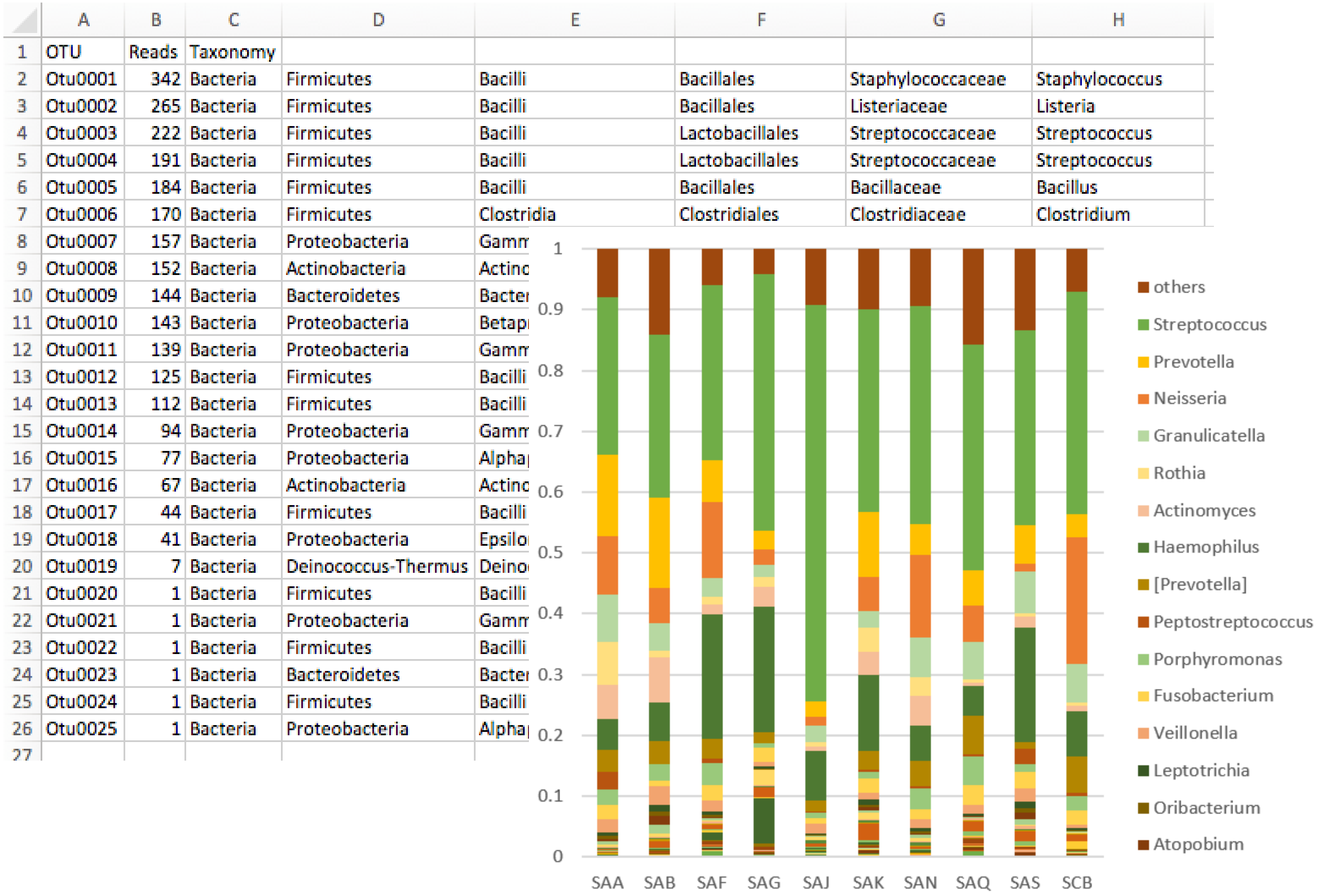
silva
high quality ribosomal RNA databases



OTU	Count	
🟦	3	Accumulibacter
🟧	11	Unkown
🟩	3	Competibacter
🟥	1	Bacillus anthracis

rDNA数据库(Ribosomal Database Project): http://rdp.cme.msu.edu/

物种OUT聚类表: OTU table



	A	B	C	D	E	F	G	H
1	OTU	Reads	Taxonomy					
2	Otu0001	342	Bacteria	Firmicutes	Bacilli	Bacillales	Staphylococcaceae	Staphylococcus
3	Otu0002	265	Bacteria	Firmicutes	Bacilli	Bacillales	Listeriaceae	Listeria
4	Otu0003	222	Bacteria	Firmicutes	Bacilli	Lactobacillales	Streptococcaceae	Streptococcus
5	Otu0004	191	Bacteria	Firmicutes	Bacilli	Lactobacillales	Streptococcaceae	Streptococcus
6	Otu0005	184	Bacteria	Firmicutes	Bacilli	Bacillales	Bacillaceae	Bacillus
7	Otu0006	170	Bacteria	Firmicutes	Clostridia	Clostridiales	Clostridiaceae	Clostridium
8	Otu0007	157	Bacteria	Proteobacteria	Gamm			
9	Otu0008	152	Bacteria	Actinobacteria	Actino			
10	Otu0009	144	Bacteria	Bacteroidetes	Bacter			
11	Otu0010	143	Bacteria	Proteobacteria	Betapr			
12	Otu0011	139	Bacteria	Proteobacteria	Gamm			
13	Otu0012	125	Bacteria	Firmicutes	Bacilli			
14	Otu0013	112	Bacteria	Firmicutes	Bacilli			
15	Otu0014	94	Bacteria	Proteobacteria	Gamm			
16	Otu0015	77	Bacteria	Proteobacteria	Alpha			
17	Otu0016	67	Bacteria	Actinobacteria	Actino			
18	Otu0017	44	Bacteria	Firmicutes	Bacilli			
19	Otu0018	41	Bacteria	Proteobacteria	Epsilo			
20	Otu0019	7	Bacteria	Deinococcus-Thermus	Deino			
21	Otu0020	1	Bacteria	Firmicutes	Bacilli			
22	Otu0021	1	Bacteria	Proteobacteria	Gamm			
23	Otu0022	1	Bacteria	Firmicutes	Bacilli			
24	Otu0023	1	Bacteria	Bacteroidetes	Bacter			
25	Otu0024	1	Bacteria	Firmicutes	Bacilli			
26	Otu0025	1	Bacteria	Proteobacteria	Alpha			
27								

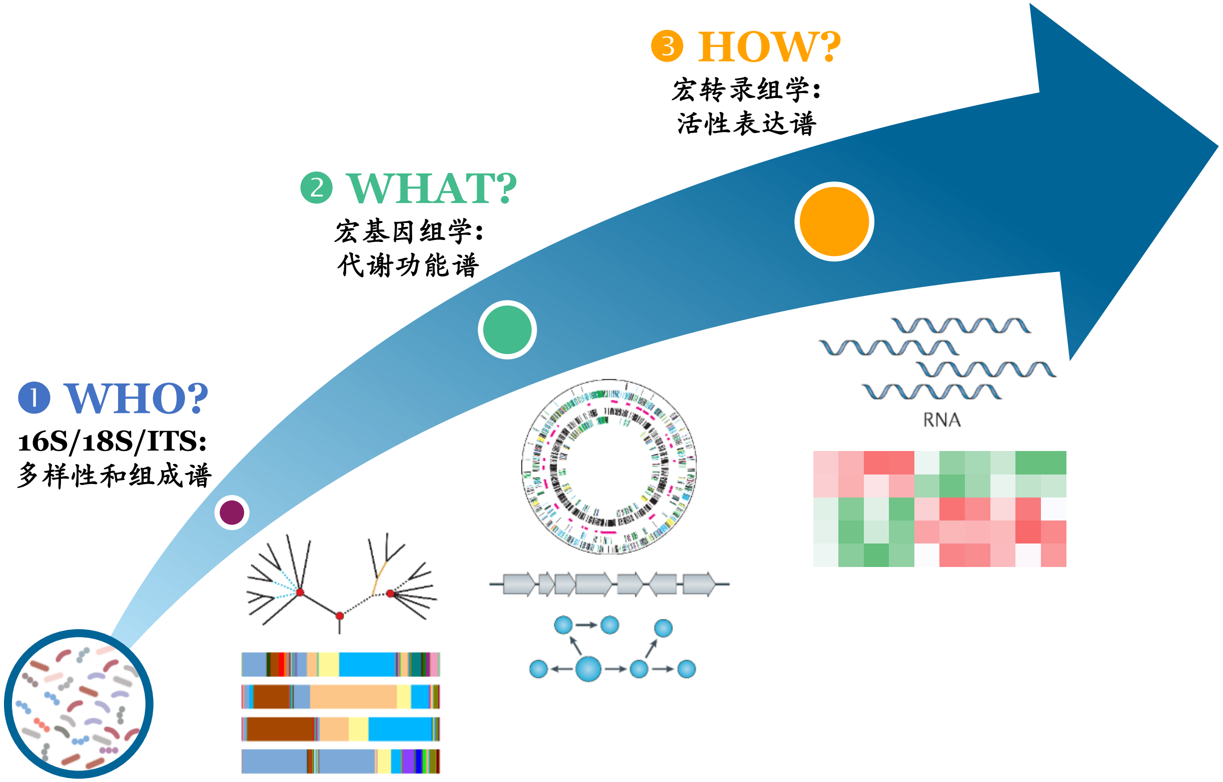
物种分类柱状堆积图

微生物组软件（USEARCH/QIIME）

- USEARCH是好用的扩增子分析软件，但是代码不开源，用于分析较大数据的64位版本收费。VSEARCH是USEARCH的免费、开源代替品。VSEARCH主要功能有: 嵌合体检测、聚类、去冗余、两两比对、排序、抽样、物种分类等。

- QIIME(Quantitative Insights Into Microbial Ecology)是一个用于比较和分析微生物基因组的开源软件，其开发者是美国科罗拉多大学的Rob Knight团队。QIIME能够处理各种测序平台上扩增子高通量测序结果。

基于高通量测序技术的微生物组学研究



❶ WHO?
16S/18S/ITS:
多样性和组成谱

❷ WHAT?
宏基因组学:
代谢功能谱

❸ HOW?
宏转录组学:
活性表达谱

微生物组仿真实验操作练习

扫描右边二维码查看操作说明👉

超星在线课程《微生物育种实验》