ExaBiome: Exascale Solutions to Microbiome Analysis

https://sites.google.com/lbl.gov/exabiome/

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LBNL, LANL, and JGI

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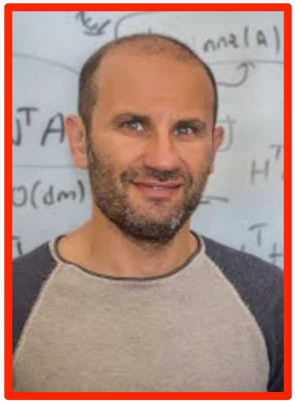
The Team from LBNL, LANL and JGI


Kathy Yelick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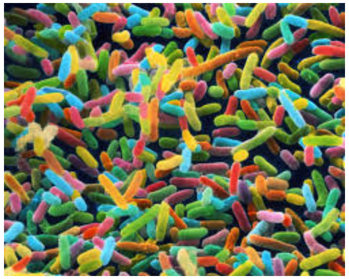
ExaBiome: Exascale Solutions for Microbiome Analysis

- **Microbes:** single cell organisms, such as bacteria and viruses
- **Microbiomes:** communities of 1000s of microbial species, less than 1% individually culturable in a lab (and thus sequenced)
- **Metagenomics:** genome sequencing on these communities (growing exponentially)

Environment



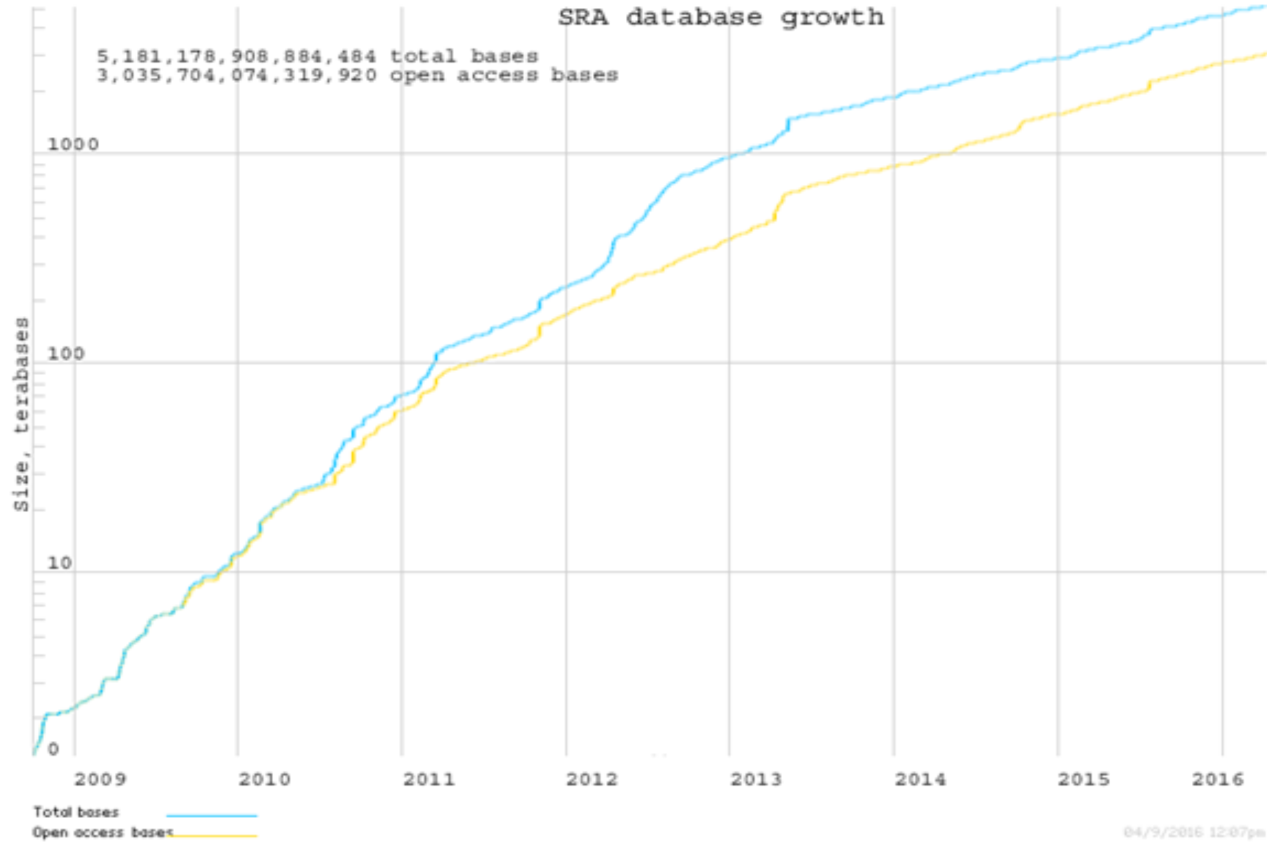
(Health)

Bio-Energy

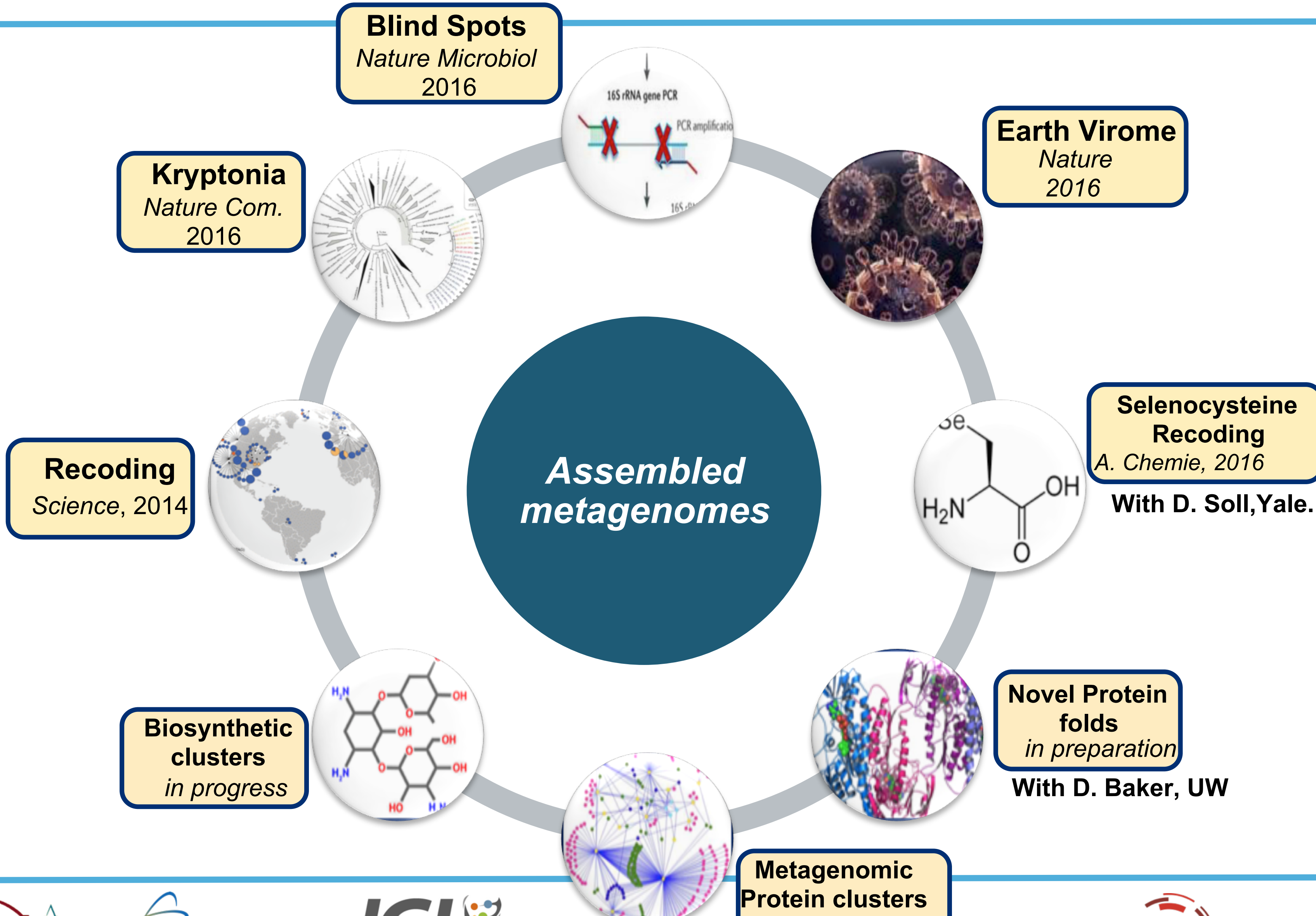
Bio-Manufacturing

Metagenome database growth

- **Metagenomic data double every 11 months**



Metagenomics data mining efforts at JGI



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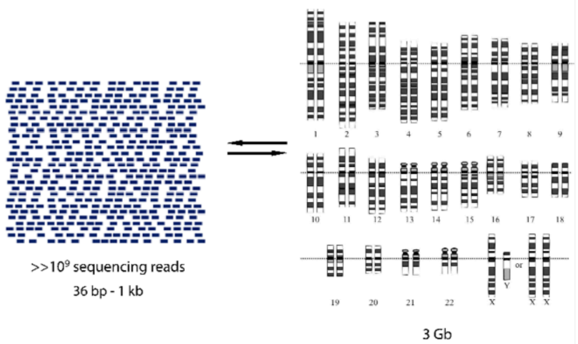
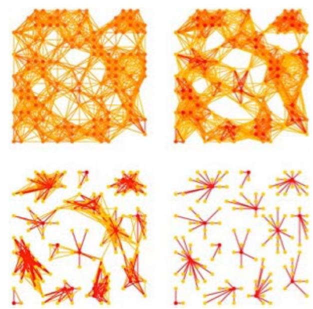
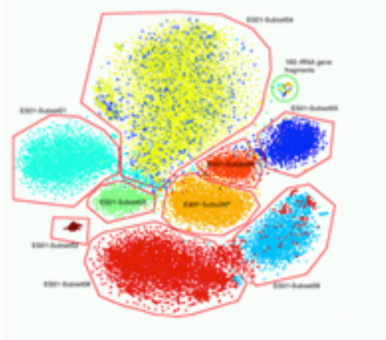






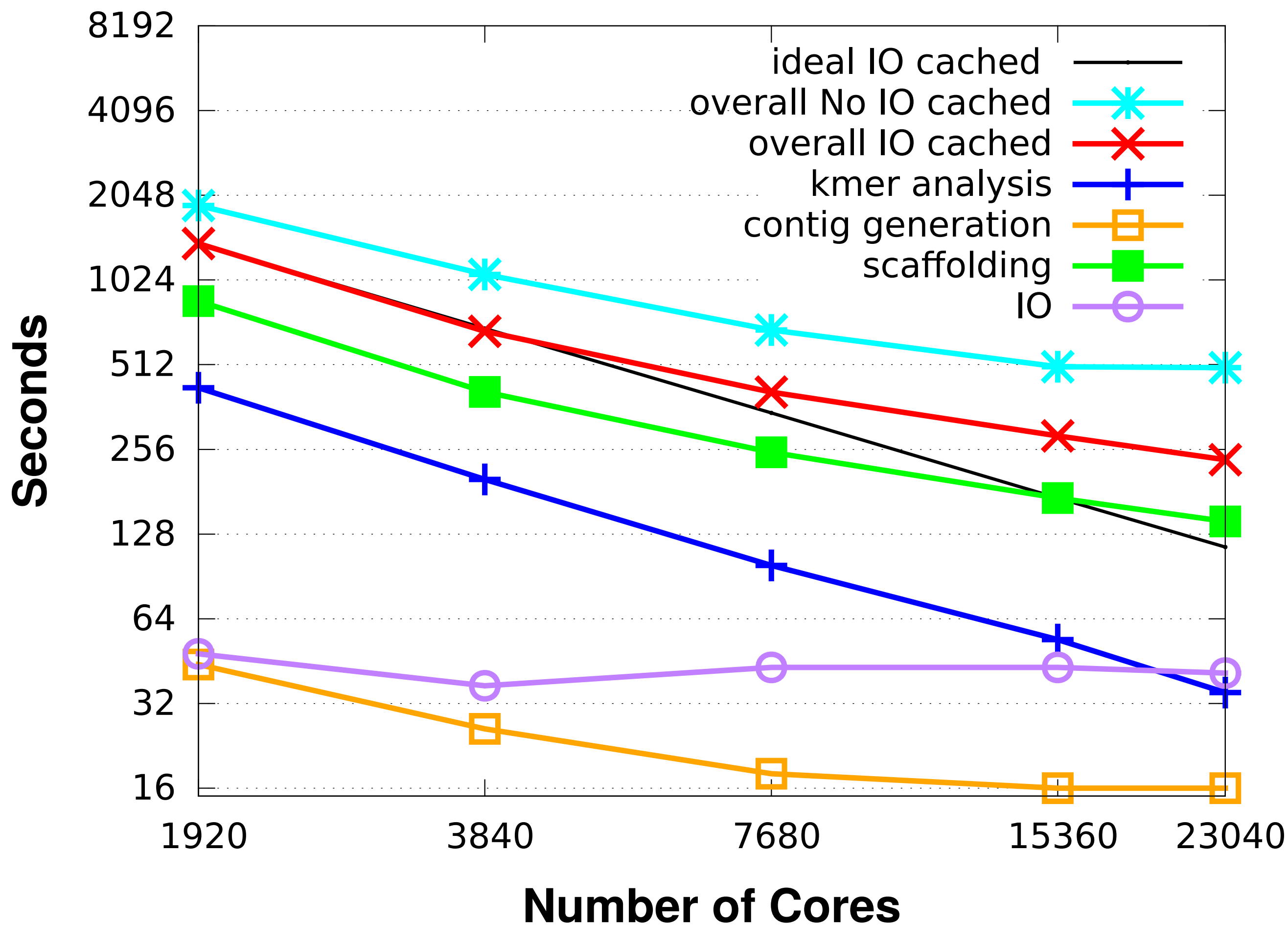
Metagenome Assembly	Protein Clustering	Comparative Analysis
Graph algorithms, Hash Tables, alignment (Smith-Waterman)	Machine learning (clustering), sparse linear algebra / graphs	Alignment, Machine learning (dimensionality reduction), linear algebra
Fine-grained comm., all-to-all, remote atomics and fast I/O	Fast barriers, subset reductions	All-to-all

PI: Katherine Yelick (LBNL)

Metagenome assembly is hard

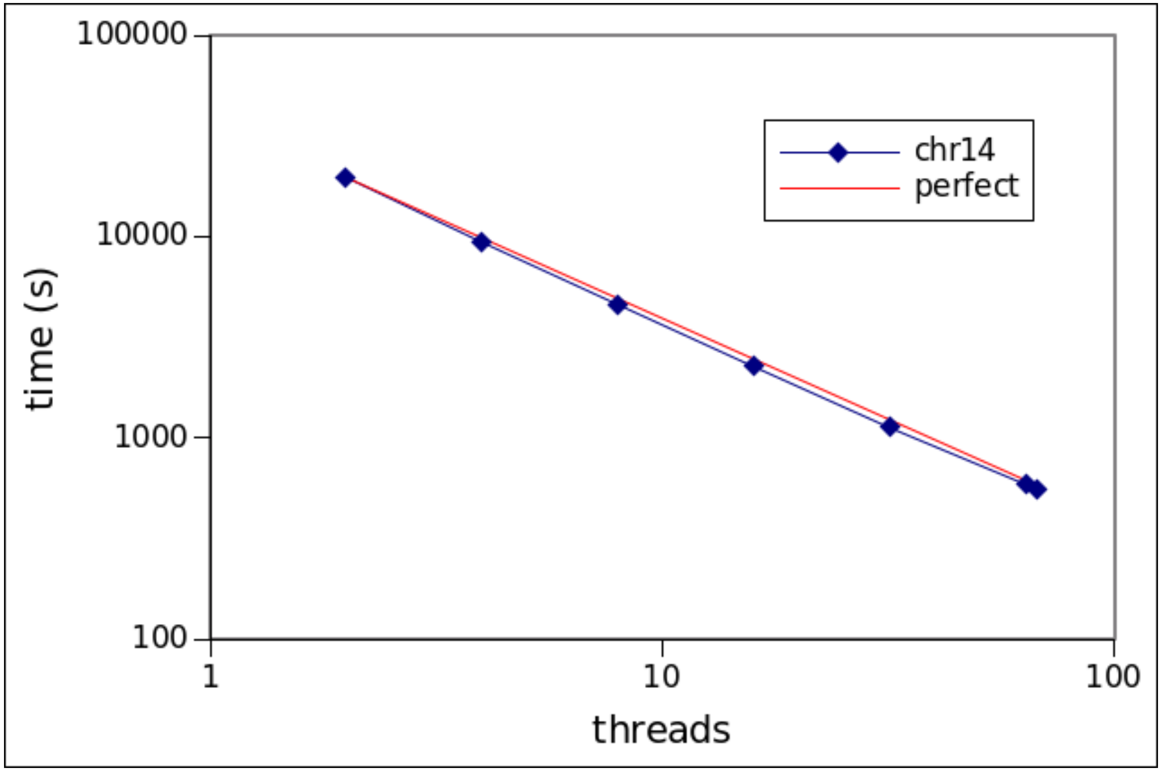
- **There is no genome reference.**
- **Reads are significantly shorter than whole genome.**
  - –Reads consist of 20 to 30K bases
  - –Genomes vary in length and complexity – up to 30G bases
- **Reads include errors.**
- **Genomes have repetitive regions.**
  - –Repetitive regions increase genome complexity
- **Microbial genomes occur with thousands of others**
  - –The coverage (frequency) may vary be orders of magnitude

Strong scaling (human genome) on Cray XC30



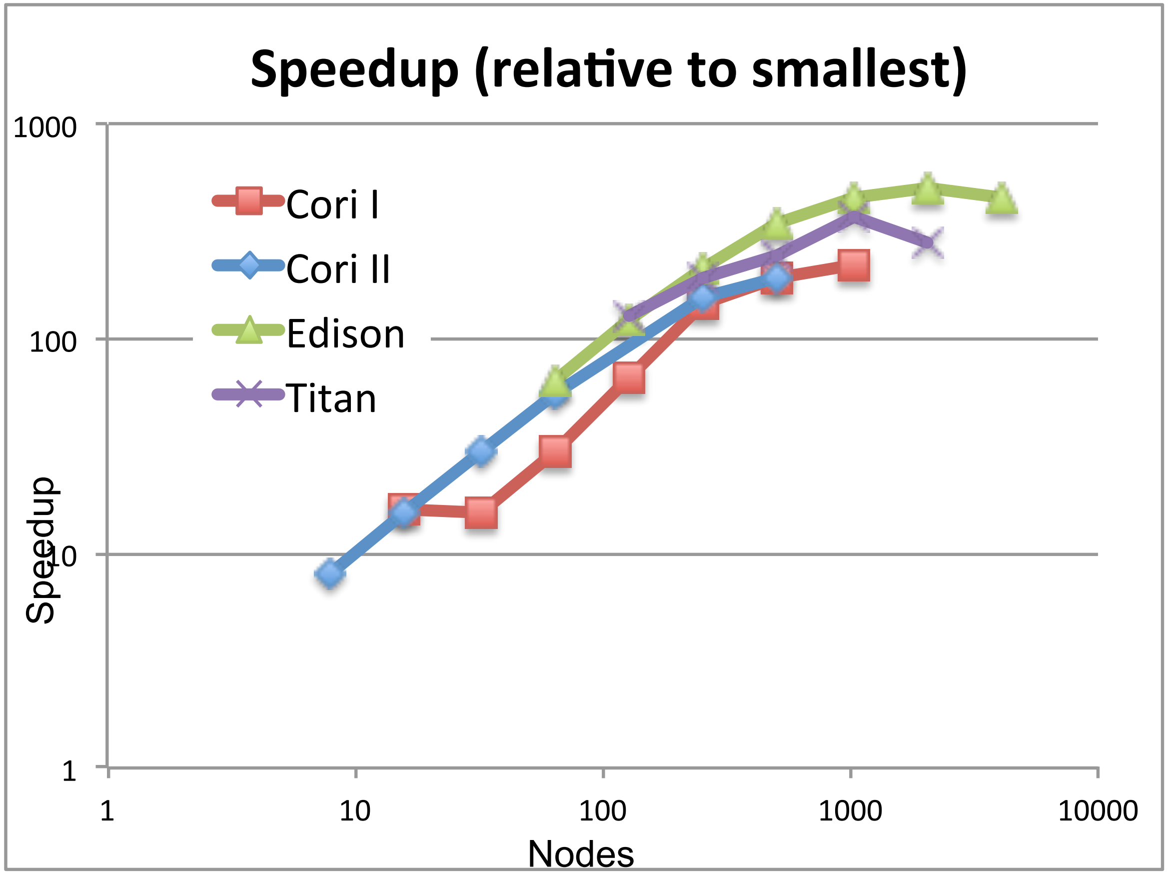
Single Node Scaling on KNL

Strong Scaling (small dataset, chromosome14)



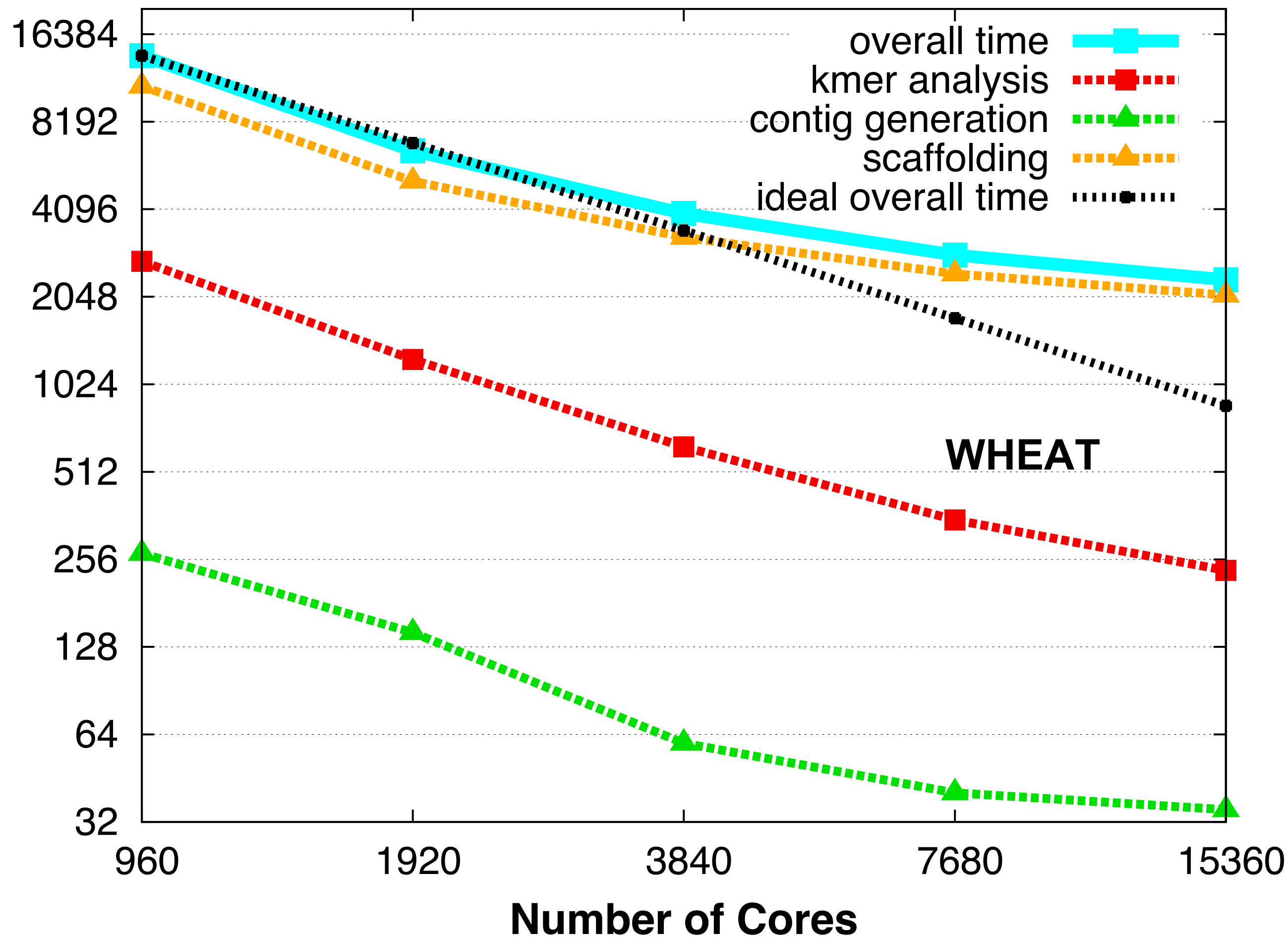
UPC (with MPI in K-mer analysis)

Speedup on Human Data (single genome)



- Complete assembly of human genome in **4 minutes using 23K cores.**
- **700x speedup over** original Meraculous code used in production (only ran on shared memory where it took **2,880 minutes**)

Strong scaling (wheat genome) on Cray XC30



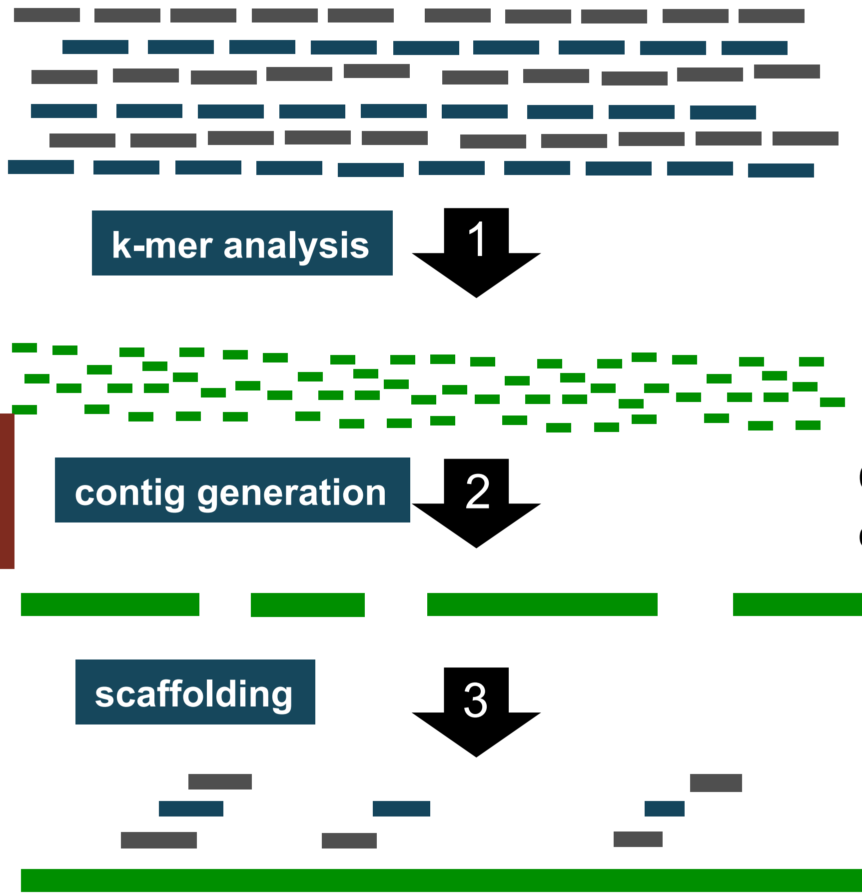
Early version of HipMer were used for the first whole genome assembly of wheat

Original HipMer Pipeline Summary (Single Genome)

reads

Input: Reads that may contain errors

Chop reads into k-mers, process k-mers to **exclude errors**



***k*-mers**

NERSC-9 “Meraculous” benchmark

Construct & traverse de Bruijn graph of k-mers, generate contigs

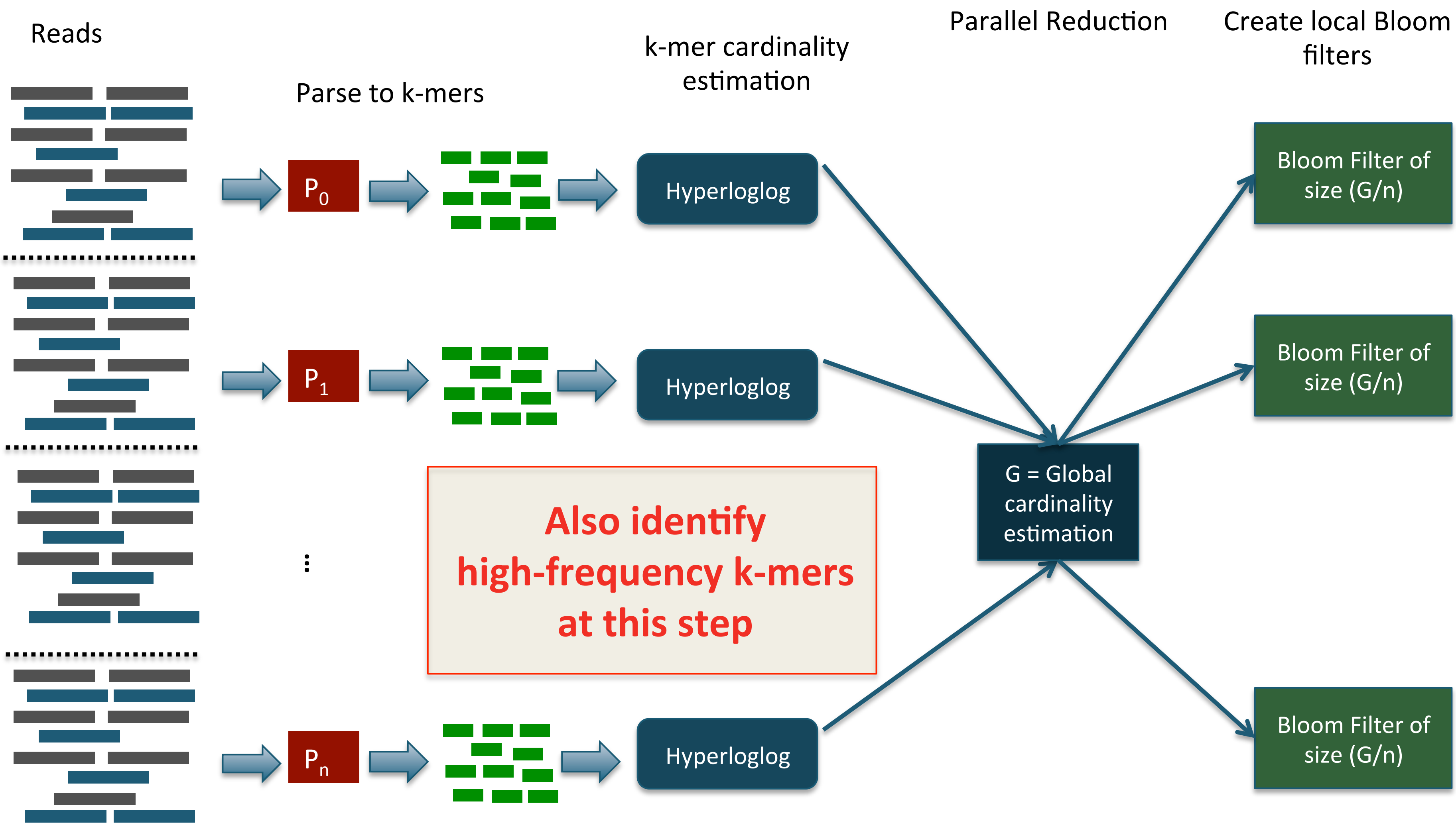
contigs

Leverage read information to link contigs and generate scaffolds.

scaffolds

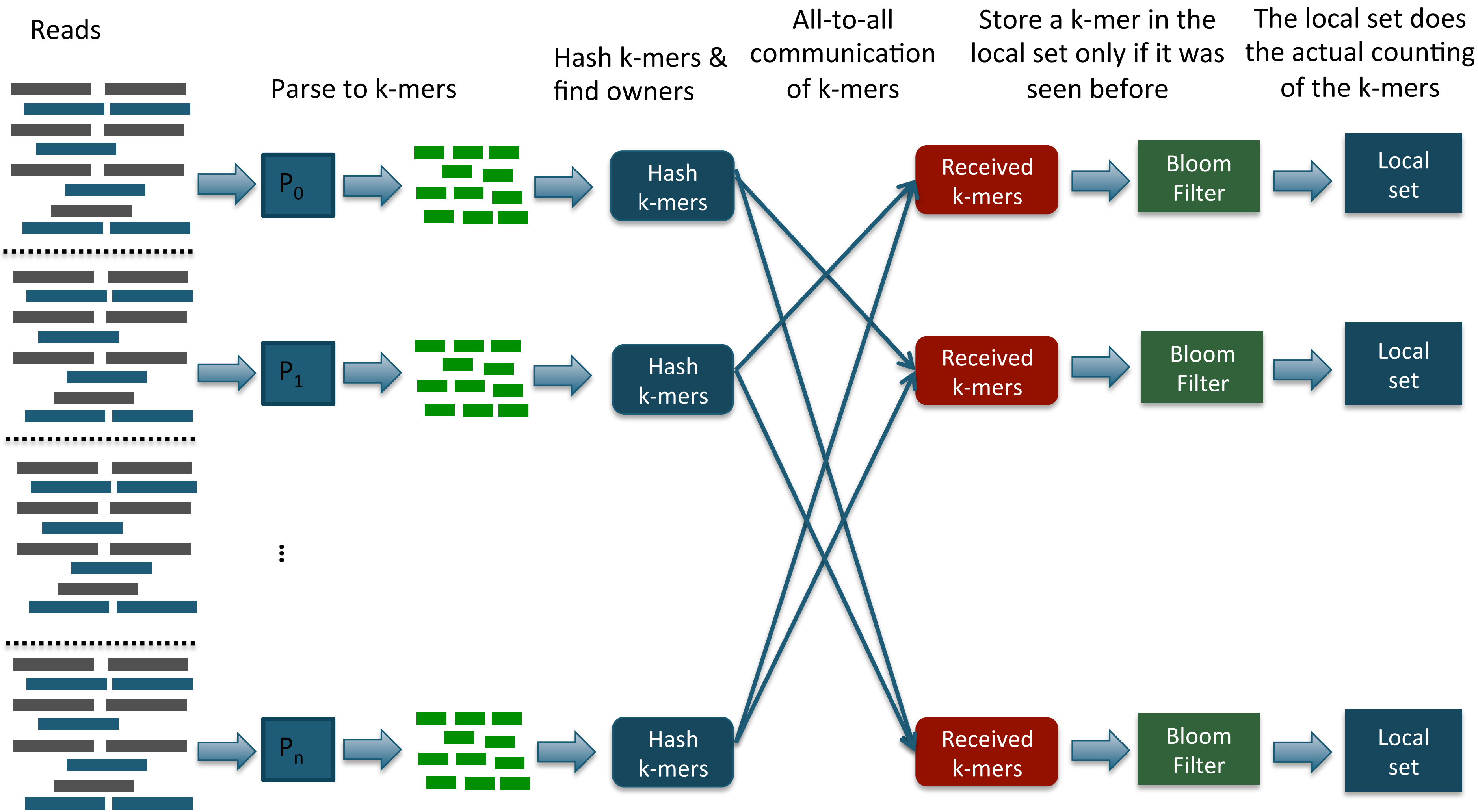
(Assembly output is generally called “contigs” even when a scaffolding phase is included.)

K-Mer Analysis: Pass 1 (I/O + Independent + Reduce)



Also identify high-frequency k-mers at this step

K-Mer Analysis: Pass 2 (Iterative All-to-All)



Bloom Filter

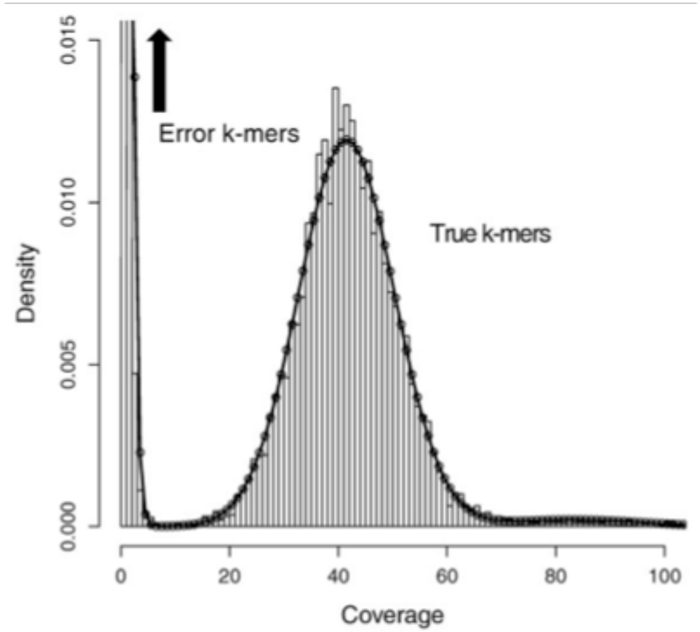
Bloom filter is a *probabilistic* data structure used for membership queries

- **Given a bloom filter, we can ask:**

"Have we seen this k-mer before?"

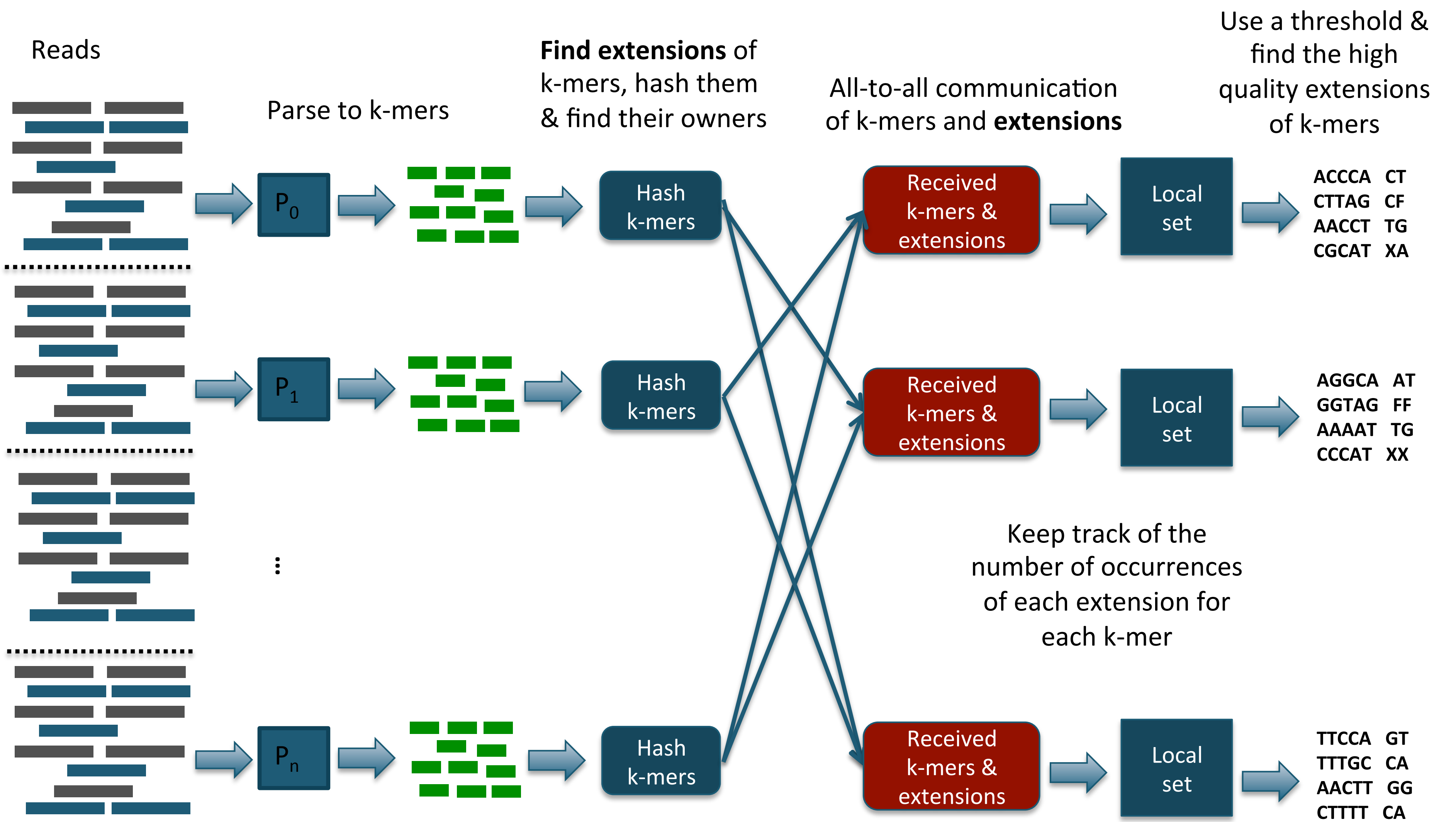
- **No false negatives.**
- **May have false positives**

(in practice 5% false positive rate)



k-mers with frequency =1 are useless (either error or can not be distinguished from error), and can safely be eliminated.

K-Mer Analysis: Step 3 (All-to-All)

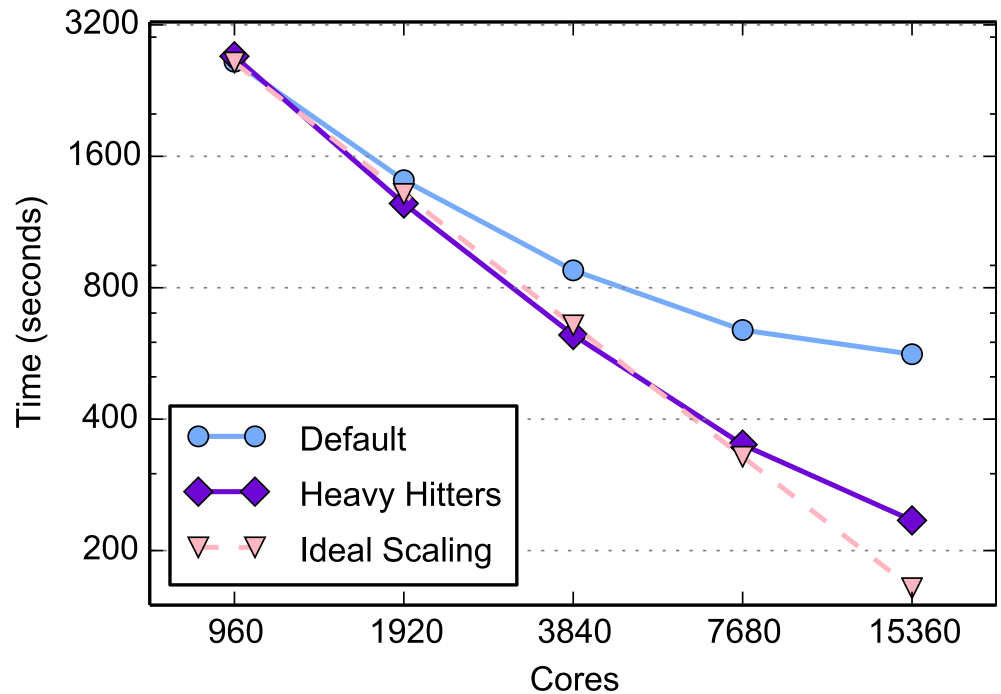


Heavy Hitters

Long-tailed distribution for genomes with repetitive content:

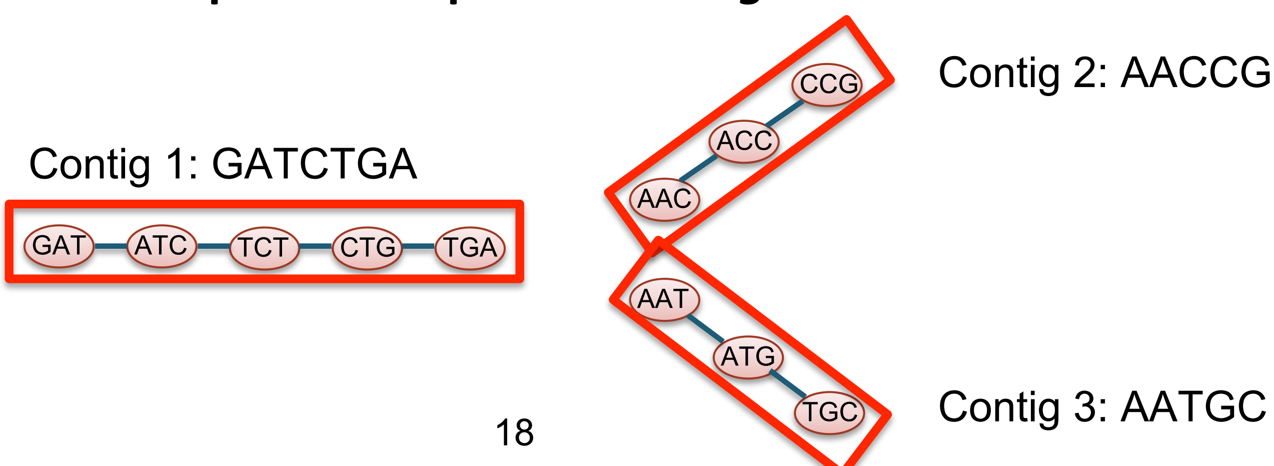
- The maximum count for any k-mer in the wheat dataset is **451 million**
- Our original scheme (SC'14) was "owner counts", after an all-to-all
- Counting an item w/ 451 million occurrences alone is **load imbalanced**

Solution: Quickly identify high-frequency k-mers using minimal communication during the "cardinality estimation" step and treat them specially by using local counters.



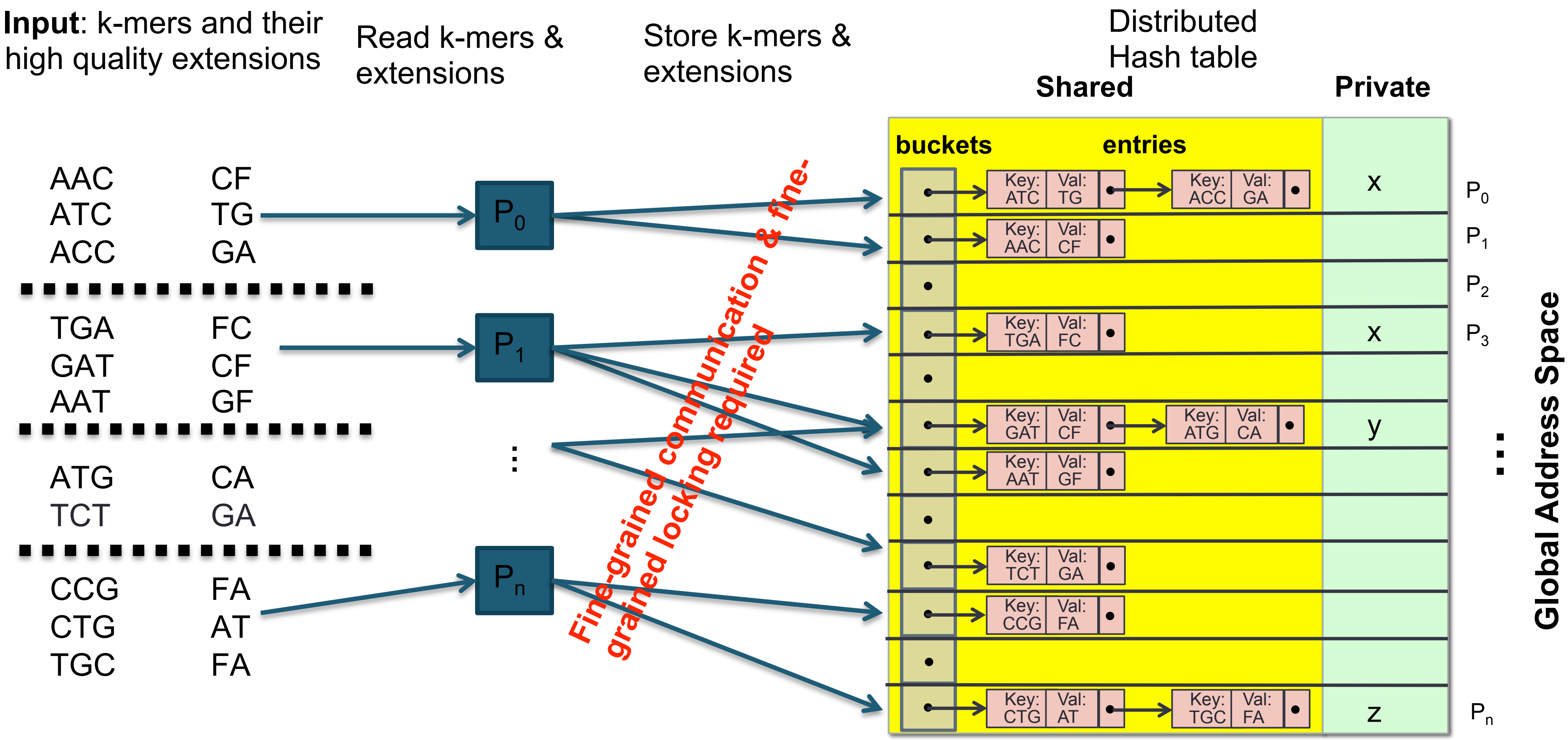
Distributed De Bruijn Graph

- **The de Bruijn graph of k-mers is represented as a hash table.**
- **A k-mer is a node in a graph ⇔ a k-mer is an entry (key) in the hash table.**
- **An edge in the graph connects two nodes that overlap in k-1 bases.**
- **The edges in the hash table can be stored efficiently by storing the extensions of the k-mers as their corresponding values.**
- **The connected components represent *contigs*.**



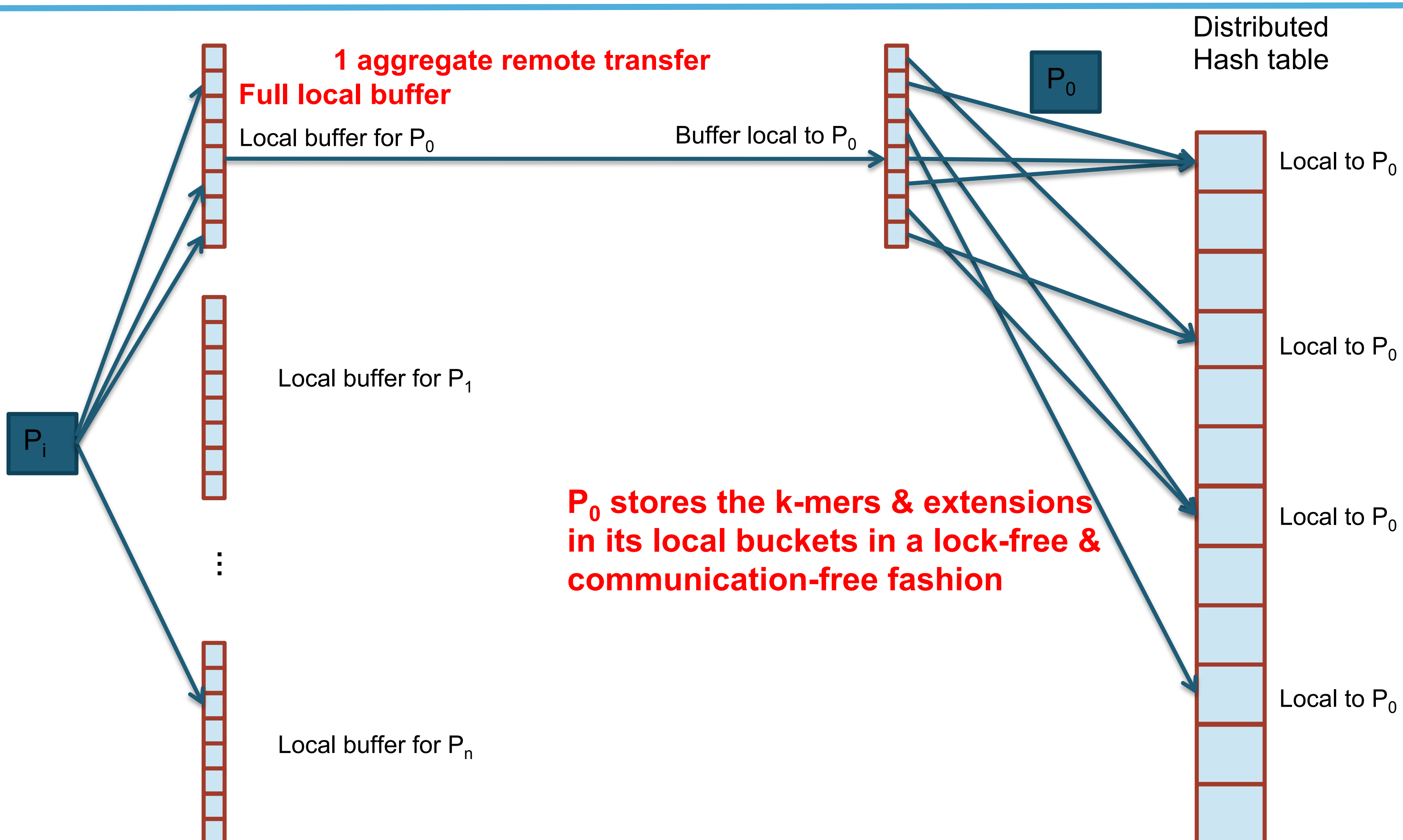
Parallel De Bruijn Graph Construction

Graph construction, traversal, and all later stages are written in UPC to take advantage of its global address space



BERKELEY LAB · Los Alamos NATIONAL LABORATORY EST.1943 · JGI JOINT GENOME INSTITUTE · ECP EXASCALE COMPUTING PROJECT

Aggregating Stores Optimization



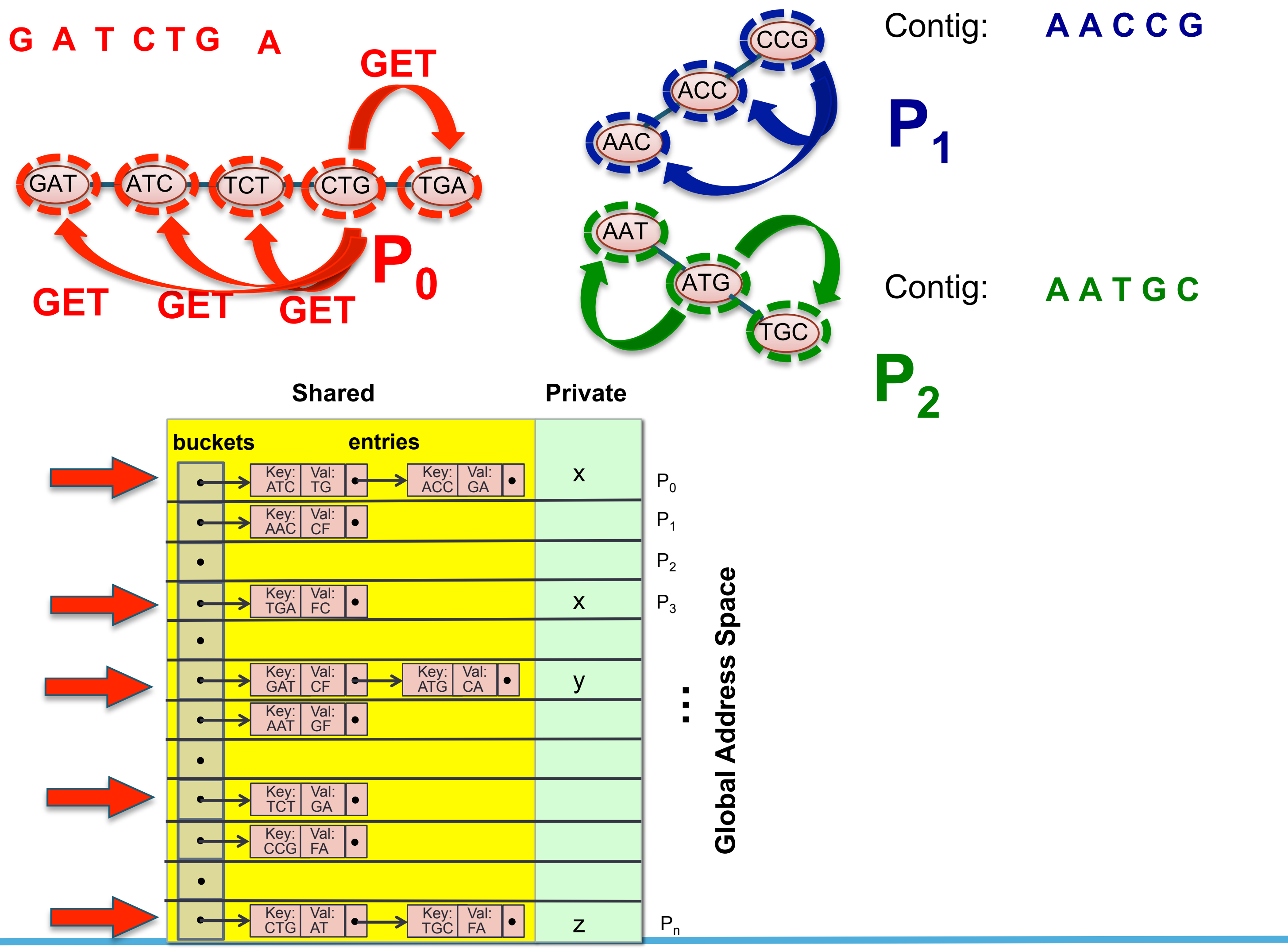
Parallel Graph Construction Challenge

- **Challenge 1: Hash table for de Bruijn graph is huge (at least multiple terabytes)**
  - **Solution:** Distribute the graph over multiple processors.

- **Challenge 2: Parallel hash table construction introduces communication and synchronization costs**
  - **Solution:** Split the construction in two phases and aggregate messages → **10x-20x performance improvement.**

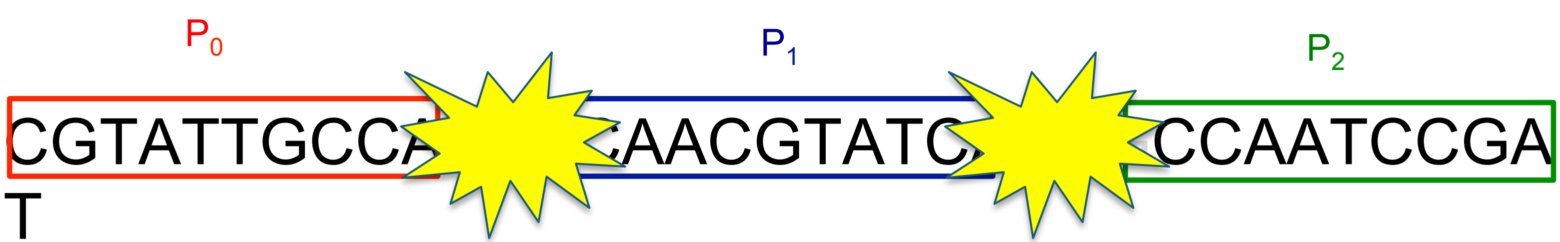
Parallel De Bruijn Graph Traversal

Algorithm: Pick a random k-mer and expand connected component by consecutive lookups in the distributed hash table.

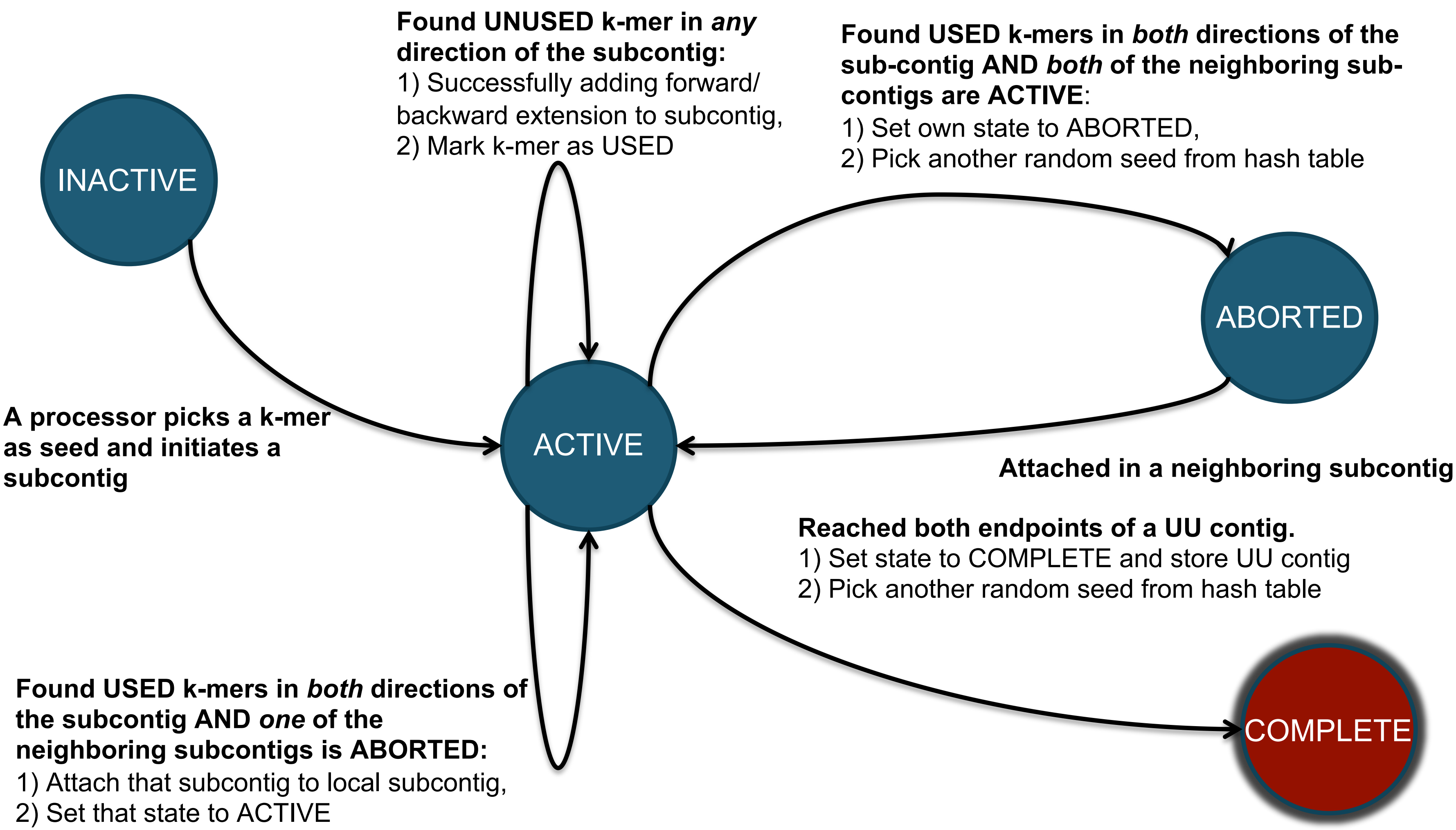


Parallel De Bruijn Graph Traversal

- **<u>Algorithm:</u> Pick a random k-mer and expand connected component by consecutive lookups in the distributed hash table.**
- **Fine-grained, irregular, remote accesses. Need fine-grained parallelization.**
  - Worst case: the result is a single very long chain (high-diameter graph).
  - Global address space and one-sided communication simplifies logic.
  - If multiple processors are working on the same connected component, they cooperate via a **lightweight synchronization protocol**.



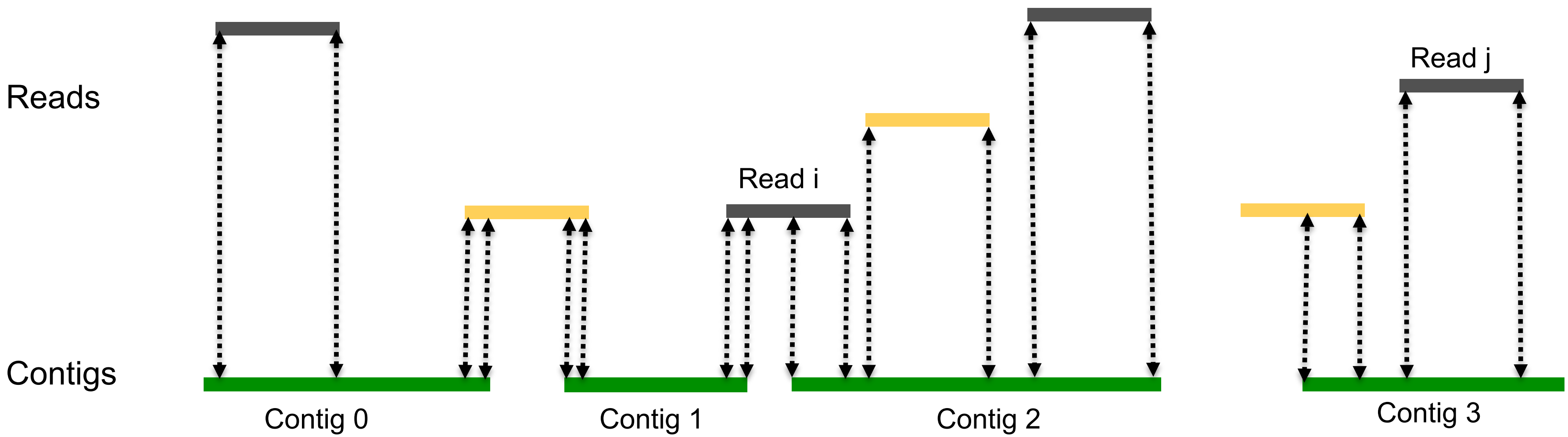
Lightweight synchronization protocol



Scaffolding: Main step maps reads to contigs

- **Input :** a set of reads and a set contigs
- **Output** : detailed alignments of the reads onto the contigs

Alignment with (large) input data; use of hash table to find possible matches



Read ID	Start-pos	End-pos	Contig ID	Start-pos	End-pos
Read i	1	4	Contig 1	152	155
Read i	130	150	Contig 2	1	21
Read j	1	150	Contig 3	101	250

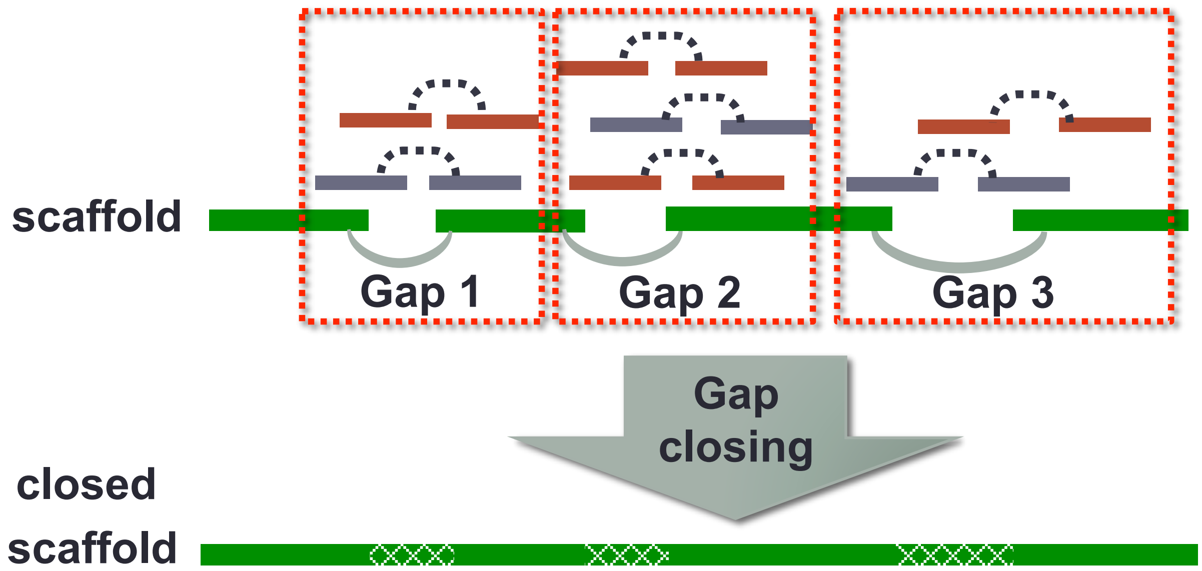
Seed-and-extend Algorithm Summary

1. Given a set of reference sequences (contigs), build an index of them using seeds (substrings) of fixed length **s**.

2. Given a query sequence (read), extract substrings of length **s** and look those up in the reference index → locate candidate contigs to be aligned.

3. Perform an extension algorithm (e.g. Smith-Waterman) on the read and the candidate contig to obtain detailed alignments.

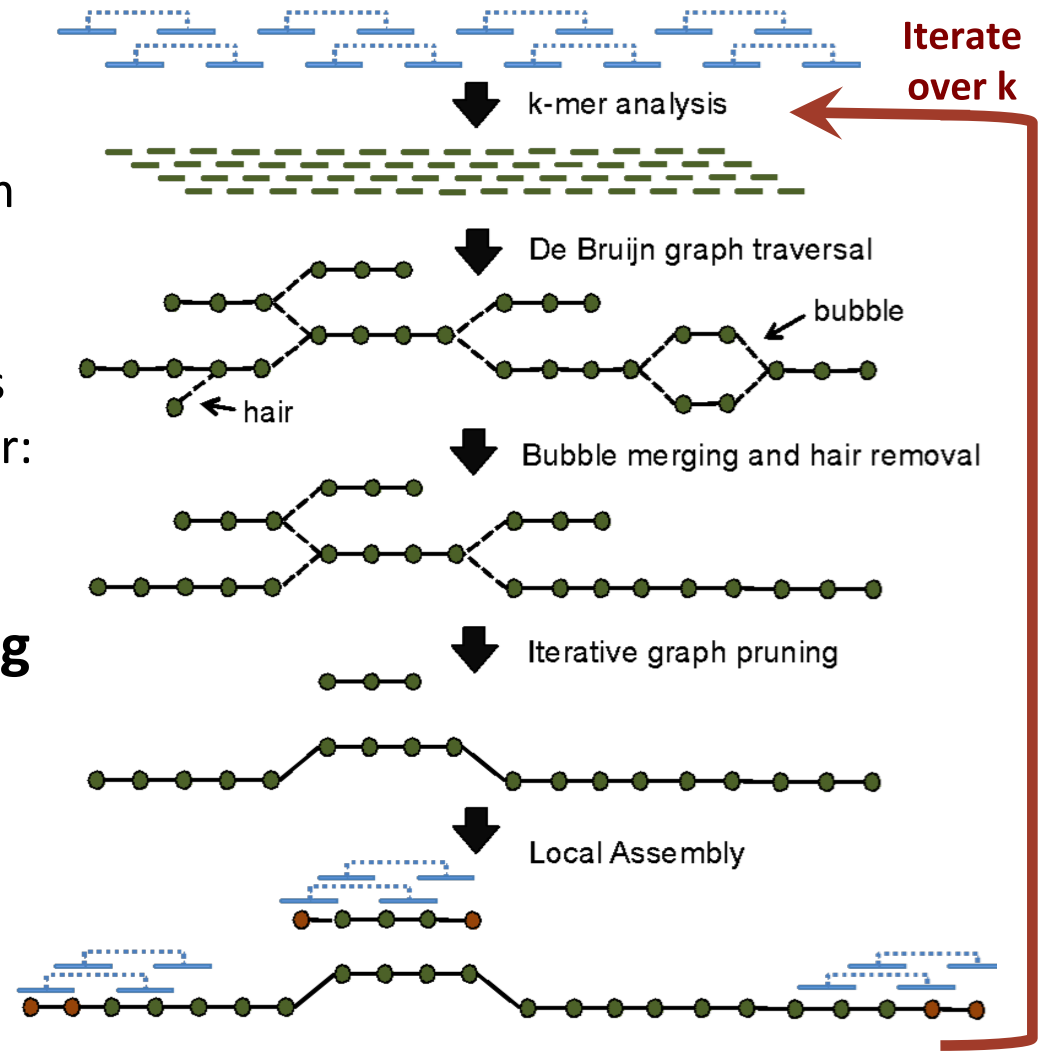
Gap Closing



- Gaps found from alignment and information about distance between paired reads
- Leads to load balancing problems (work stealing)

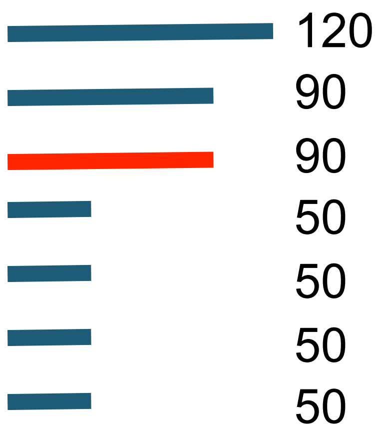
Extending HipMer for Metagenomes

- **Primary change in contig generation on k-mer graph**
  - Iterative contig generation, from small to large k-mer size
  - Small k: low coverage genomes
  - Large k: high-coverage genomes
  - Added/modified steps in HipMer: merging bubbles, iterative pruning, local assembly
- **Some changes to scaffolding**
  - Looping over scaffolding
  - Opportunities for future improvements
- **Scaffolding steps omitted from figure for simplicity**



Quality Metrics

- **A goal of this milestone was to identify good quality metrics**
- **Want to simultaneously maximize contig length and minimize errors**
  - –Sort output contigs by length and find the halfway point.
  - –Several standard metrics, simplest is N50.
  - –***N50*** is the minimal contig length X such that contigs of length at least x account for at least 50% of the total assembly length



Total assembly length = 500

First 3 contigs account for over half

N50 = 90 (length of 3rd contig)

Quality Metrics: Error-free contiguity

Contiguity

- ***N50*** is the contig length such that using longer or equal length contigs produces at least half (50%) the bases of the assembly.
- ***NG50*** is the contig length such that using longer or equal length contigs produces half (50%) the bases of the reference genome. This metric could be computed only if the reference is given.

Error-free contiguity

- ***NGA50*** similar to NG50, but uses lengths of aligned blocks rather than contigs. if a contig has a misassembly with respect to the reference, we break the contig into smaller pieces. Also, if a contig has bases that don't align to the reference, they are not counted in NGA50.
- **Median NGA50:** median NGA50 across all genomes for which NGA50 can be computed (requires sufficient coverage of genome)

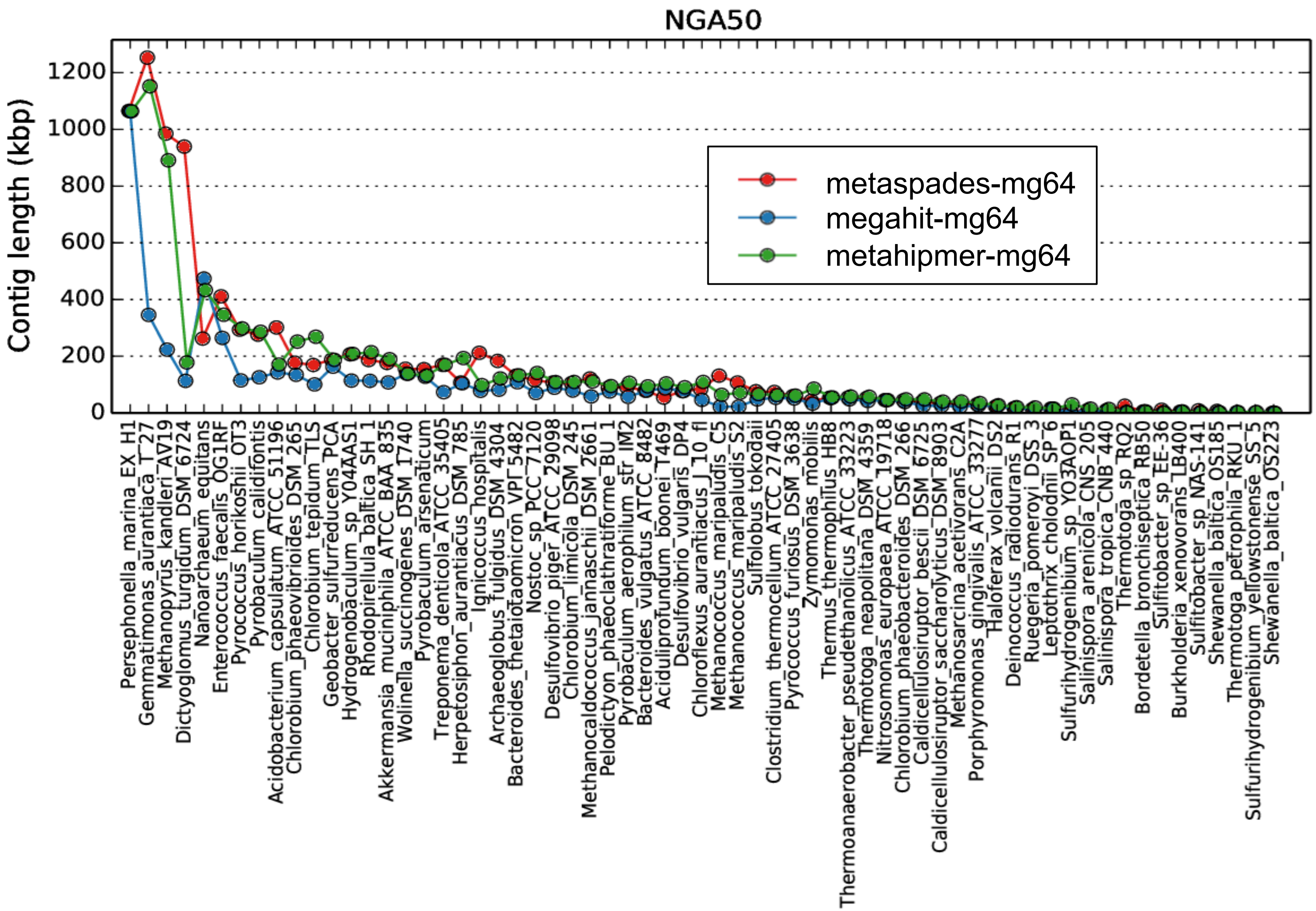
We will use NGA50 and Median NGA50 for error-free contiguity

Results for low complexity data (mg64)

Genome Statistics	MetaHipMer	metaSPAdes	MEGAHIT
Genome Fraction (%)	**95.5**	94.2	95.3
Misassemblies (↓)	456	**239**	309
Mismatches per 100 kpb (↓)	**45.6**	99.3	71.9
Median NGA50	**95129**	89288	58468
Predicted Genes	**202228**	191307	200367

- Low complexity dataset - only 64 genomes
- All assemblers find most genomes, with low errors
- MetaHipMer has more misassemblies due to small number of species (recall this is not normalized to assembly length)
- MetaHipMer has fewer mismatches, greater predicted genes and better NGA50

Error free contiguity (NGA50) on mg64



Results for medium complexity data (CAMI)

Genome Statistics	MetaHipMer	metaSPAdes	MEGAHIT
Genome Fraction (%)	66.3	68.7	**73.0**
Misassemblies (↓)	2031	**1210**	1579
Mismatches per 100 kpb (↓)	**80.3**	152.6	100.1
Median NGA50	**24906**	14219	16123
Predicted Genes	632459	635289	**637085**

- Medium complexity dataset - 225 genomes
- Lower coverage than mg64, with more errors
- MetaHipMer has a lower genome fraction, but better NGA50 than the others

Results for high complexity data (MC04)

Genome Statistics	**MetaHipMer**	**metaSPAdes**	**MEGAHIT**
Genome Fraction (%)	28.6	**32.7**	32.3
Misassemblies (↓)	12730	**9014**	17479
Mismatches per 100 kpb (↓)	**27.4**	70.3	67.9
Median NGA50	**15098**	10540	12744
Predicted Genes	**1386760**	1381479	1355607

- High complexity dataset - 800 genomes
- Low coverage from all assemblers due to challenging nature of dataset
- High rates of misassembly
- MetaHipMer has a lower genome fraction, but better NGA50 than the others

MetaHipMer Performance (preliminary results)

- **Performance on a mock community**
  - metaSPAdes: **11.5 hours** on a 32 core machine with 500 GB of RAM
  - metaHipMer: **17 minutes** on 80 Edison nodes (~2K cores) – **41x faster**

- metaSPAdes can't scale to the massive datasets (up to 80x larger)
  - For assembling TaraOceans dataset (8 TBytes, 80x larger), it would take **38 days**, on a machine with **40 TBytes** of memory.

- metaHipMer modules scale to full machine !
  - Assuming 40% efficiency, the TaraOceans dataset could be assembled in < **1 hour** using full Edison

Summary on MetaGenomeAssembly

- **Assembly is an HPC problem**
  - (Meta)HipMer can handle previously unassembled data sets
- **Hardware support**
  - High injection rate RDMA communication; low latency; remote atomics
  - High bisection bandwidth (synchronous and asynchronous all-to-all)
- **Software support**
  - PGAS model for distributed data structures, one-sided communication
  - Hierarchical algorithms probably useful, but scaling well on/off node with UPC
- **Benchmarks and proxy applications**
  - NERSC-9 "Meraculous" benchmark is contig generation (graph/hash table)
  - Latency / bandwidth tests ("roofline like") for remote atomics, async all-to-all
- **MetaHipMer is a parameterized toolkit for HPC genome analysis**
  - Quality results comparable to other state-of-art assemblers, but can solve problems they cannot due to memory/performance scaling

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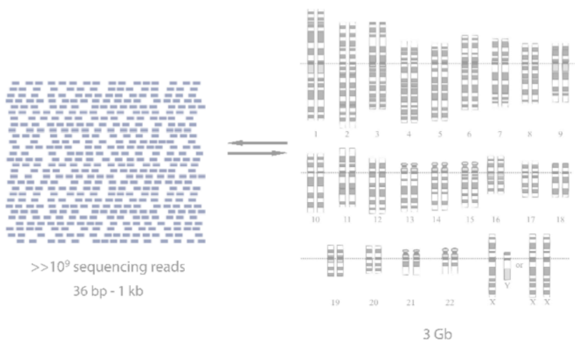

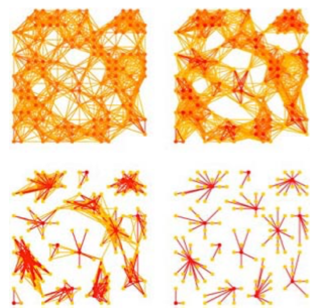

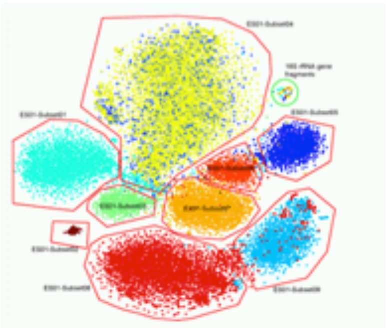

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Graph algorithms, Hash Tables, alignment (Smith-Waterman)	Machine learning (clustering), sparse linear algebra / graphs	Alignment, Machine learning (dimensionality reduction), linear algebra
Fine-grained comm., all-to-all, remote atomics and fast I/O	Fast barriers, subset reductions	All-to-all

PI: Katherine Yelick (LBNL)

Discovering Biosynthetic Gene Clusters

"A **biosynthetic gene cluster** is a physically clustered group of two or more genes in a particular genome that *together encode a biosynthetic pathway* for the production of a specialized metabolite (including its chemical variants)"

[Medema et al. "Minimum information about a biosynthetic gene cluster." Nature chemical biology, 2015]

Input: Sparse matrix encoding genes and their nonzero pairwise similarities

Method: High-performance Markov clustering (HipMCL)

Desired scale: 10s of billions of genes, trillions of nonzero pairwise similarities

- Use proteins rather than genes (constant factor)
- Find connected components first (heuristic)
- Requires supercomputers

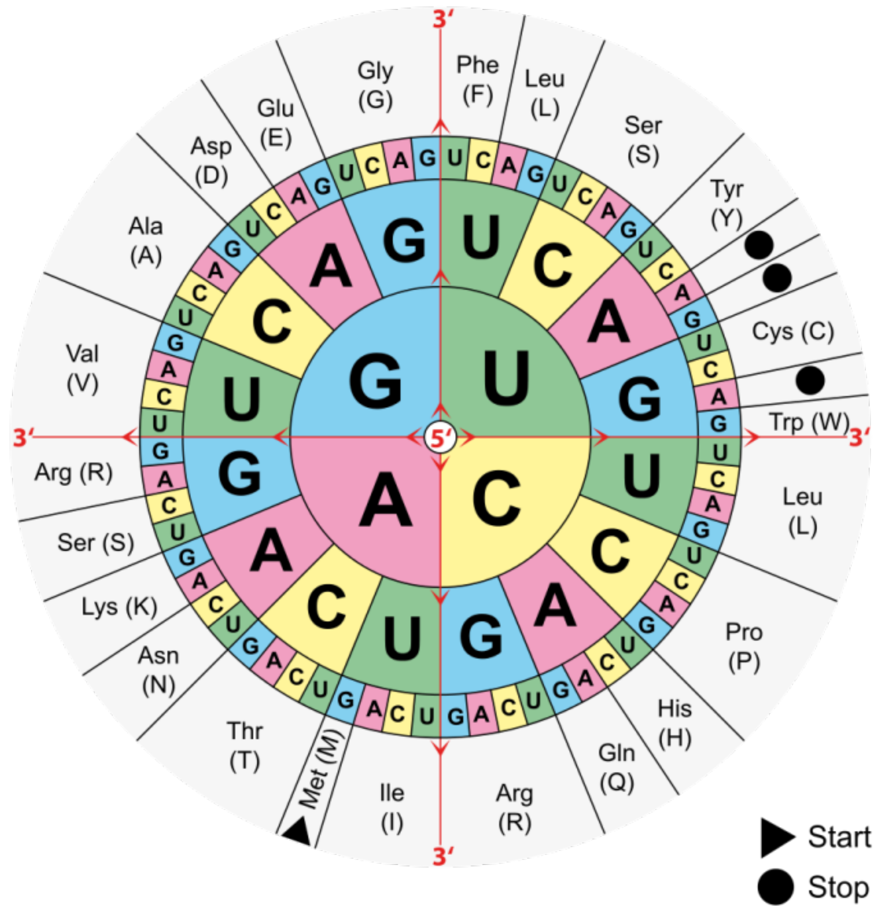
MCL is the de-facto algorithm in community for finding gene/protein families

"…MCL is remarkably robust to graph alterations…"

[Brohée, van Helden, "Evaluation of clustering algorithms for protein-protein interaction networks", 2006]

DNA to Proteins

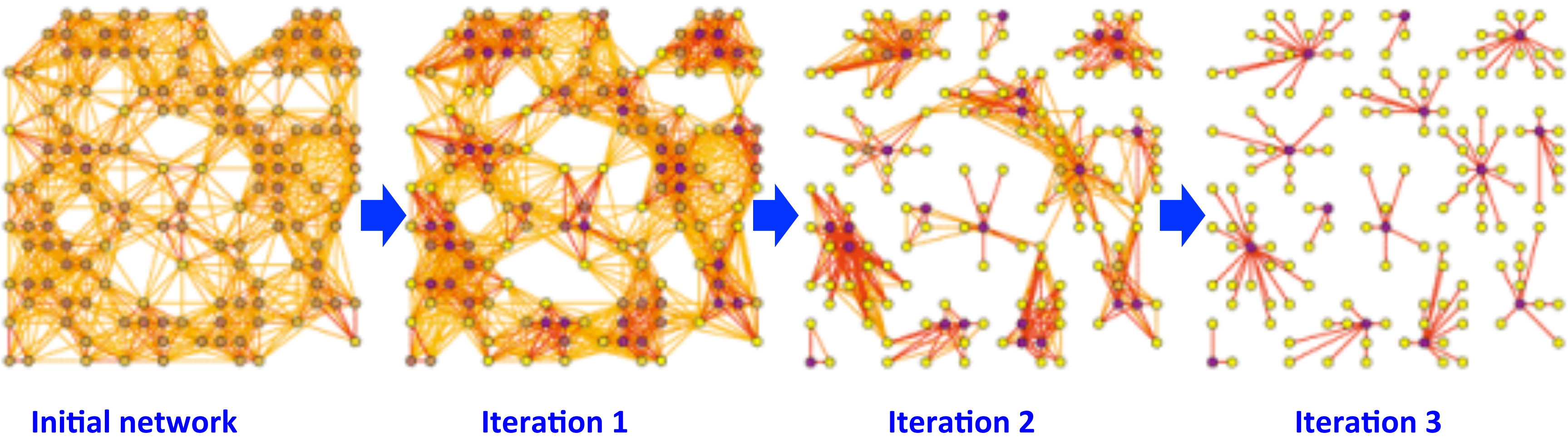
- DNA based on 4-character alphabet
- Three-letter codons represent one of the 20 regularly used amino acids
- Start and Stop codons mark the beginning/ end of genes



Markov Cluster algorithm

Input: Adjacency matrix A (sparse)

Image source: http://micans.org/mcl/



At each iteration:

Step 1 (Expansion): Squaring the matrix while
pruning (a) small entries, (b) denser columns

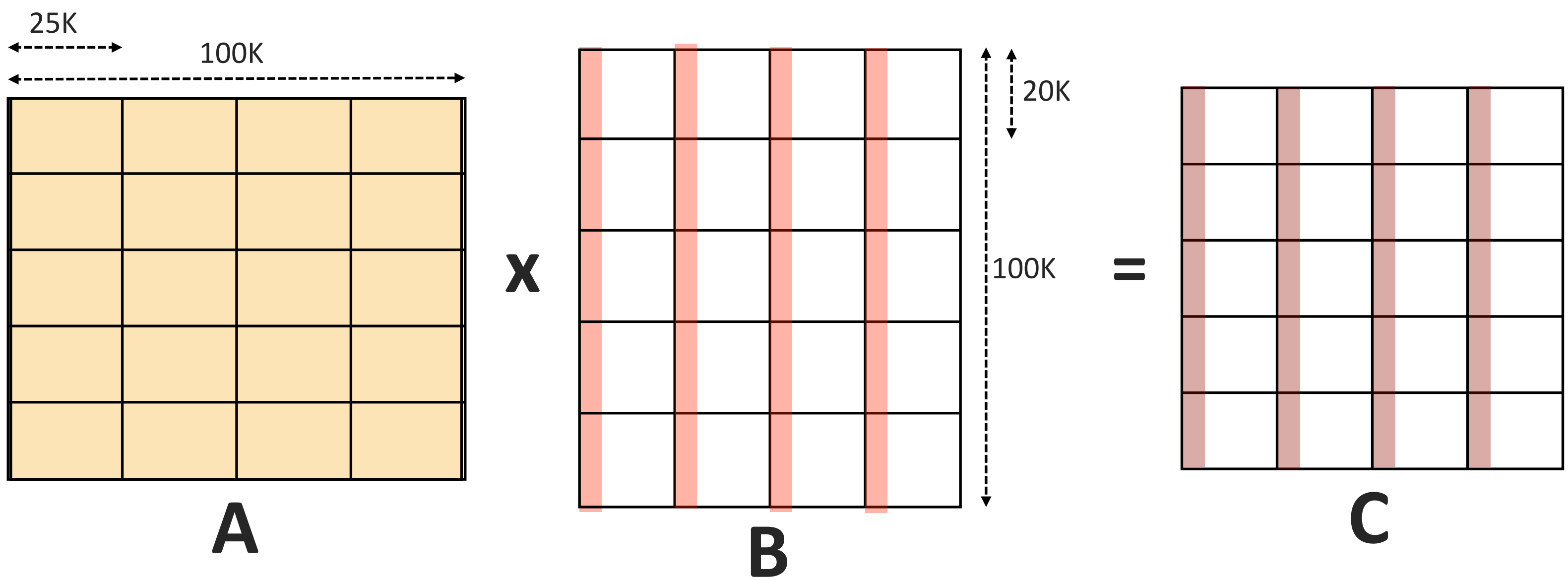
Naïve implementation: sparse matrix-matrix product (SpGEMM), followed by column-wise top-K selection and column-wise pruning

Step 2 (Inflation) : taking powers entry-wise

Scalable Distributed Memory, Memory Efficient SpGEMM

$\sqrt{p} \times \sqrt{p}$ Processor Grid

Split B into k pieces

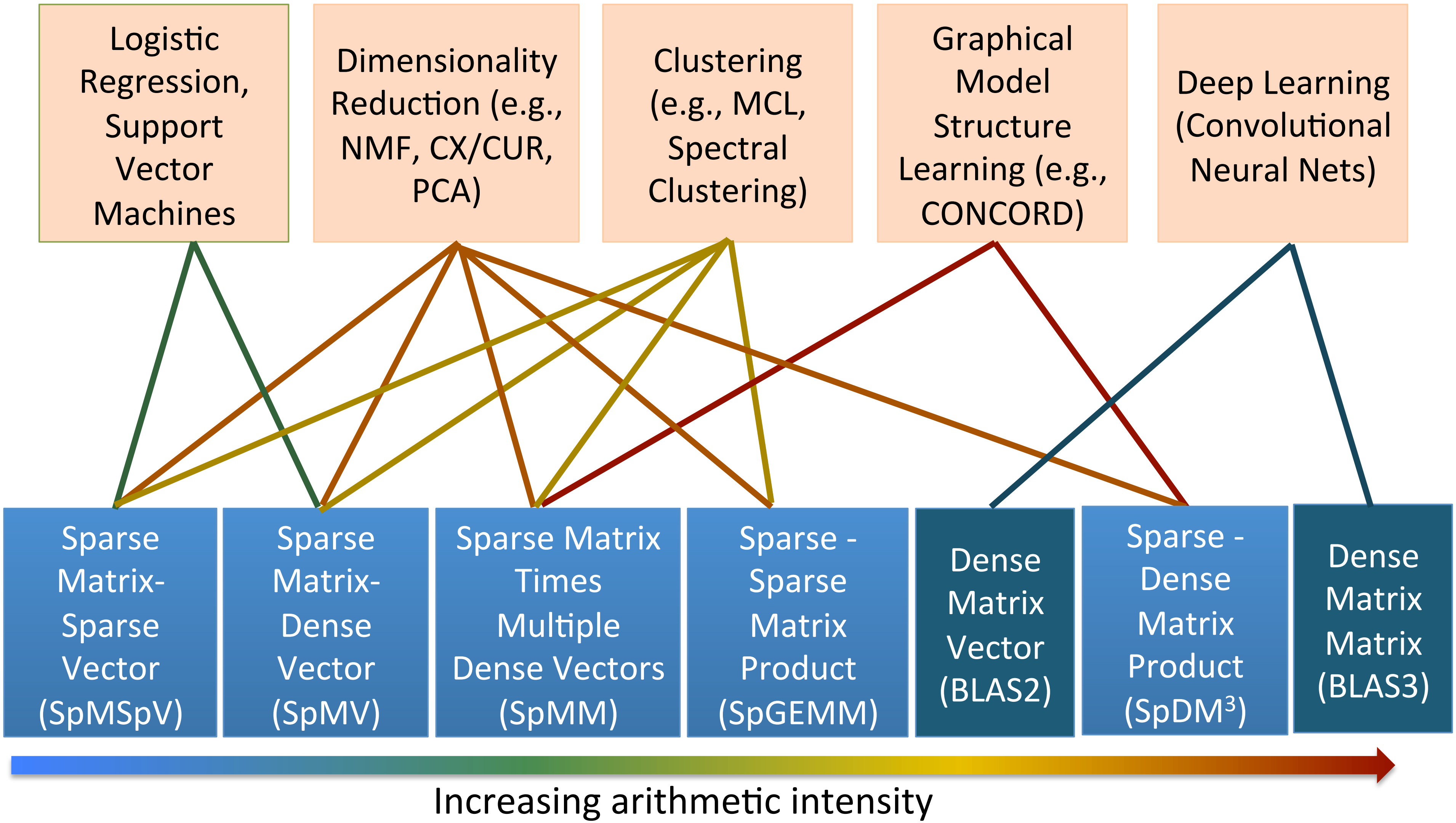


- Parts of the result is produced and pruned
- Memory requirement can be significantly reduced by increasing k
- However, A is needed to be broadcasted k times
- With k=20: MCL ran on 64 nodes of Cori in about 20 minutes

Challenges in distributed-memory MCL

- **"Small" test dataset from Joint Genome Institute**
  – 47 Million proteins
  – 14 Billion interaction
  – 4.2 Trillion expected nonzeros in A^2
  – Memory requirement of naïve implementation: ~**100 TB**
- **Ultimate dataset is 1000 times larger.**
- **Memory efficient SpGEMM algorithm being developed**
  – Since the output is sparsified by column-wise pruning, we ***create A² part by part and prune on the fly*** to save memory.
  – Need to find ***the kth largest entry of each column at every iteration***: Trivial in single node, harder in distributed memory.
- **Interpretation of final clusters need distributed-memory connected components**
  – Not a bottleneck: cheaper and only done once (not per iteration)

Machine Learning Mapping to Linear Algebra in General



Aydin Buluc, Sang Oh, John Gilbert, Kathy Yelick

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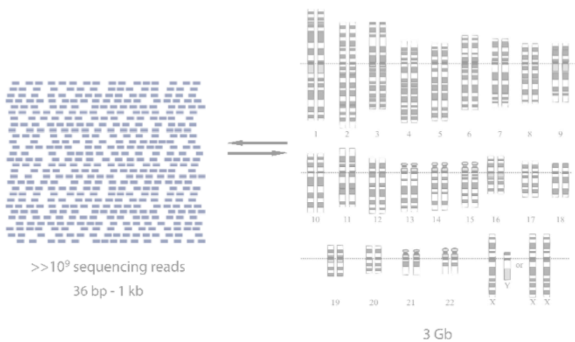

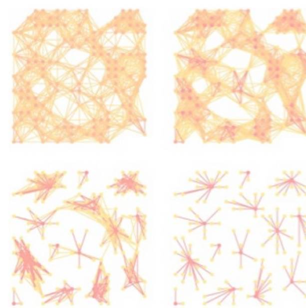

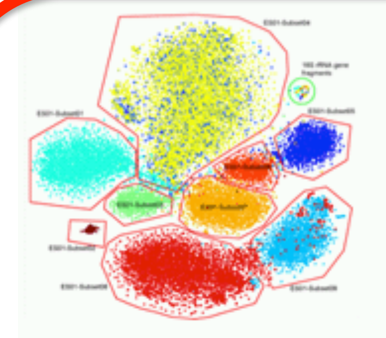

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PI: Katherine Yelick (LBNL)

Metagenome composition description

Three approaches to describe and analyze the metagenome sequencing dataset:

"features"

1) taxonomic composition-based (who is in the metagenome)
2) functional annotation-based (what can this metagenome do)
3) k-mers (shingles) composition-based (i.e., shingle frequency vector-based)

K-mer based approach (MASH) is:

– More resilient to errors /degradation of DNA, coverage bias, composition, short and redundant fragments

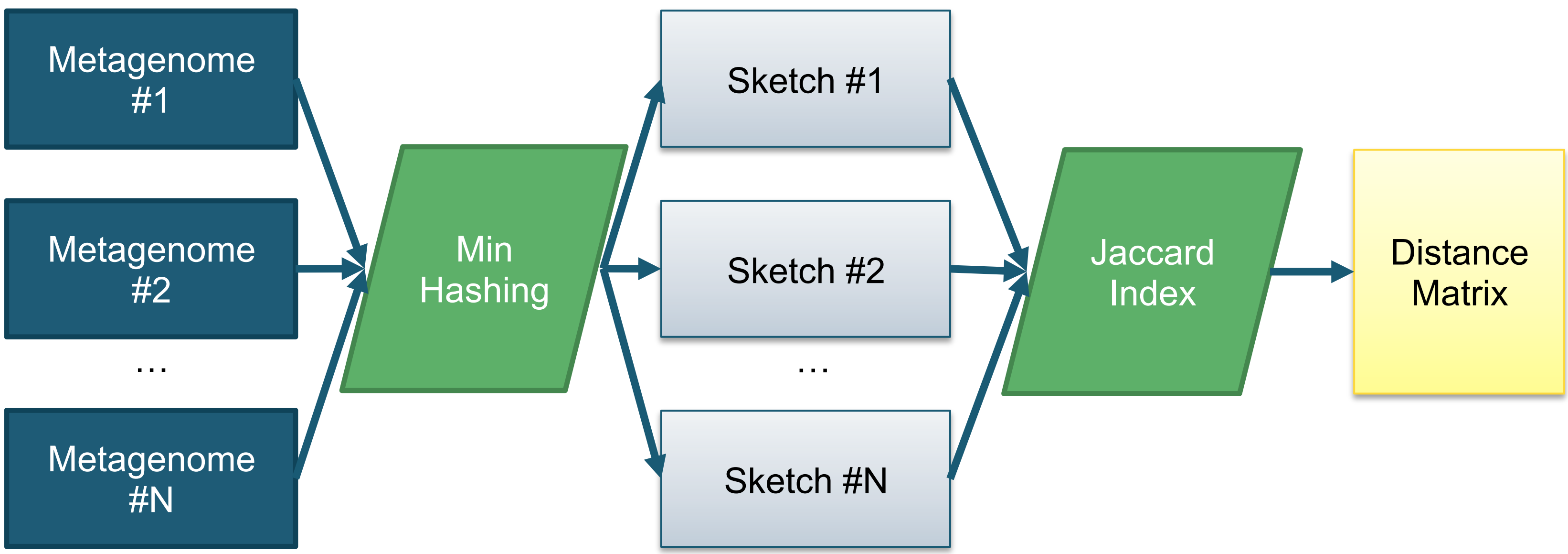
Taxonomic (GOTTCHA) or functional approaches:

– Fast and memory efficient; also gives some semantic information about differences

MASH – an alternate MinHash based approach

- **"…extends MinHash dimensionality-reduction technique to include a pairwise mutation distance and *P* value significance test, enabling the efficient clustering and search of massive sequence collections…"**

Ondov et al. Genome Biology (2016) 17:132 DOI 10.1186/s13059-016-0997-x



MASH distance is sufficiently accurate

Ondov et al. Genome Biology (2016) 17:132 DOI 10.1186/s13059-016-0997-x

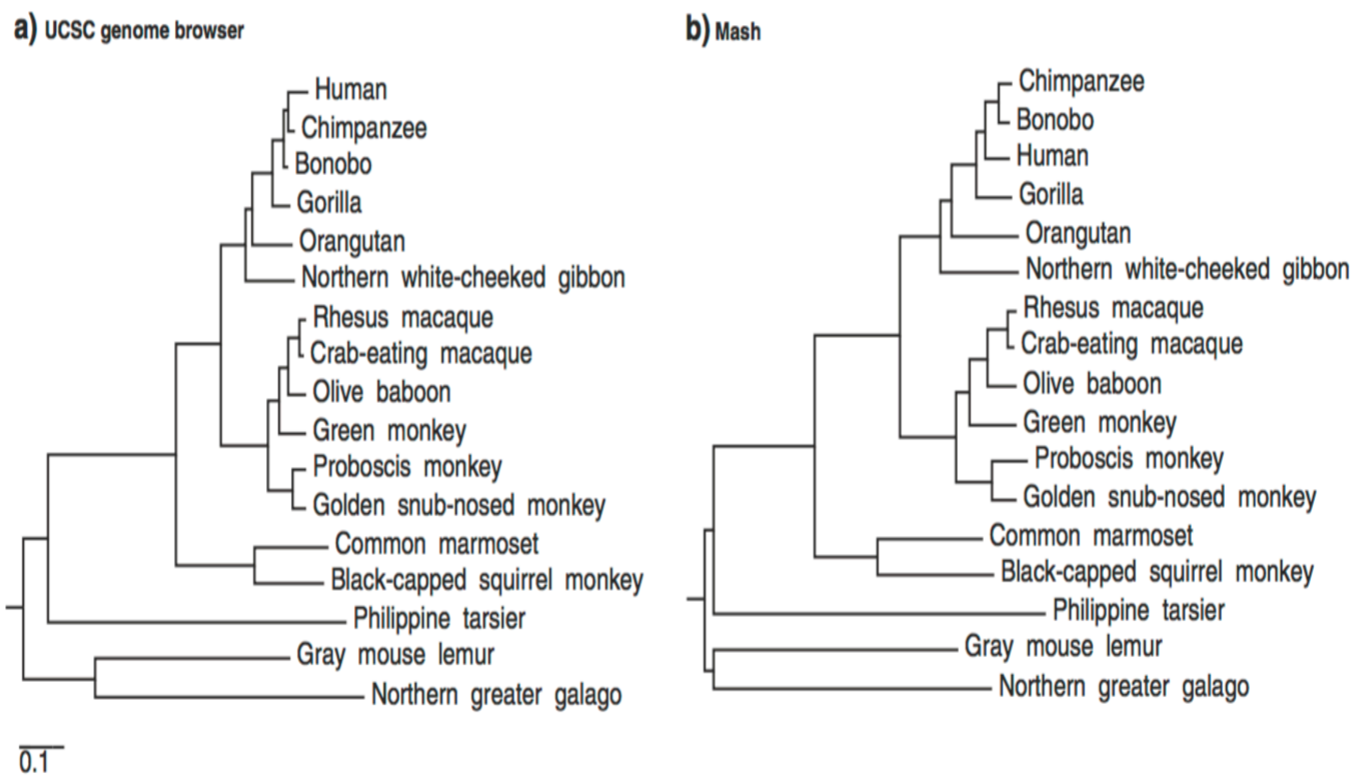
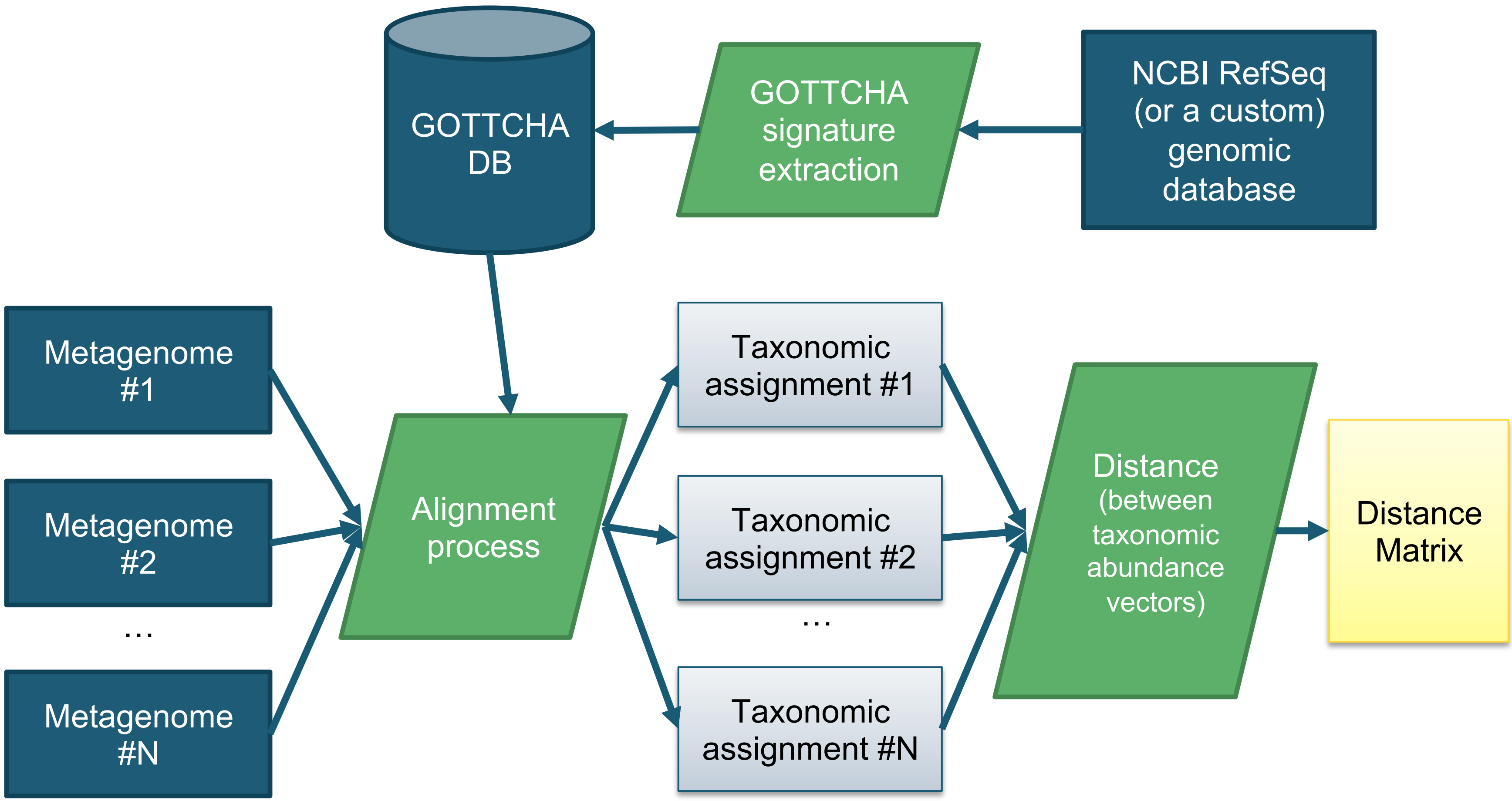


Fig. 4 Primate trees from the UCSC genome browser and Mash. **a** A primate phylogenetic tree model from the UCSC genome browser, with branch lengths derived from fourfold degenerate sites extracted from reference gene multiple alignments. **b** A comparable Mash-based tree generated from whole genomes using a sketch size of $s = 1000$ and k-mer size of $k = 21$. Additional file 1: Figure S6 includes this Mash tree with five additional mammals of increasing divergence

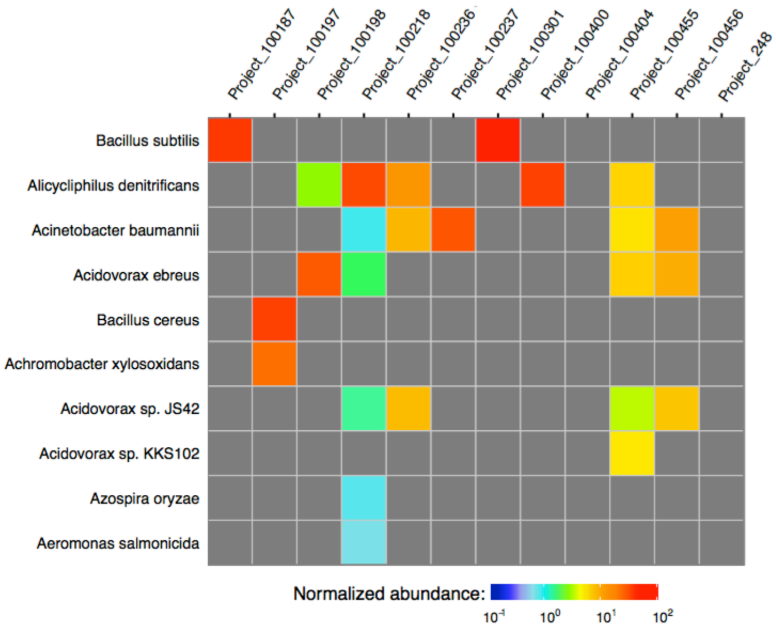
MASH pros and cons

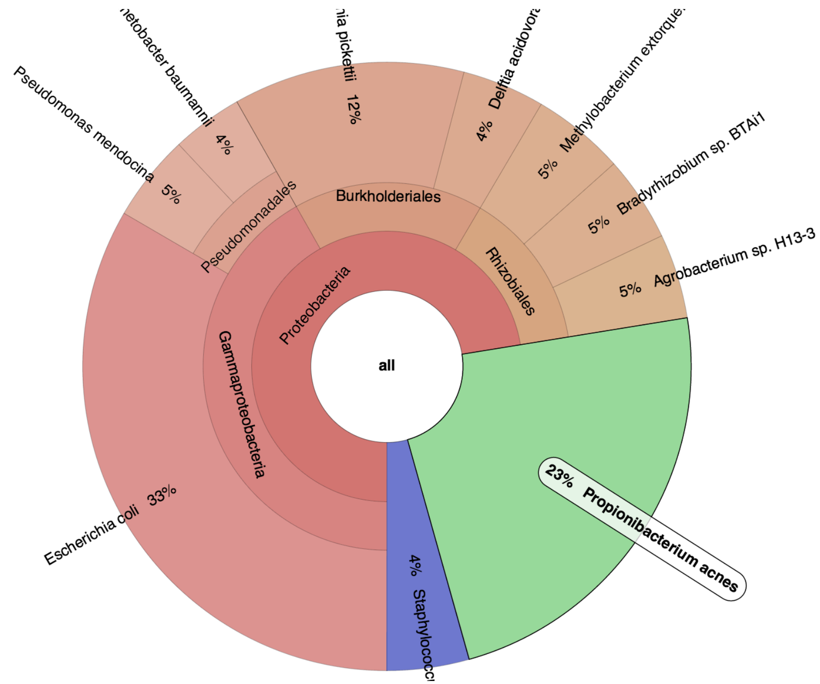
- **+ REALLY fast (just four simple consecutive steps).**
  - –K-mer extraction -> MinHash -> sketch -> distance
- **+ Reduces metagenome data size significantly**
  - –|sketch|<<|metagenome|
- **+Can cluster metagenomes with a lot of unknowns (un-annotatable)**
- **–There is a threshold in k-mer and sketch sizes which affects the tradeoff between specificity/sensitivity and the computational complexity/space requirements**
  - –The threshold can be estimated if the divergence between metagenomes can be roughly approximated
- **–Provides *no insight* into the genome taxonomic or functional composition**

GOTTCHA - Genome Signature Discovery



GOTTCHA – an interactive comparative visual display





GOTTCHA2 implementation

- Developed at LANL, will be released with GPL-2
- The database creation tool is not yet publicly available, coded in MPI C/C++ (we distribute binary database files)
- The taxonomic assignment pipeline is GPL-2 open sourced https://github.com/poeli/GOTTCHA2
- Currently relies on BWA for alignment https://github.com/lh3/bwa, distributed under GPL-3, we are working on the MerAligner integration from MetaHipMer

Summary on Post-Assembly Analytics

Finding gene clusters related groups (ML – clustering)

- Large graph / sparse matrix problems (sparse matrix-matrix multiply)
- Needs memory-efficient as well as scalable distributed memory approach

- **Hardware support (preliminary)**
  - High bisection bandwidth; low latency; fast I/O
- **Software support**
  - Sparse matrix and graph libraries (e.g., Graph BLAS approach)
  - MPI (bulk synchronous so far)

Comparative metagenome analysis (ML – dimensionality reduction)

- Dominated by alignment; see MerAligner discussion
- Also statistical (counting) and sparse matrix algorithms

ExaBiome: Exascale Solutions for Microbiome Analysis (1.2.1.20)

Application Domain

- **Application Area:** Microbiomes are integral to the environment, health, and biomanufacturing. They occur in communities that are collected and sequenced as a group, called metagenomes. These metagenomes lead to some of the most computationally demanding problems in bioinformatics, including assembly, protein clustering and comparative whole-metagenome analysis.
- **Challenge Problem**:
- *4 Year:* Provide scalable tools for and complete assembly and analysis of metagenome data in SRA and IMG.
- *10 Year:* Perform high quality assembly and analysis of 1 million metagenomes on an early exascale system to reveal microbial functions for environmental remediation and other applications.

Physical Models and Code(s)

- **Physical Models**: *Assembly of genomes using DNA sequence fragments from metagenomes. Clustering of genes culled from metagenomes to identify novel protein families. Comparative analysis across whole metagenomes.*
- **Codes**: (Meta)*HipMer, HipMCL, Mash, GOTCCHA, Combinatorial BLAS*
- **Motifs:** *Graph Traversal, Sparse Matrices, Dynamic Programming, (possibly Graphical Models)*

Partnerships

Co-Design Centers:

- *ExaGraph (if funded) and possibly AMReX*

Software Technology Centers:

- *Programming: PAGODA, PROTEAS, Legion, ExaMPI and OMPI-X*
- *Performance/Systems: HPCToolkit, PAPI, Qthreads, ExaHDF5*
- *Libraries: xSDK4ECP, Sparse Solvers, Trilinos*

Application Projects:

- *Cancer, NWChem*

First Year Development Plans

Develop high quality, scalable metagenome assembly

- *Extend HipMer with features for metagenomes, now called MetaHipMer (currently a research prototype)*
- *Evaluate and improve quality and scalability of MetaHipMer; compare to other metagenome assemblers*
- *Release MetaHipMer*

Use machine learning algorithms for metagenome analysis

- *Develop and demonstrate HipMCL clustering algorithm for genes from metagenomes*
- *Evaluate use of MerAligner (a phase in MetaHipMer) for GOTTCHA's metagenome characterization*

PI