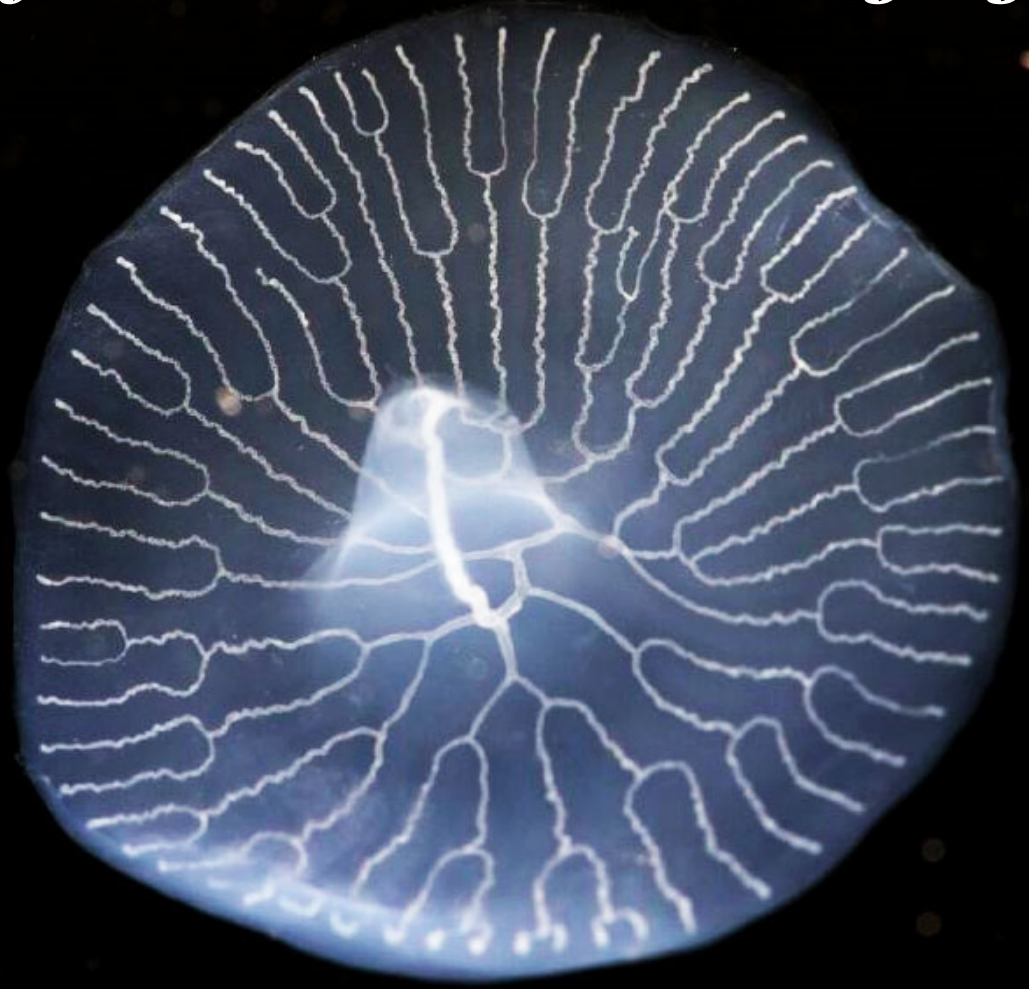
History of & Introduction to Phylogenomics



Antonis Rokas
Department of Biological Sciences, Vanderbilt University
http://www.rokaslab.org
@RokasLab

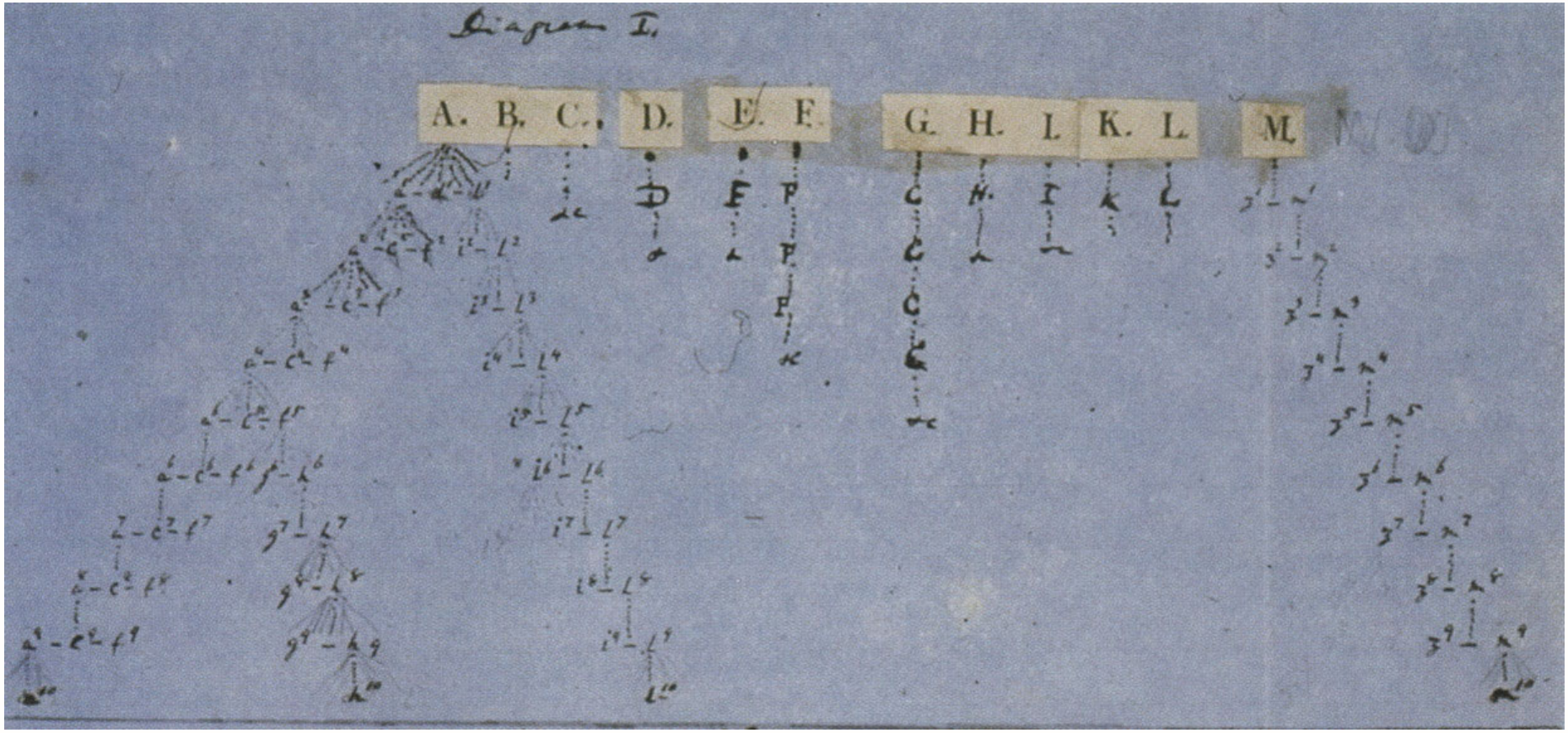
Lecture Outline

- **From Darwin to Phylogenomics**

---------- Coffee Break ----------

- **Prospects and Challenges of Phylogenomics**

Darwin's Tree



"As buds give rise by growth to fresh buds, and these, if vigorous, branch out and overtop on all sides many a feebler branch, so by generation I believe it has been with the great Tree of Life, which fills with its dead and broken branches the crust of the earth, and covers the surface with its ever branching and beautiful ramifications"

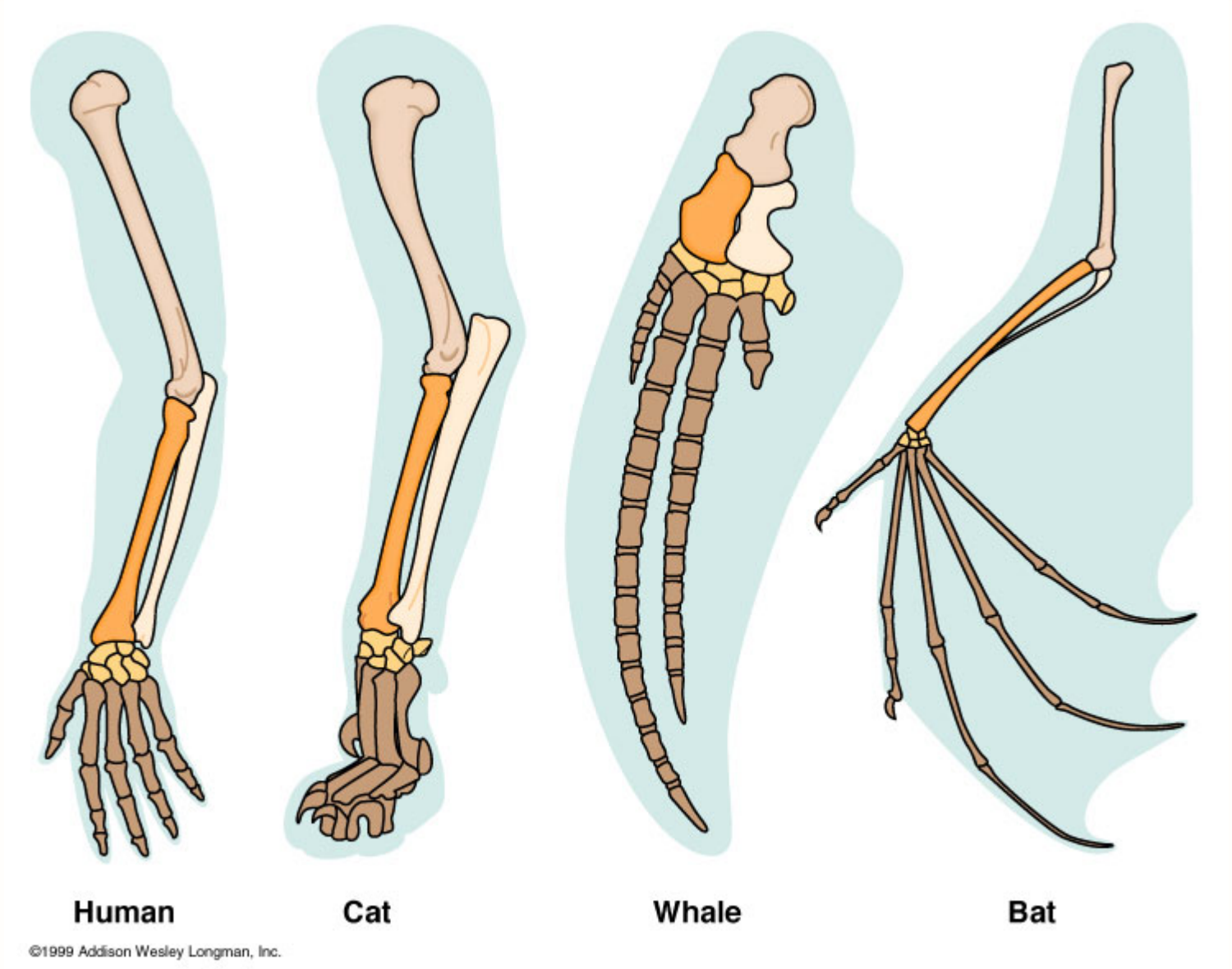
Darwin's hand-made proof of the famous diagram in his Origin of Species; Maderspacher (2006) Curr. Biol.

and instinct as the summing up of many contrivances, each useful to the possessor, nearly in the same way as when we look at any great mechanical invention as the summing up of the labour, the experience, the reason, and even the blunders of numerous workmen; when we thus view each organic being, how far more interesting, I speak from experience, will the study of natural history become!

A grand and almost untrodden field of inquiry will be opened, on the causes and laws of variation, on correlation of growth, on the effects of use and disuse, on the direct action of external conditions, and so forth. The study of domestic productions will rise immensely in value. A new variety raised by man will be a far more important and interesting subject for study than one more species added to the infinitude of already recorded species. Our classifications will come to be, as far as they can be so made, genealogies; and will then truly give what may be called the plan of creation. The rules for classifying will no doubt become simpler when we have a definite object in view. We possess no pedigrees or armorial bearings; and we have to discover and trace the many diverging lines of descent in our natural genealogies, by characters of any kind which have long been inherited. Rudimentary organs will speak infallibly with respect to the nature of long-lost structures. Species and groups of species, which are called aberrant, and which may fancifully be called living fossils, will aid us in forming a picture of the ancient forms of life. Embryology will reveal to us the structure, in some degree obscured, of the prototypes of each great class.

When we can feel assured that all the individuals of the same species, and all the closely allied species of most genera, have within a not very remote period de-

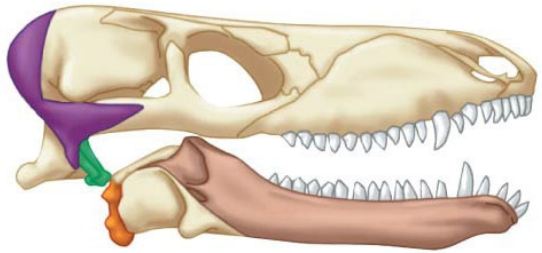
Comparative Morphology of Extant Organisms



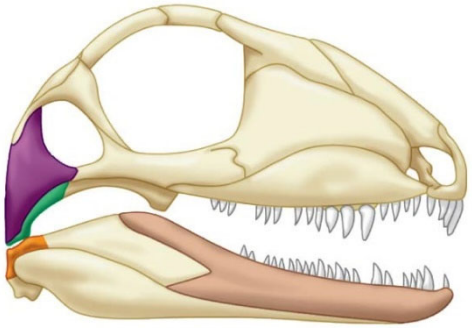
http://www.mun.ca/biology/scarr/139393_forelimb_homology.jpg

Comparative Anatomy of Fossils

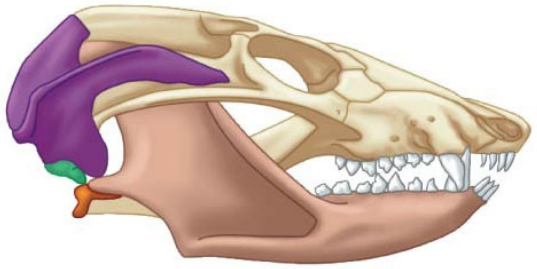
Early cynodont (260 mya)



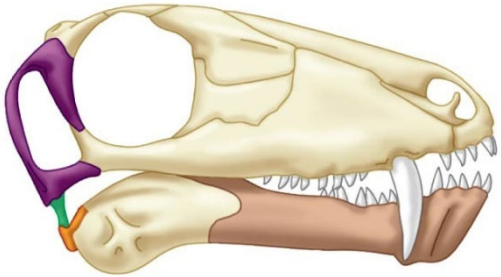
Synapsid (300 mya)



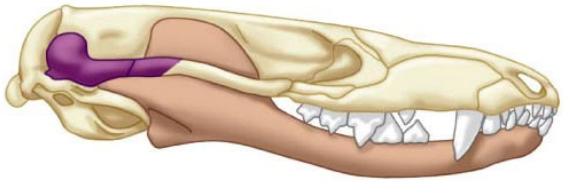
Later cynodont (220 mya)



Therapsid (280 mya)



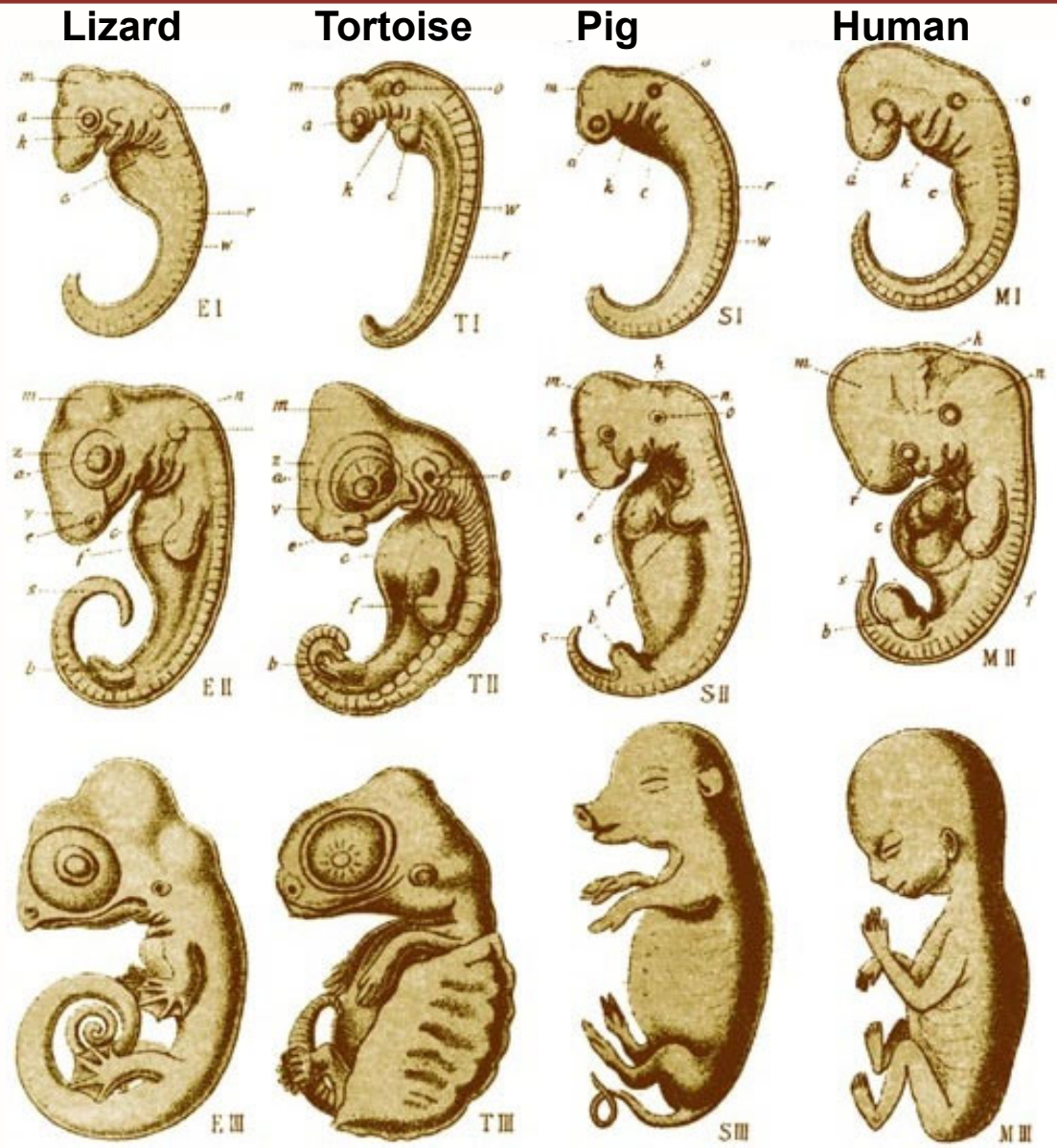
Very late cynodont (195 mya)



Key to skull bones

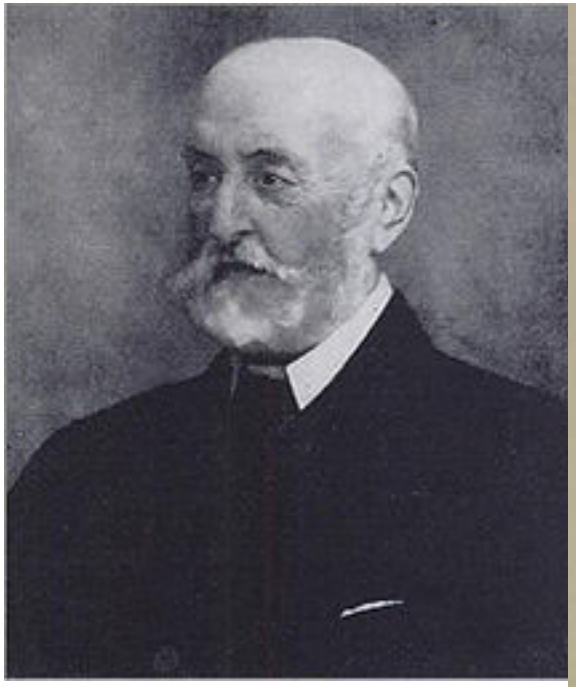
- Articular
- Quadrate
- Dentary
- Squamosal

Comparative Embryology

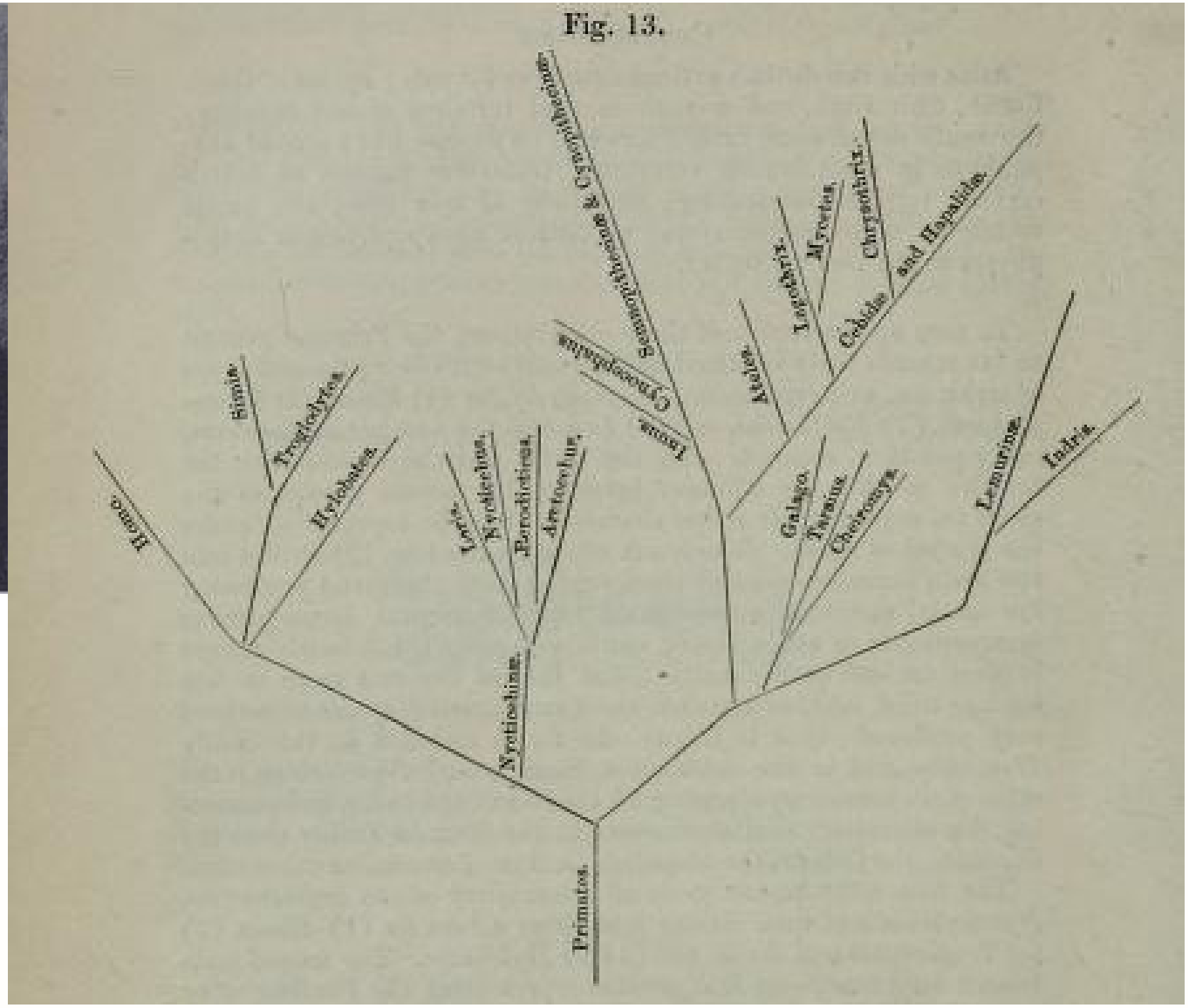


http://www.nature.com/nrg/journal/v7/n11/images/nrg1918-f2.jpg

The First Published Phylogeny



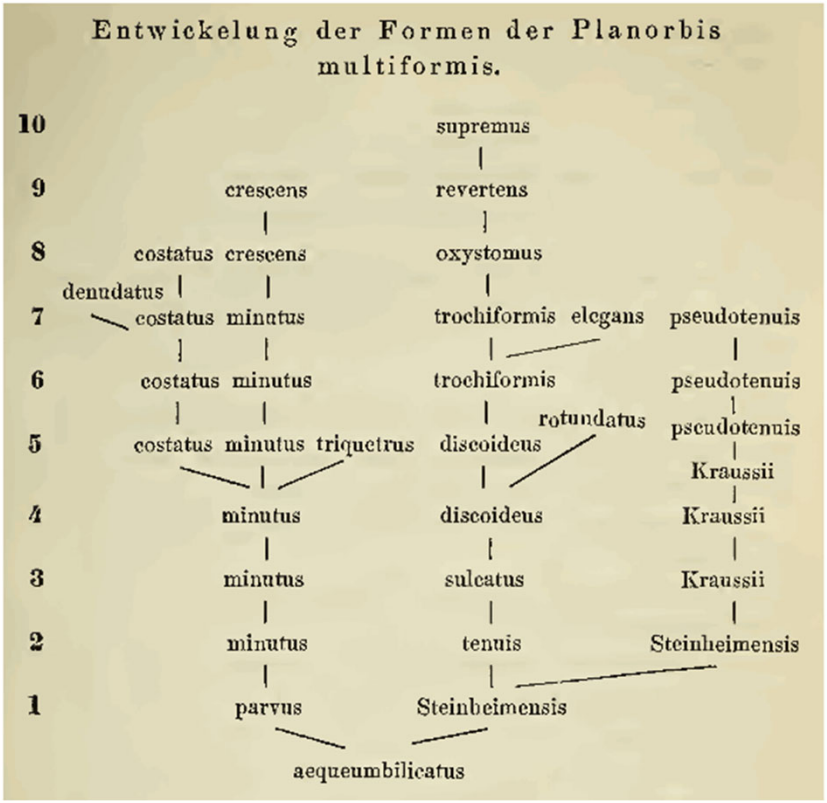
St. George Jackson Mivart



Mivart (1865) Proc. Zool. Soc. London

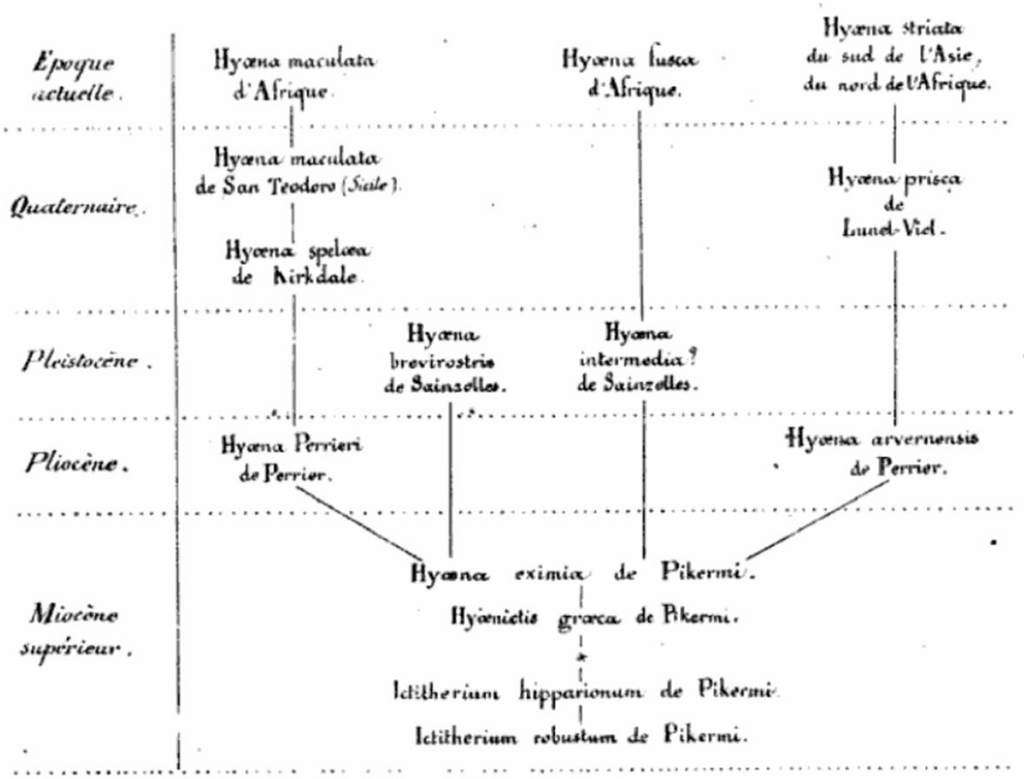
Inferring Phylogenies Becomes a Cottage Industry

Fossil gastropods



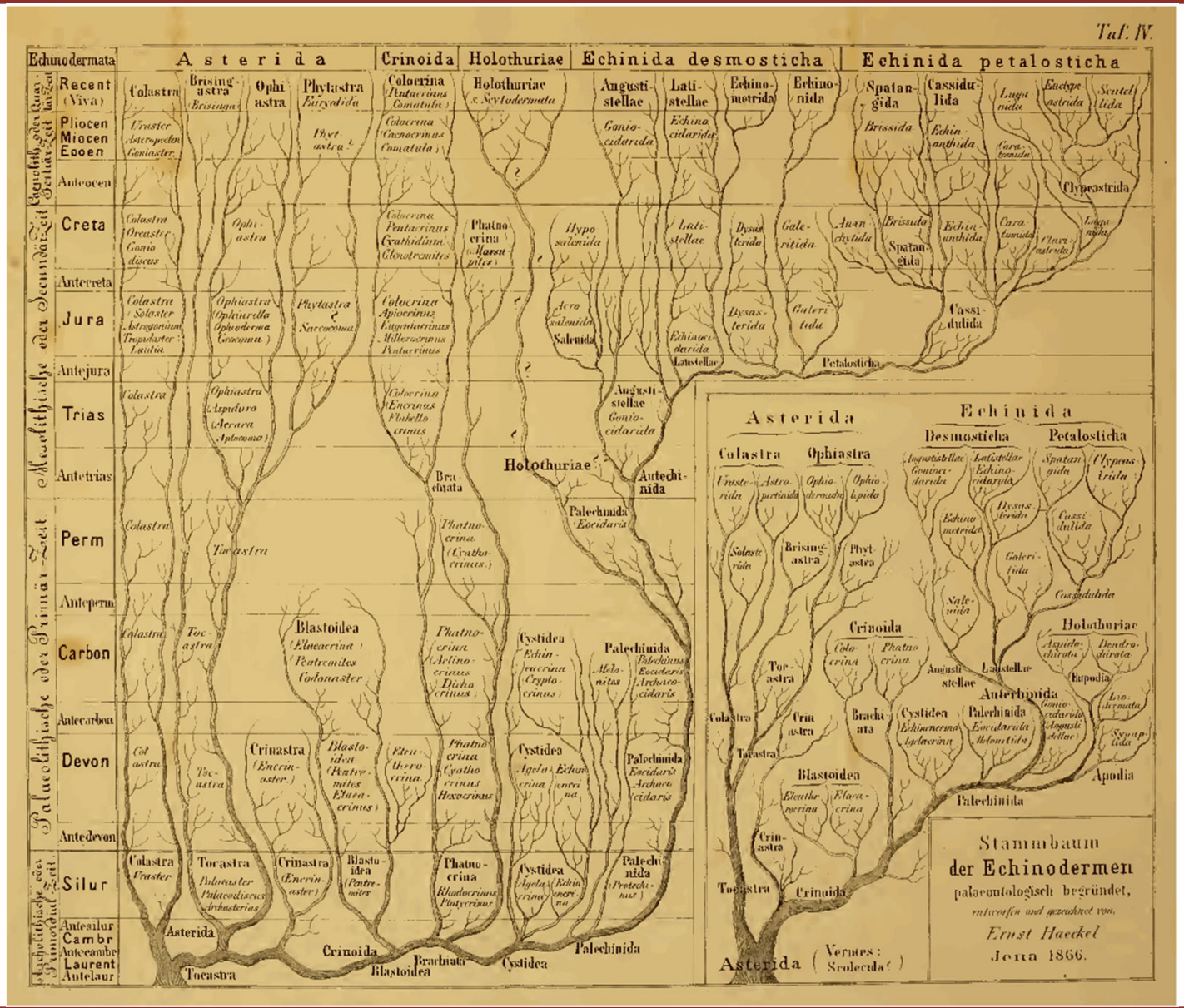
Hilgendorf, 1867

Extant and extinct mammals



Gaudry, 1866

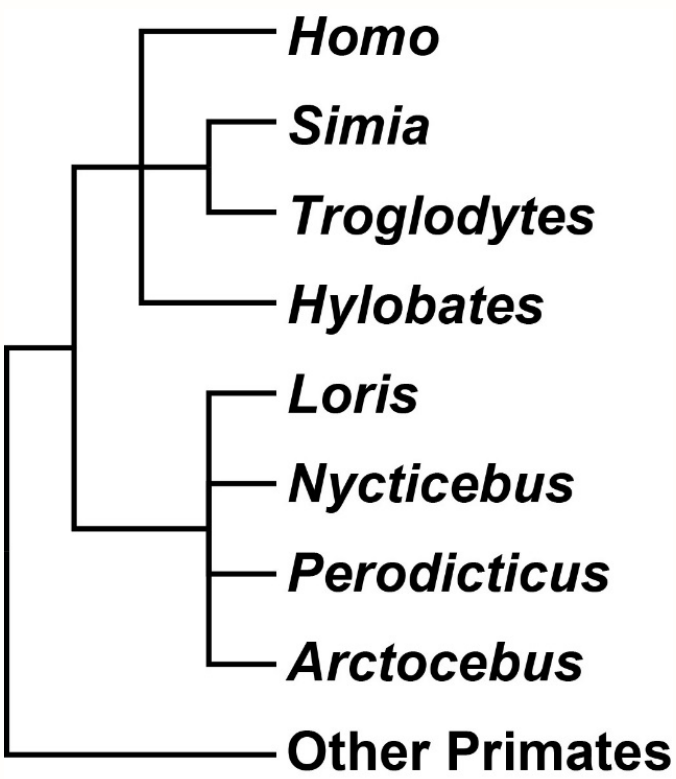
Haeckel's Phylogenies



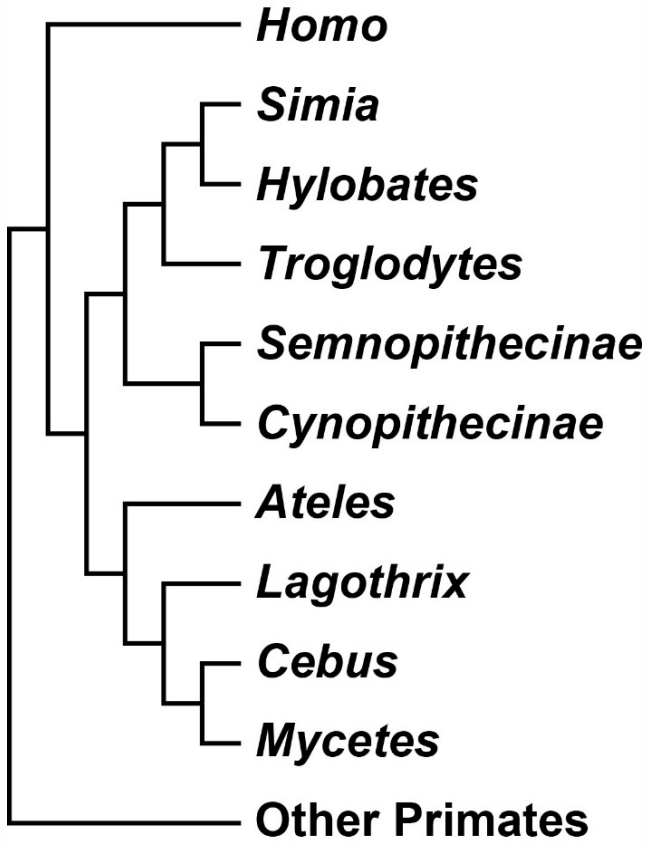
Haeckel (1866)

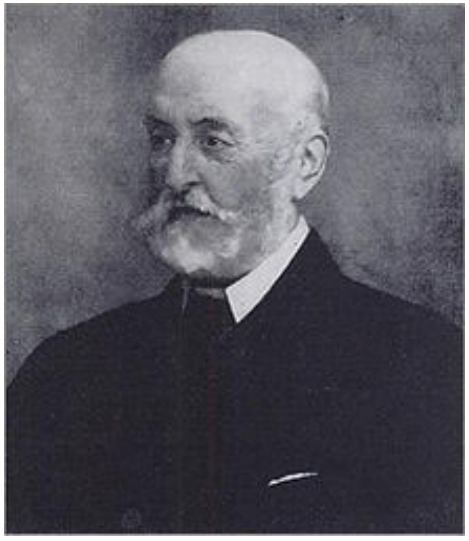
Disagreement Between Phylogenies

1865: SPINAL COLUMN



1867: LIMBS





St. George Jackson Mivart

In some M.S. [… I say] that on genealogical principles alone, & considering whole organisation man probably diverged from the Catarhine stem a little below the branch of the anthropo:apes […]. I have then added in my M.S. that this is your opinion […]. Is this your opinion?

I have really expressed no opinion as to Man's origin nor am I prepared to do so at this moment. The [1865] diagram […] expresses what I believe to be the degree of resemblance as regards the spinal column *only*. The [1867] diagram expresses what I believe to be the degree of resemblance as regards the appendicular skeleton *only*

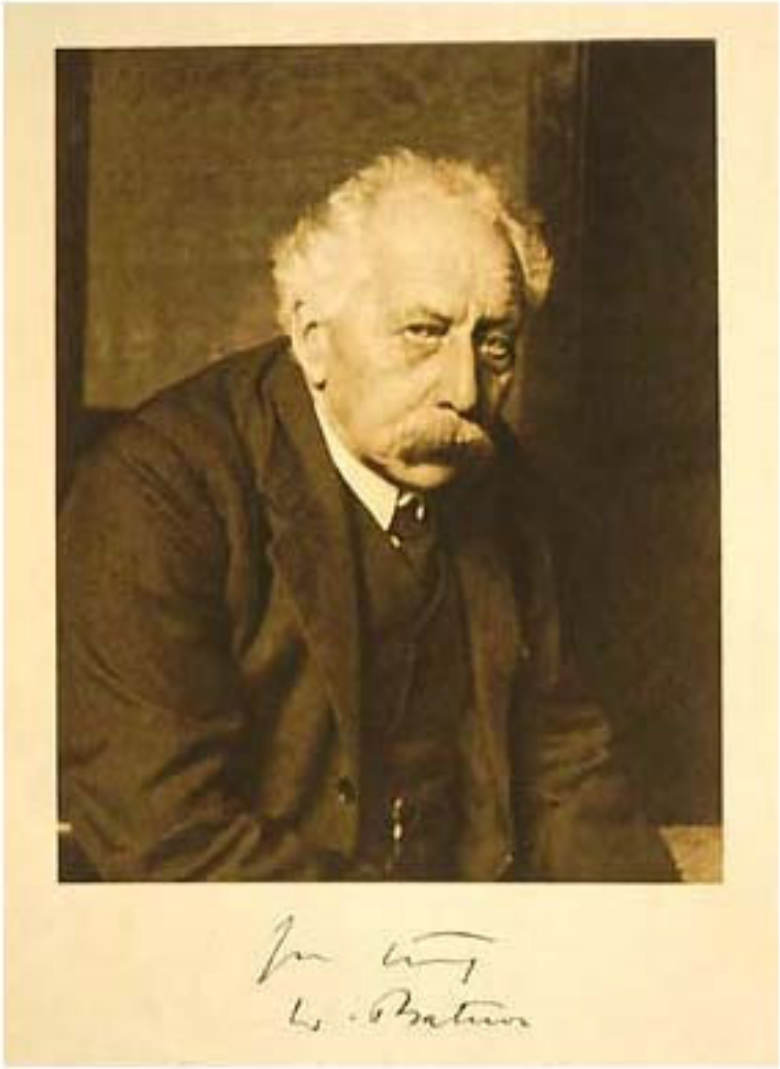
Comparative Morphology & Embryology in Trouble

By the turn of the century, the conflicting phylogenies produced by comparative morphology and embryology data have given rise to a sense of despair among the community

"From the same facts, opposite conclusions are drawn; facts of the same kind will take us no further. Need we waste more effort in these vain and sophistical disputes"

William Bateson (1894)

Materials for the Study of Variation



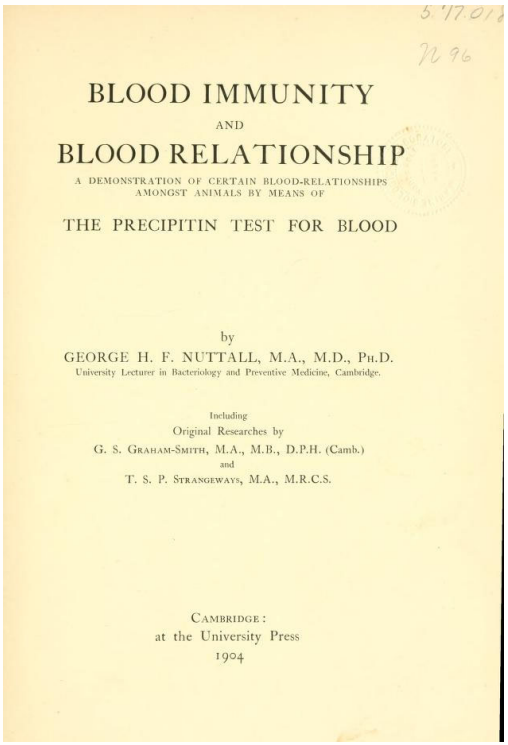
Courtesy of American Philosophical Society, Curt Stern Papers. Noncommercial, educational use only.

https://www.dnalc.org/view/16197-Gallery-5-William-Bateson-Portrait.html

The Origins of "Molecular" Phylogenetics

Studies in immunochemistry were showing that serological cross-reactions were stronger for more closely related organisms

Nuttall, realizing the evolutionary implications, used this approach to reconstruct the phylogenetic relationships among various groups of animals



BLOOD IMMUNITY
AND
BLOOD RELATIONSHIP
A DEMONSTRATION OF CERTAIN BLOOD-RELATIONSHIPS AMONGST ANIMALS BY MEANS OF
THE PRECIPITIN TEST FOR BLOOD

by
GEORGE H. F. NUTTALL, M.A., M.D., PH.D.
University Lecturer in Bacteriology and Preventive Medicine, Cambridge.

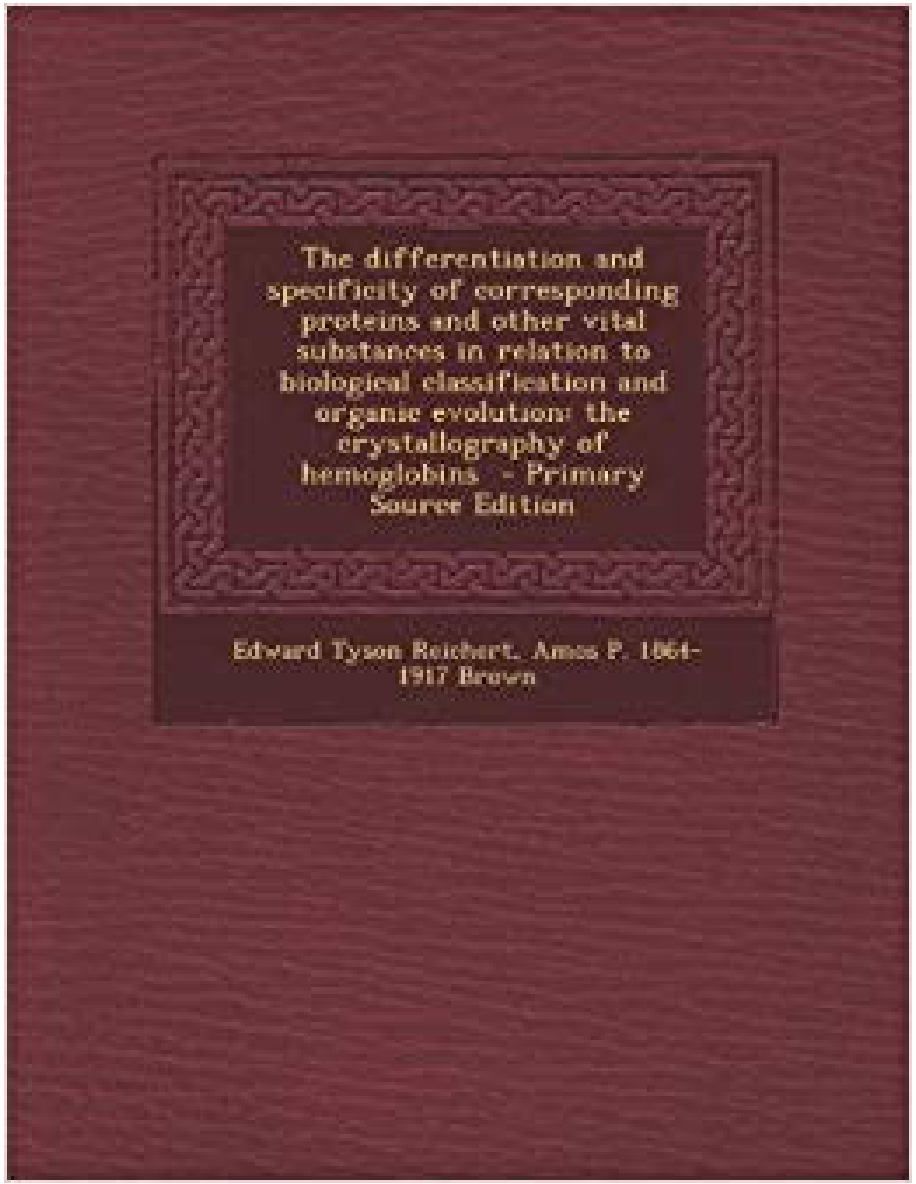
Including
Original Researches by
G. S. GRAHAM-SMITH, M.A., M.B., D.P.H. (Camb.)
and
T. S. P. STRANGEWAYS, M.A., M.R.C.S.

CAMBRIDGE:
at the University Press
1904

He determined that humans' closest relatives were the apes, followed, in order of relatedness, by Old World monkeys, New World monkeys, and prosimians (lemurs and tarsiers)!

The First Large-Scale Molecular Investigation of Species Differences

- Different crystals of hemoglobin from the same species differed in size & shape, but angles between faces constant
- But interfacial angles differed from species to species -> similarities in angle values were consistent with taxonomy-based phylogeny
- 600 photomicrographs of crystals of hemoglobin from >100 species
- All this before discoveries of X-ray diffraction / protein sequencing / DNA & DNA sequencing



Reichert & Brown (1909) The Crystallography of Hemoglobins

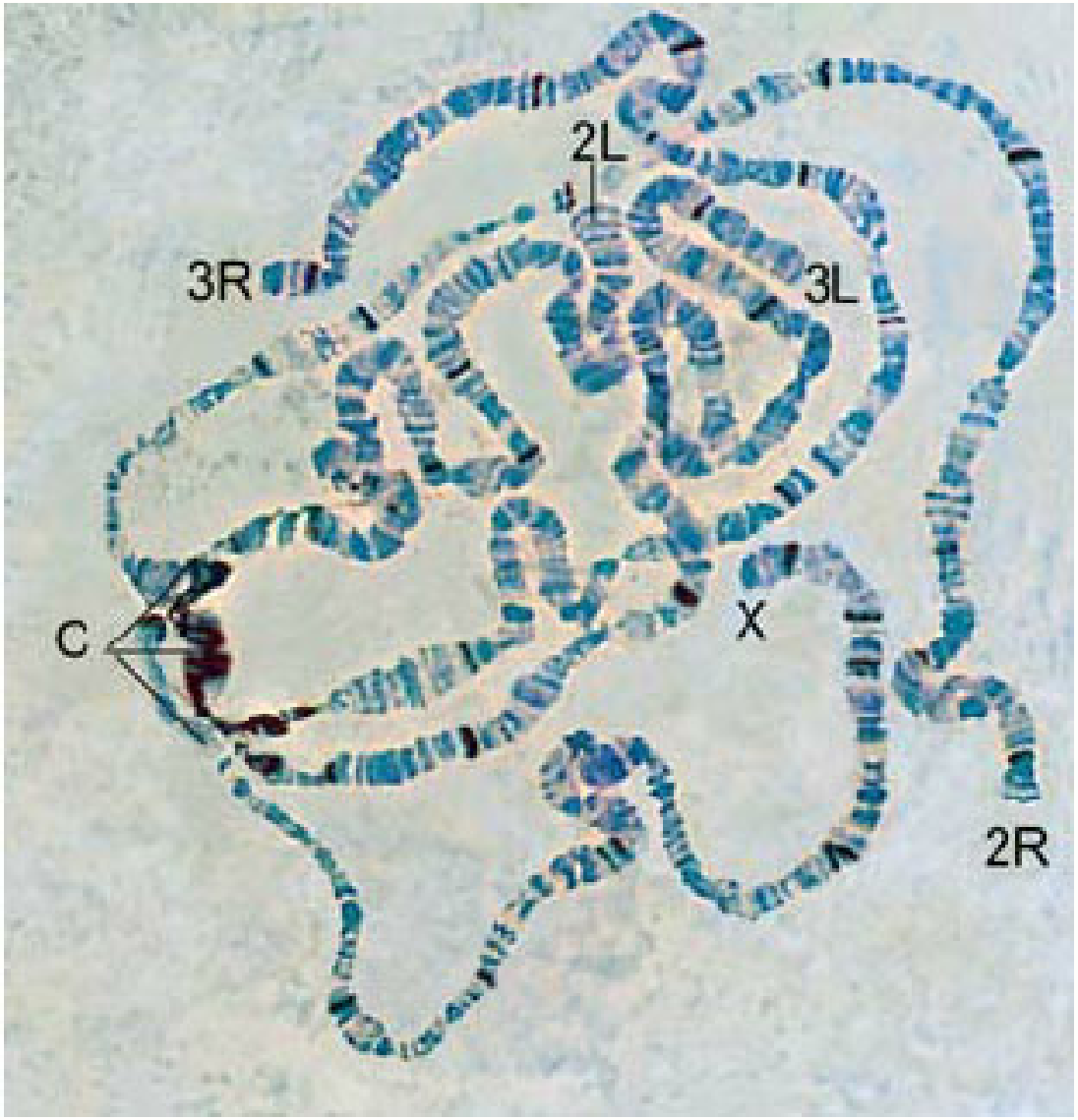
"Molecular" Phylogenetics of Drosophila

Theodosius Dobzhansky

Alfred Sturtevant

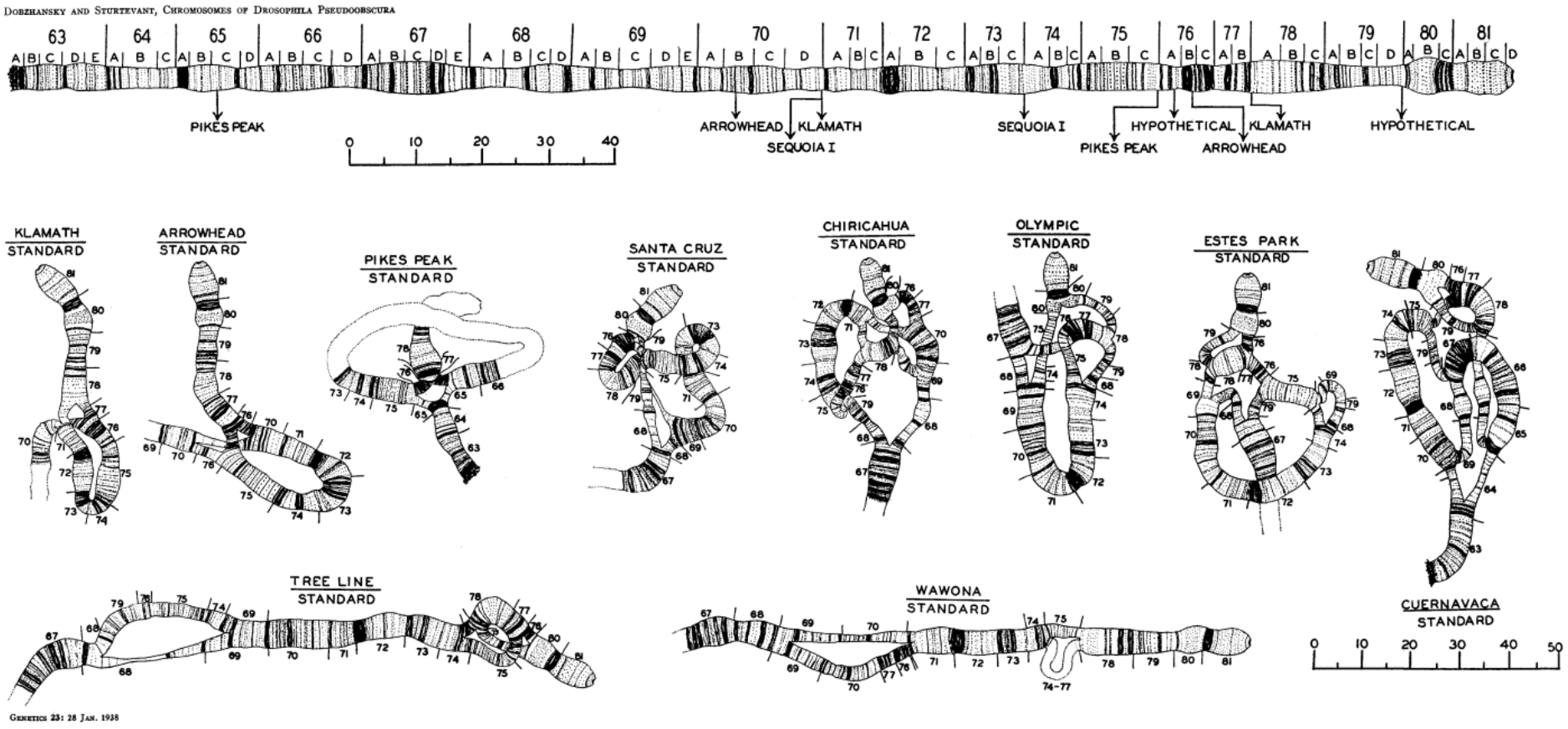
https://www-tc.pbs.org/wgbh/evolution/library/06/2/images/l_062_04_l.jpg;
http://www.caltech.edu/news/first-genetic-linkage-map-38798

Polytene Chromosomes



Viera et al. (2009) Transposons and the Dynamic Genome

Using Chromosomal Rearrangements as Markers…



Dobzhansky & Sturtevant (1938) Genetics

...To Infer the History of Species

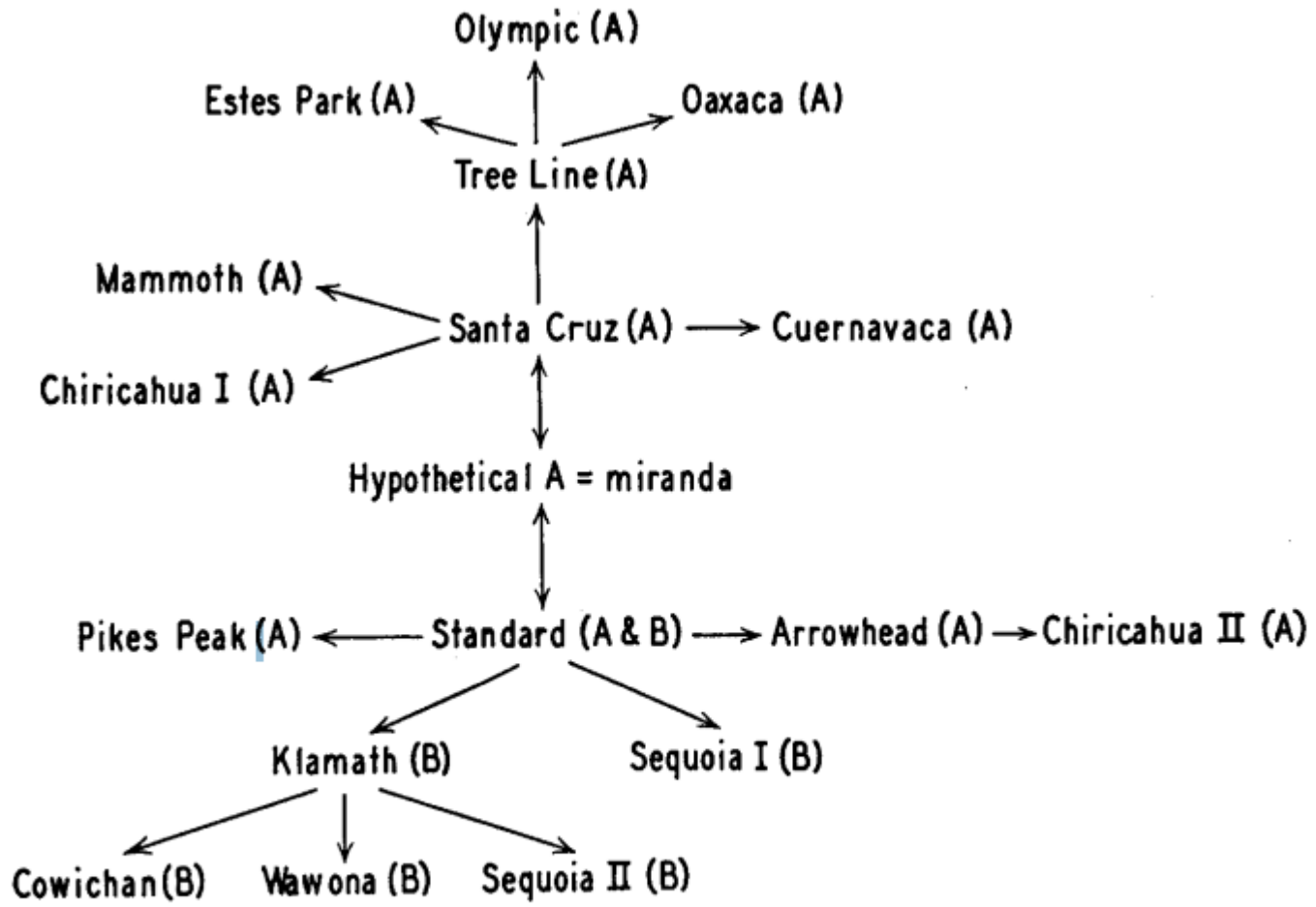
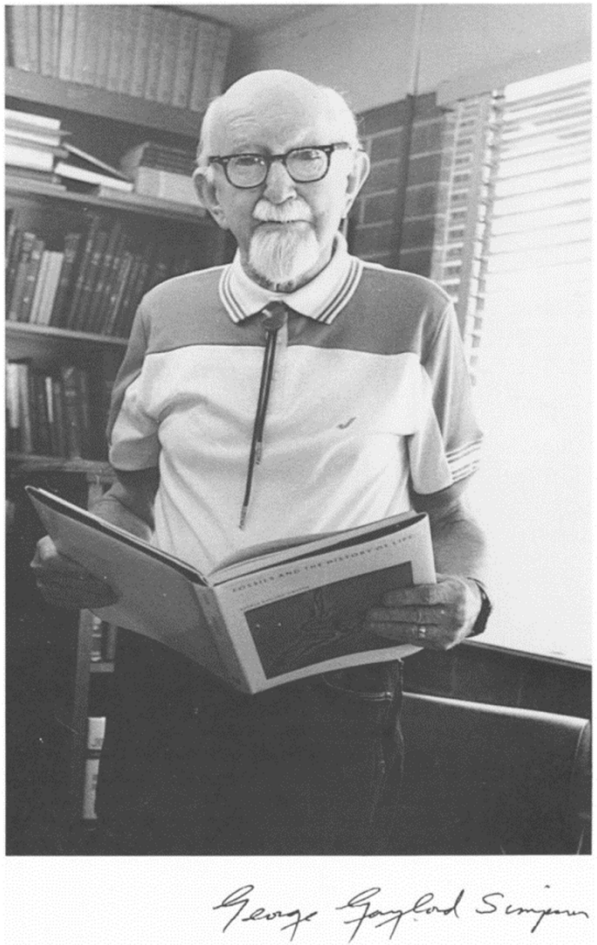
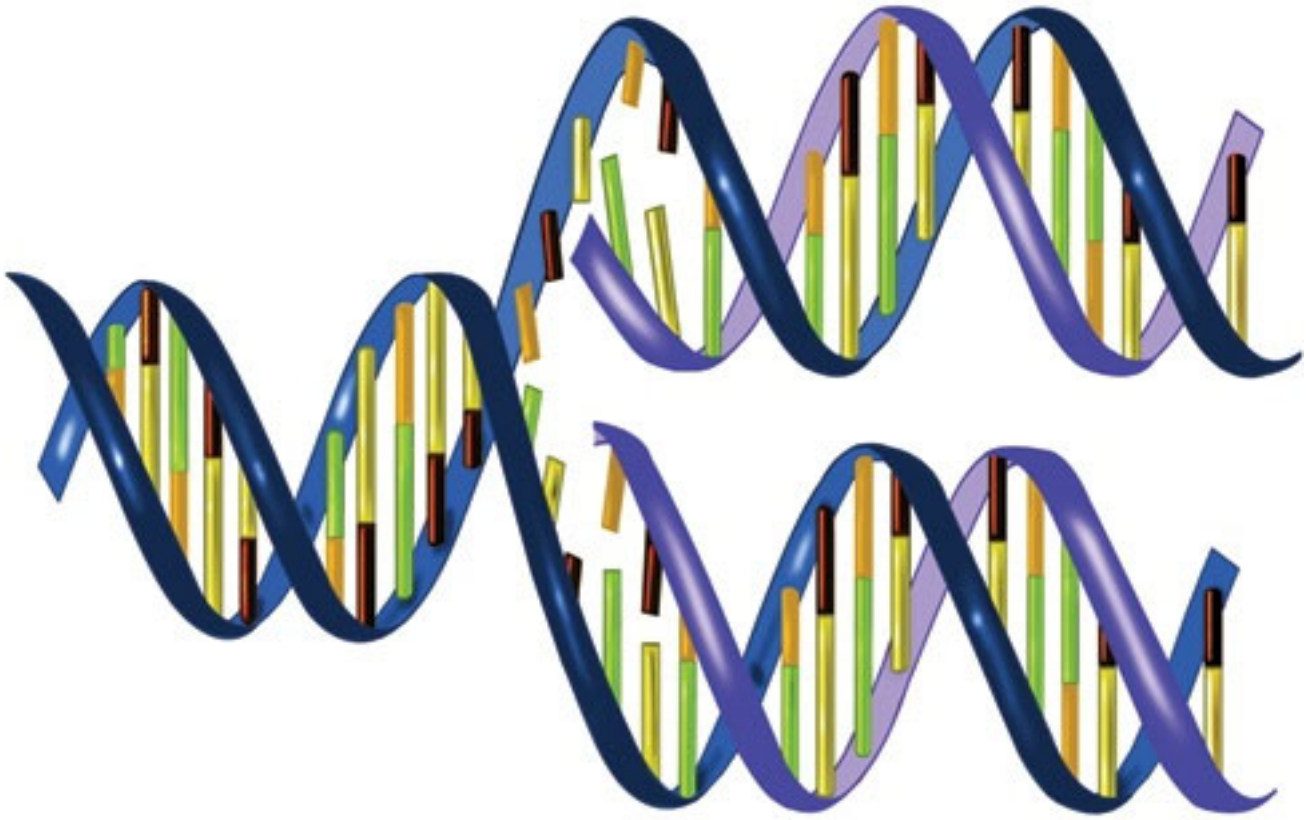


FIGURE 3.—Phylogeny of the gene arrangements in the third chromosome of *Drosophila pseudoobscura*. Any two arrangements connected by an arrow in the diagram differ by a single inversion. Further explanation in text.

Dobzhansky & Sturtevant (1938) Genetics



George Gaylord Simpson



“The stream of heredity makes phylogeny; in a sense, it is phylogeny. Complete genetic analysis would provide the most priceless data for the mapping of this stream”

G. G. Simpson, 1945

Early '50s: Discovery of Protein Sequencing

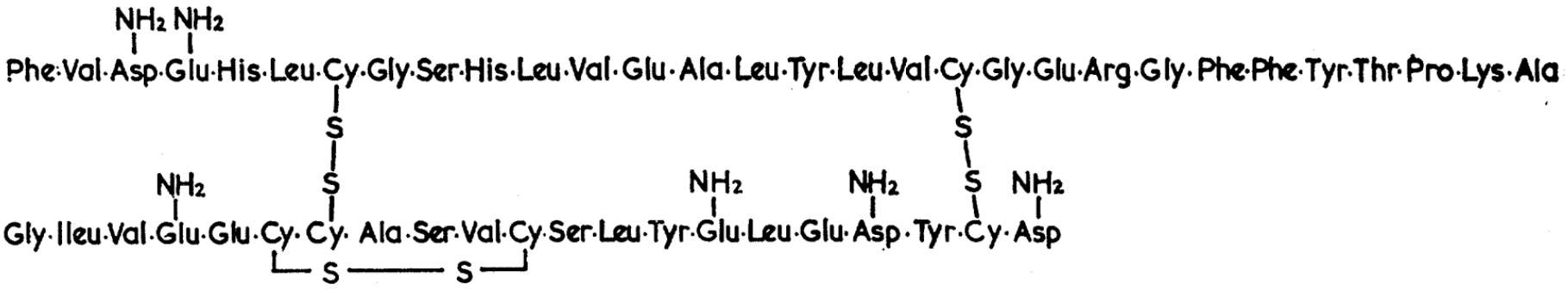
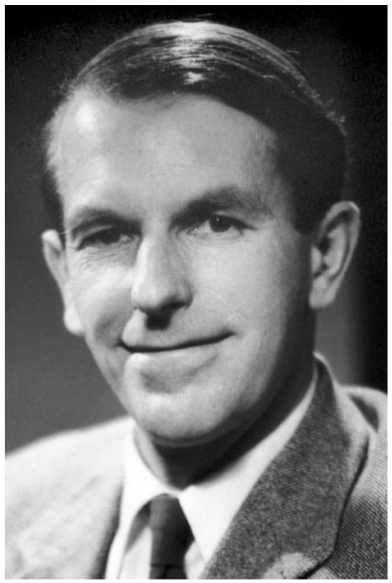


Fig. 2. Structure of insulin.



Frederick Sanger
The Nobel Prize in Chemistry 1958

Born: 13 August 1918, Rendcombe, United Kingdom

Died: 19 November 2013, Cambridge, United Kingdom

Affiliation at the time of the award: University of Cambridge, Cambridge, United Kingdom

Prize motivation: "for his work on the structure of proteins, especially that of insulin."

Prize share: 1/1

"...before long we shall have a subject which might be called "protein taxonomy"; the study of amino acid sequences of the proteins of an organism and the comparison of them between species. It can be argued that these sequences are the most delicate expression possible of the phenotype of an organism and that vast amounts of evolutionary information may be hidden away within them"

Francis Crick (1957) Nature

Elucidating the Sequence of Proteins

NATURE March 4, 1961 VOL. 189

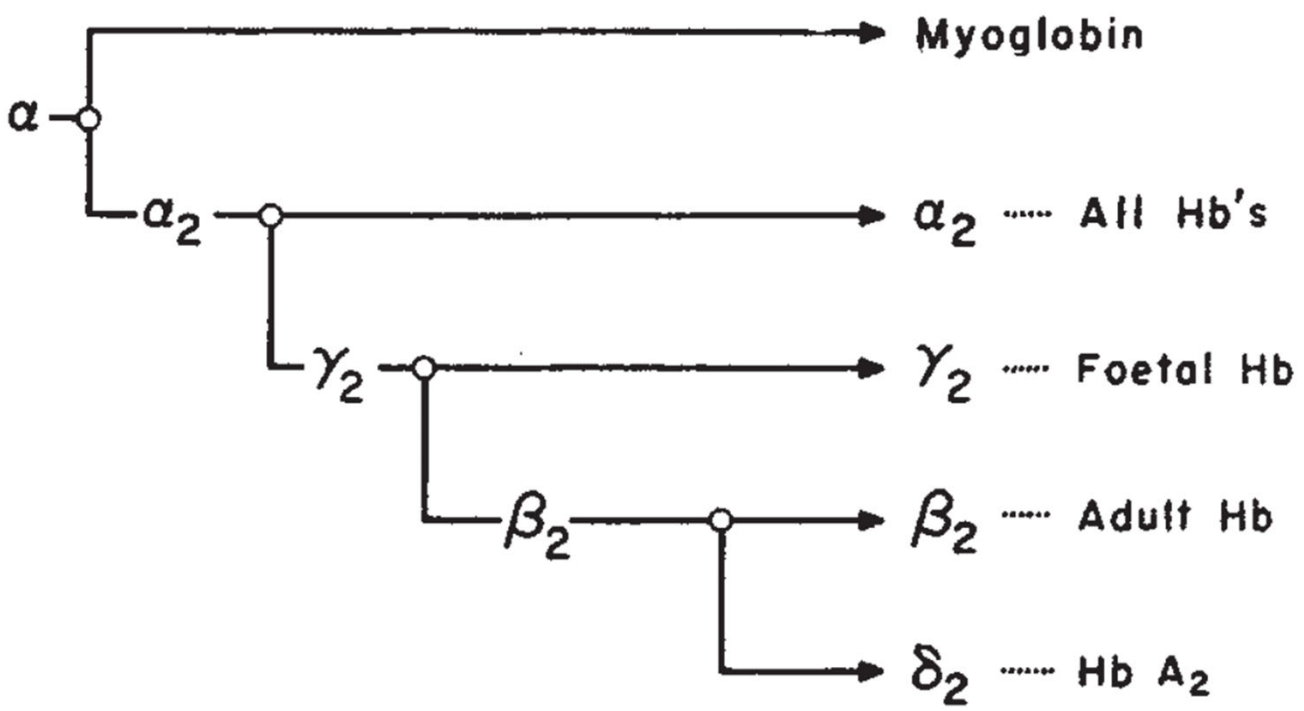
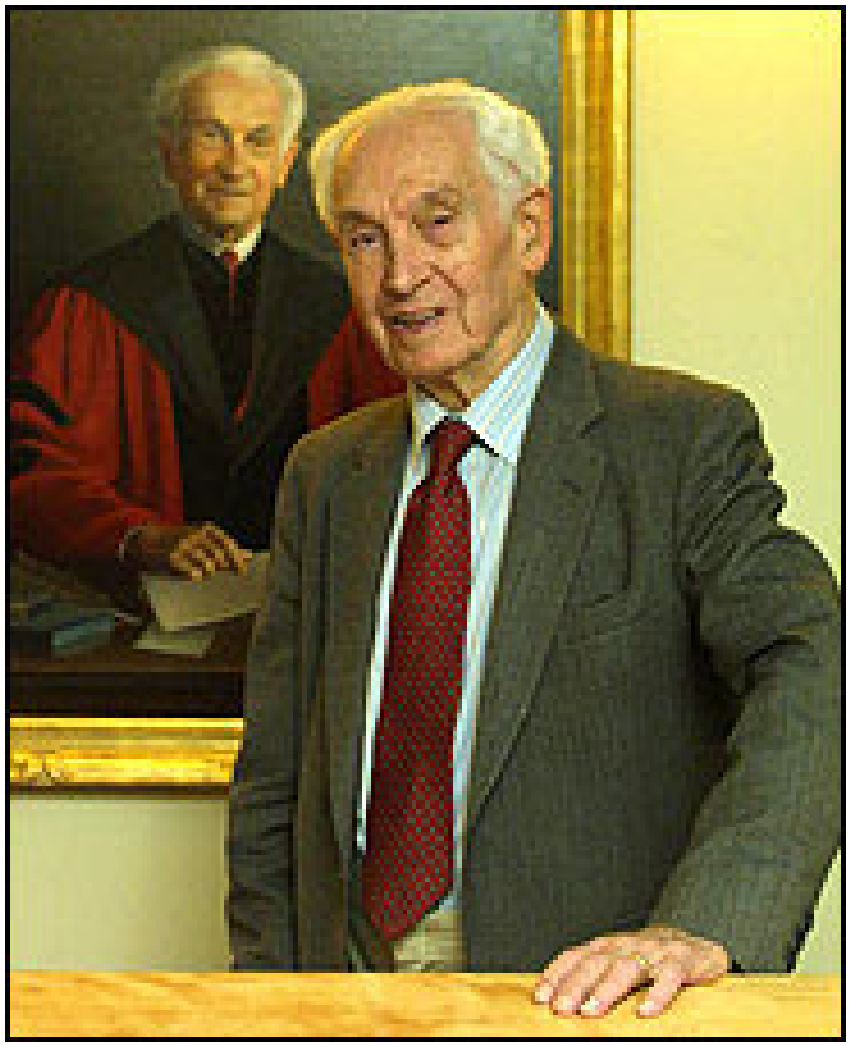


Fig. 1. Evolution of the hæmoglobin chains. The α-chain is the ancestral peptide chain. —○— indicates a point of gene duplication followed by translocation of the new gene

Ingram (1961) Nature

“…the search for homologous genes is quite futile except in very close relatives”

Ernst Mayr, 1963



DNA & Protein Sequences Record Evolutionary History

Molecules as Documents of Evolutionary History

EMILE ZUCKERKANDL AND LINUS PAULING

Gates and Crellin Laboratories of Chemistry,
California Institute of Technology, Pasadena, California, U.S.A.

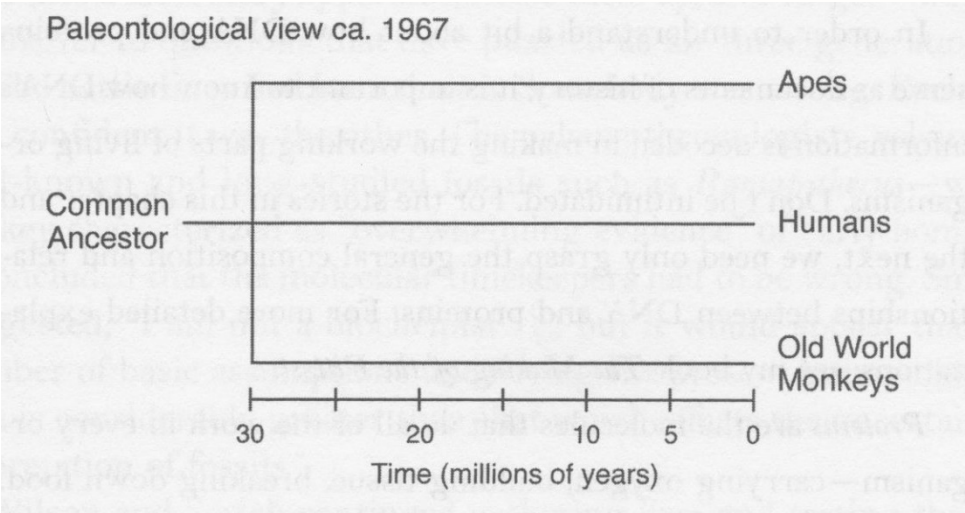
(*Received* 17 *September* 1964)

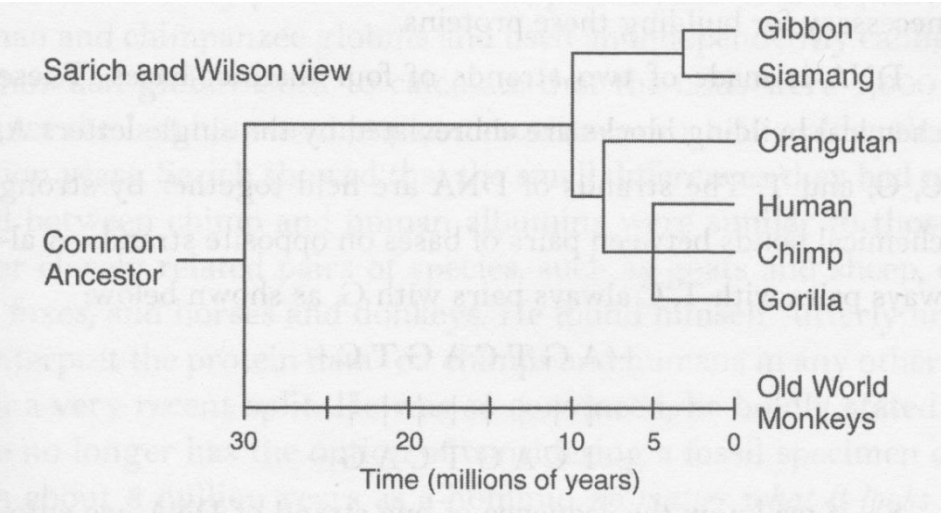
Different types of molecules are discussed in relation to their fitness for providing the basis for a molecular phylogeny. Best fit are the "semantides", i.e. the different types of macromolecules that carry the genetic information or a very extensive translation thereof. The fact that more than one coding triplet may code for a given amino acid residue in a polypeptide leads to the notion of "isosemantic substitutions" in genic and messenger polynucleotides. Such substitutions lead to differences in nucleotide sequence that are not expressed by differences in amino acid sequence. Some possible consequences of isosemanticism are discussed.

Zuckerkandl & Pauling (1965) J. Theoret. Biol.

Estimating the Divergence of Humans and Chimps

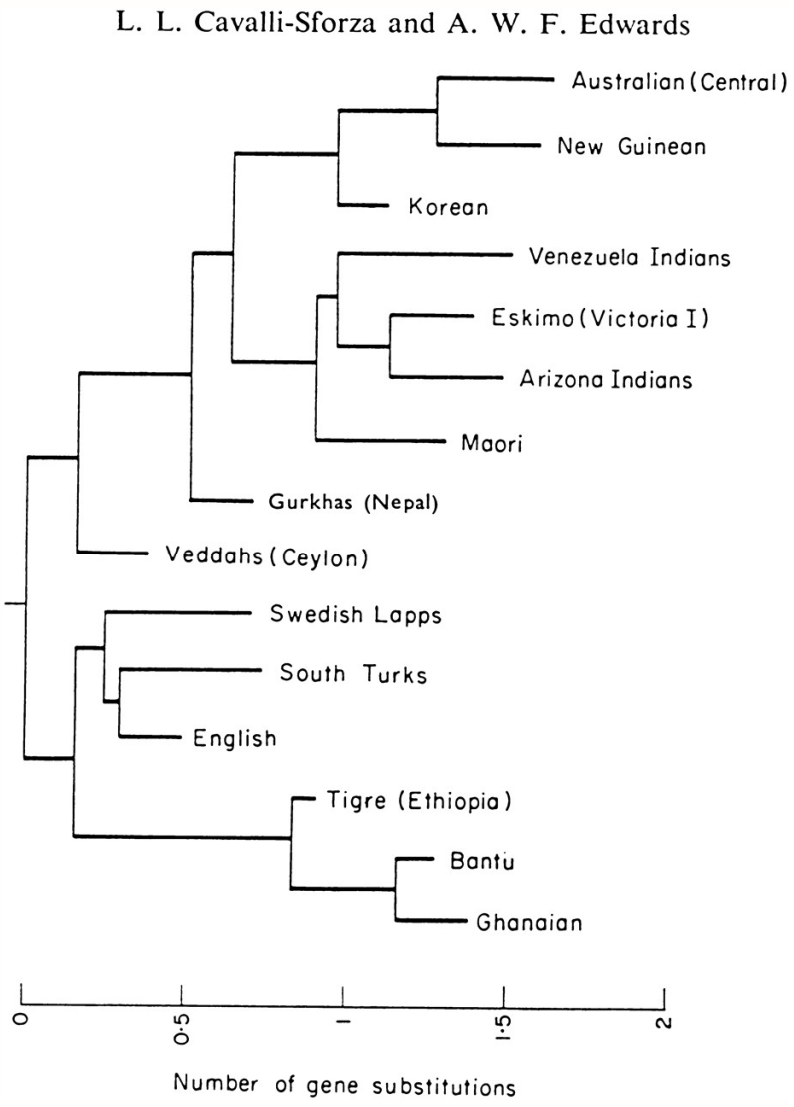
Divergence times were estimated by measuring the immunological cross-reaction of blood serum albumin between pairs of primates





"no fuss, no muss, no dishpan hands. Just throw some proteins into a laboratory apparatus, shake them up, and bingo! – we have an answer to questions that have puzzled us for three generations."

The Phylogeny of Human Populations

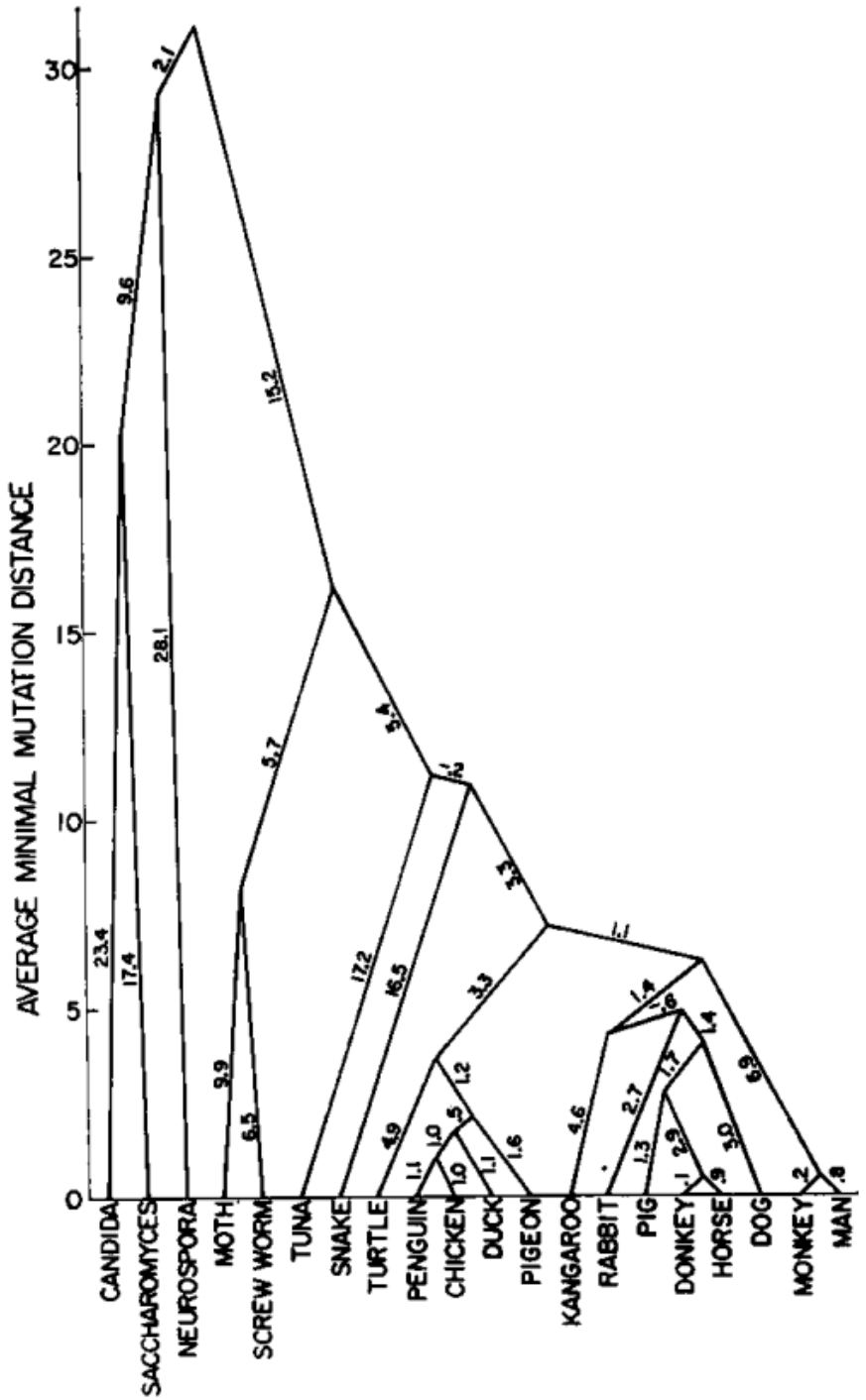


Phylogeny inferred from blood group allele frequencies from 15 populations

Sequence-based Phylogenies

Used protein sequences from a large number of organisms

Developed a computational approach for efficient analysis of large numbers of taxa (distance matrix method)



Fitch & Margoliash (1967) Science

Proc. Natl. Acad. Sci. USA
Vol. 74, No. 11, pp. 5088–5090, November 1977
Evolution

Phylogenetic structure of the prokaryotic domain: The primary kingdoms

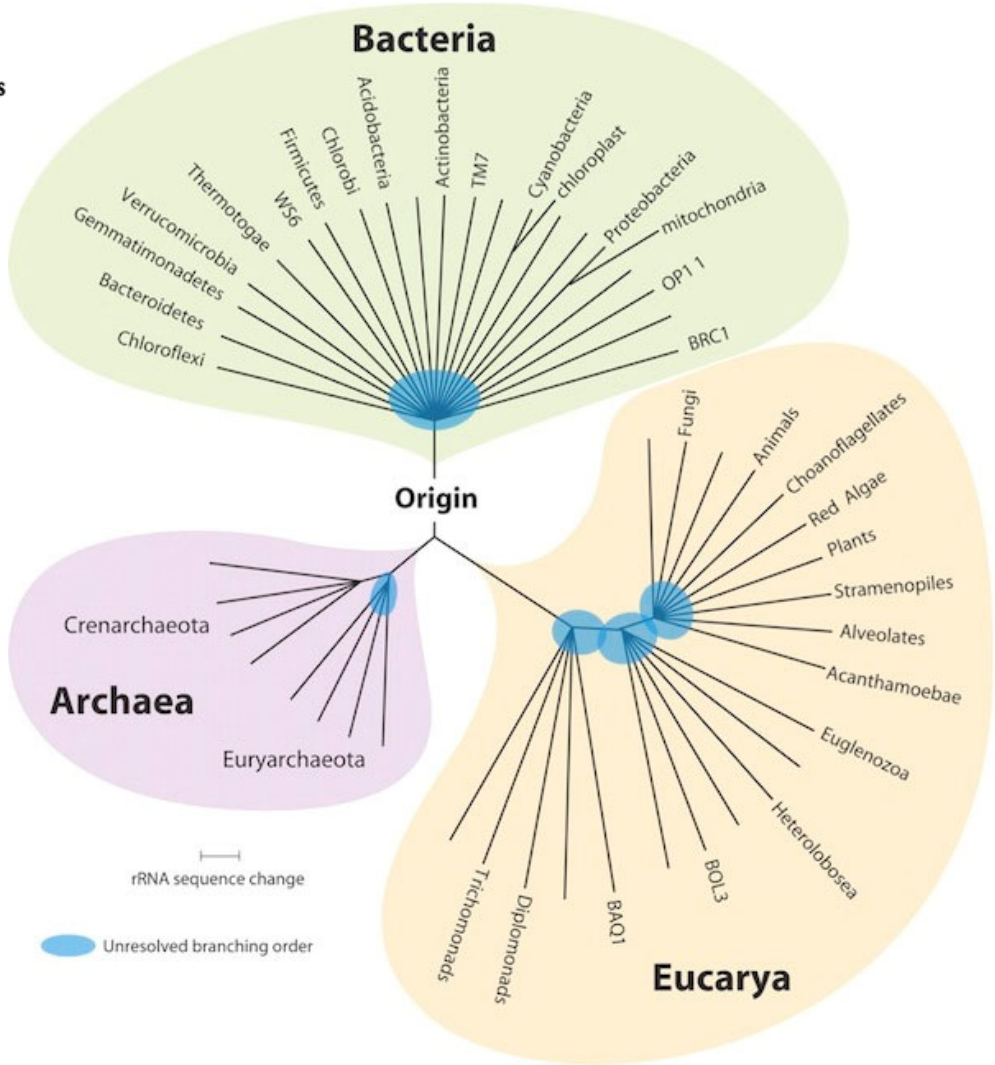
(archaebacteria/eubacteria/urkaryote/16S ribosomal RNA/molecular phylogeny)

CARL R. WOESE AND GEORGE E. FOX*

Department of Genetics and Development, University of Illinois, Urbana, Illinois

Communicated by T. M. Sonneborn, August 18, 1977

ABSTRACT **A phylogenetic analysis based upon ribosomal RNA sequence characterization reveals that living systems represent one of three aboriginal lines of descent: (*i*) the eubacteria, comprising all typical bacteria; (*ii*) the archaebacteria, containing methanogenic bacteria; and (*iii*) the urkaryotes, now represented in the cytoplasmic component of eukaryotic cells.**



~~Gene tree ≈ Species phylogeny~~

Gene tree ≠ Species phylogeny

Two Types of Factors Influence the Relationship

Analytical factors

They lead to failure in accurately inferring a gene tree; these can be either due to **stochastic error** (e.g., insufficient sequence length or taxon samples) or due to **systematic error** (e.g., observed data far depart from model assumptions)

Biological factors

They lead to gene trees whose histories may differ from each other and from the species tree. Known factors include **stochastic lineage sorting**, **hidden paralogy**, **horizontal gene transfer**, **recombination** and **natural selection**

Sampling Error

```
10  50
Cow         MAYPMQLGFQ DATSPIMEEL LHFHDHTLMI VFLISSLVLY IISLMLTTKL
Carp        MAHPTQLGFK DAAMPVMEEL LHFHDHALMI VLLISTLVLY IITAMVSTKL
Chicken     MANHSQLGFQ DASSPIMEEL VEFHDHALMV ALAICSLVLY LLTLMLMEKL
Human       MAHAAQVGLQ DATSPIMEEL ITFHDHALMI IFLICFLVLY ALFLTLTTKL
Loach       MAHPTQLGFQ DAASPVMEEL LHFHDHALMI VFLISALVLY VIITTVSTKL
Mouse       MAYPFQLGLQ DATSPIMEEL MNFHDHTLMI VFLISSLVLY IISLMLTTKL
Rat         MAYPFQLGLQ DATSPIMEEL TNFHDHTLMI VFLISSLVLY IISLMLTTKL
Seal        MAYPLQMGLQ DATSPIMEEL LHFHDHTLMI VFLISSLVLY IISLMLTTKL
Whale       MAYPFQLGFQ DAASPIMEEL LHFHDHTLMI VFLISSLVLY IITLMLTTKL
Frog        MAHPSQLGFQ DAASPIMEEL LHFHDHTLMA VFLISTLVLY IITIMMTTKL
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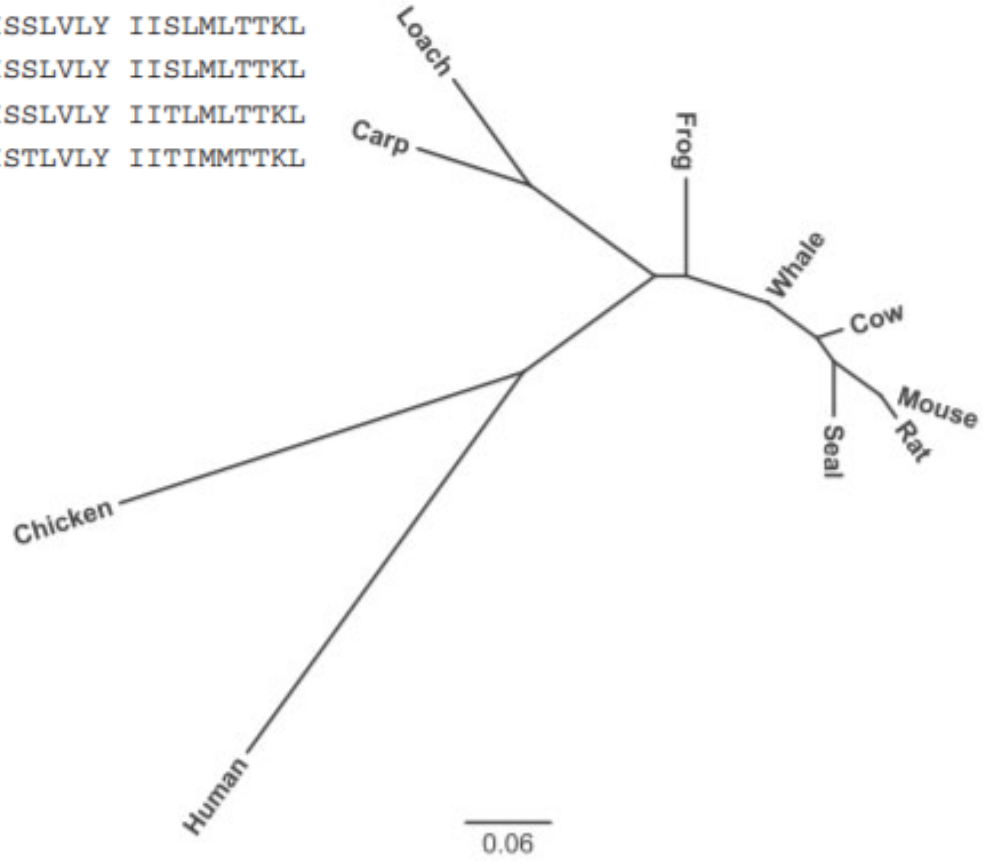
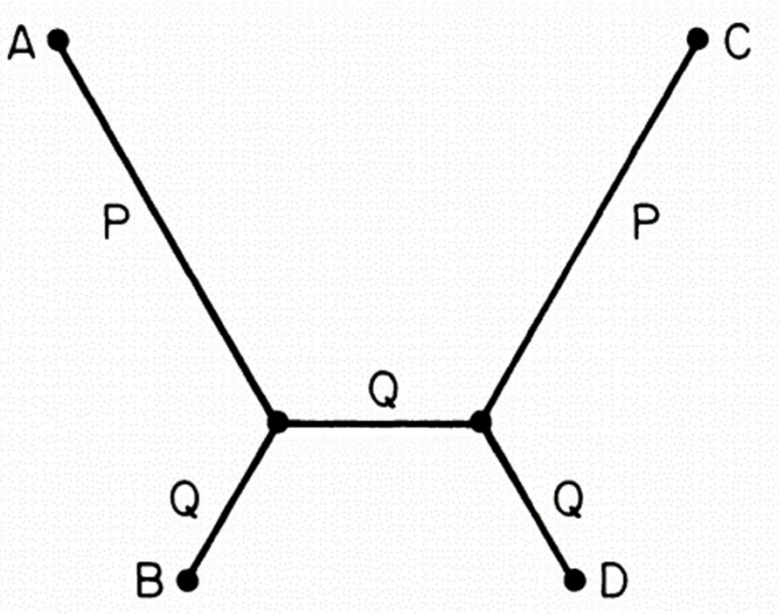
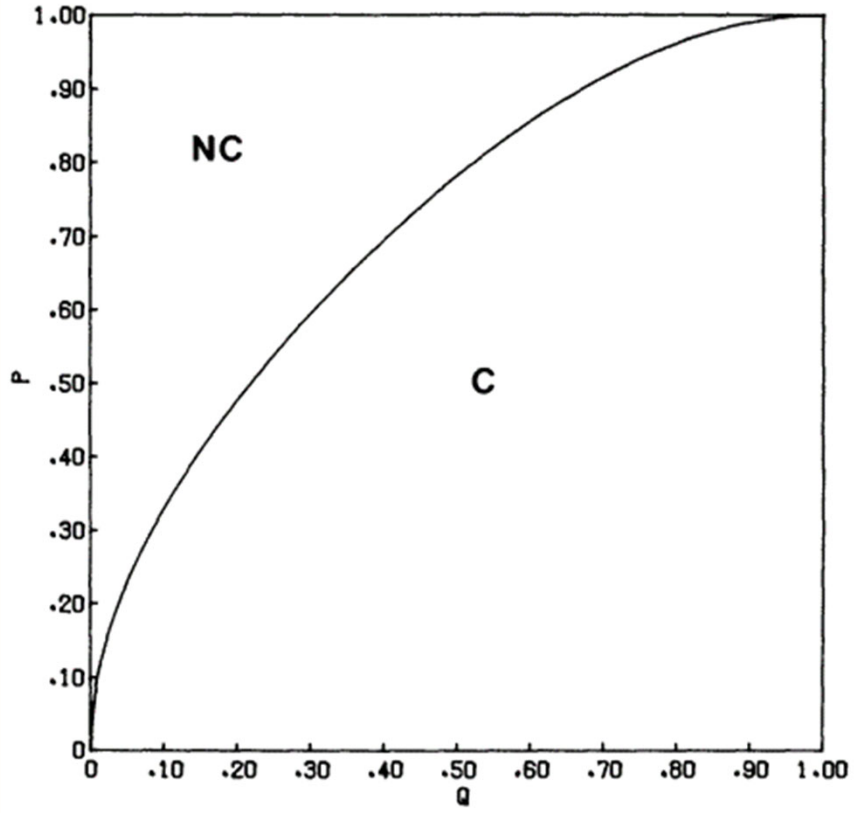


Systematic Error

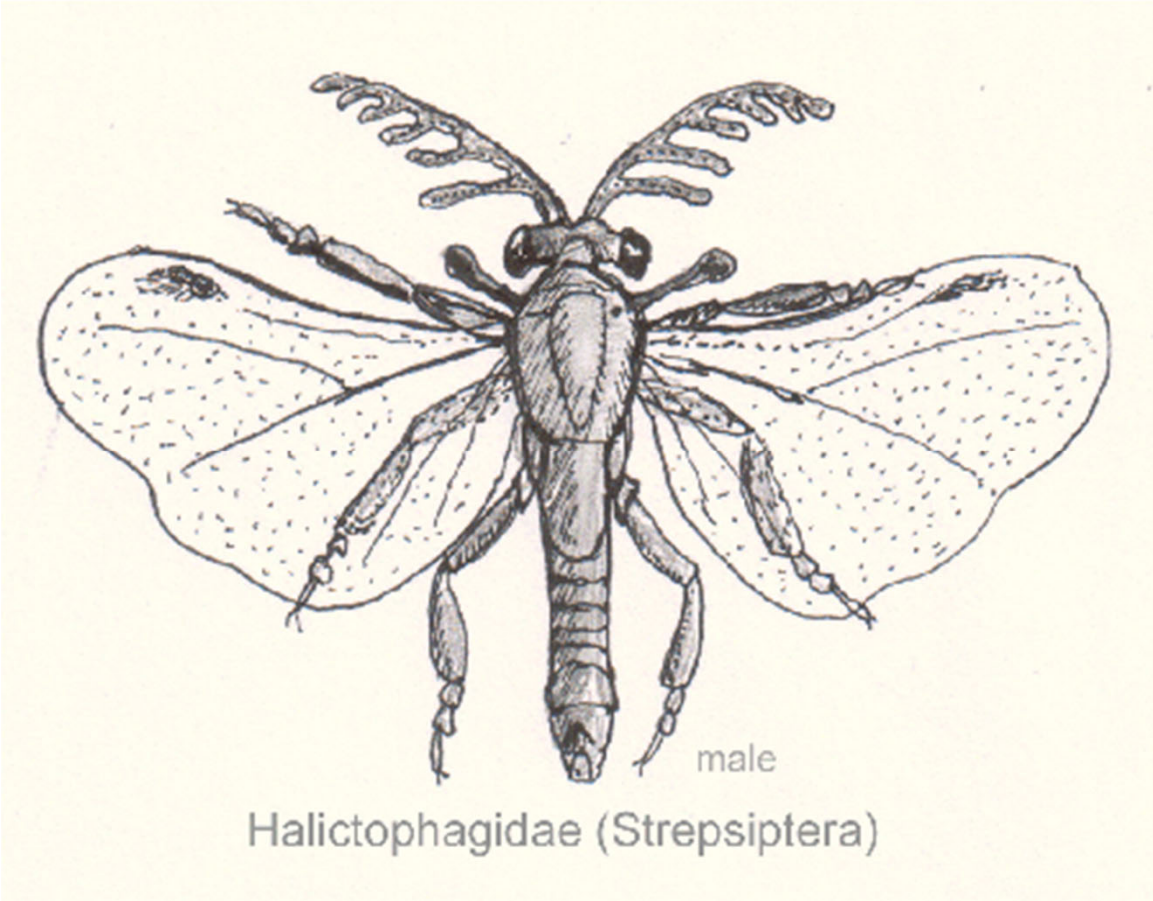




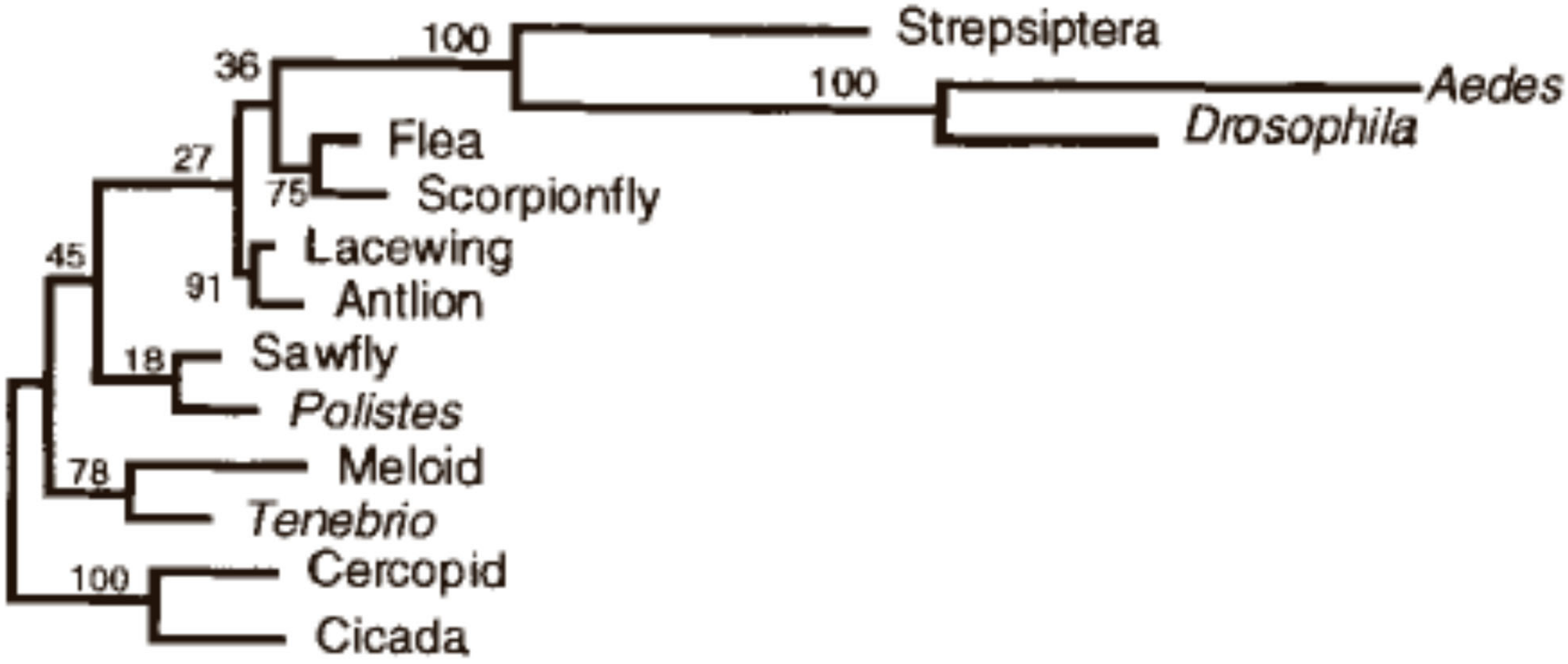
Long branch attraction

Felsenstein (1978) Syst. Zool.

"The Strepsiptera Problem" is a Classic Example of LBA

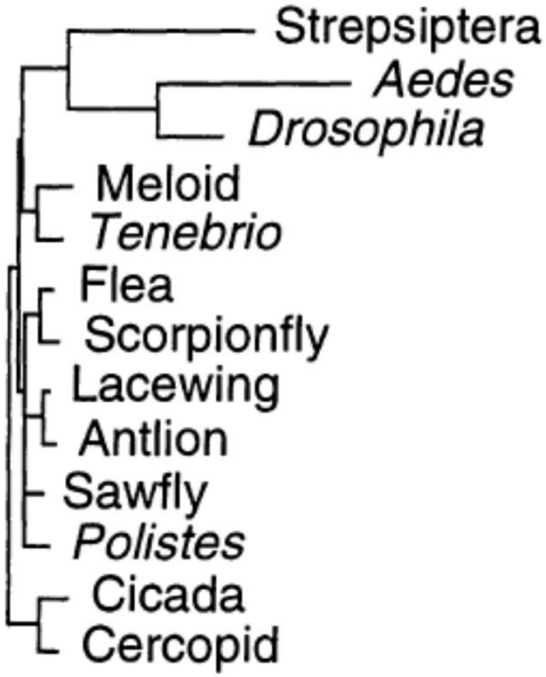


The Strepsiptera Problem

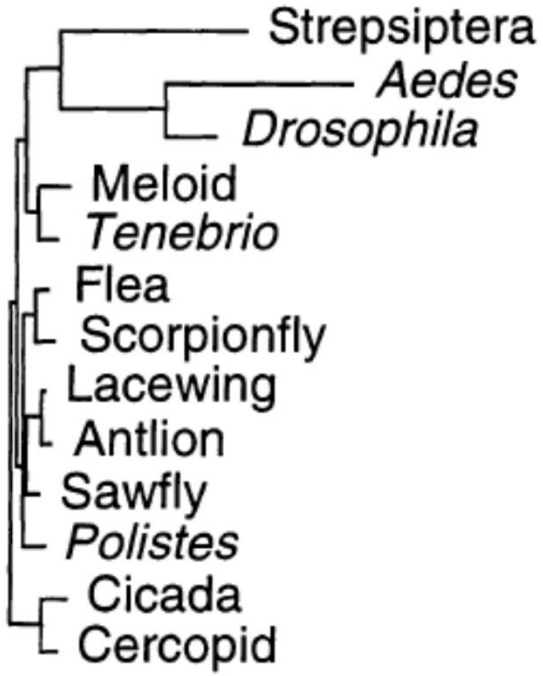


Carmean & Crespi (1995) Nature

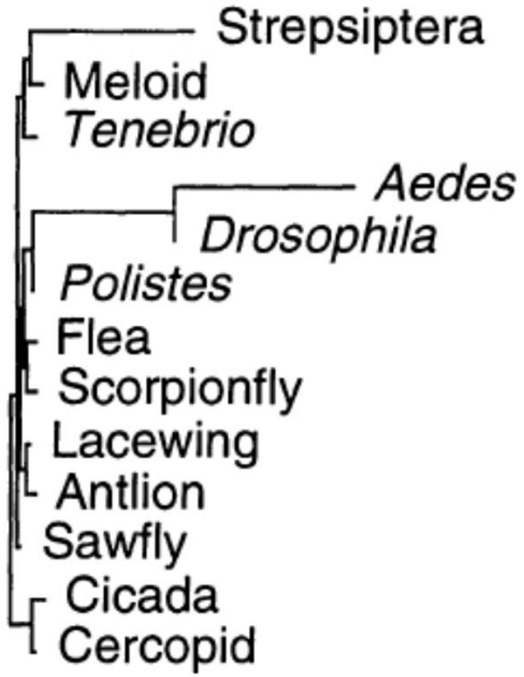
The Strepsiptera Problem



p distance

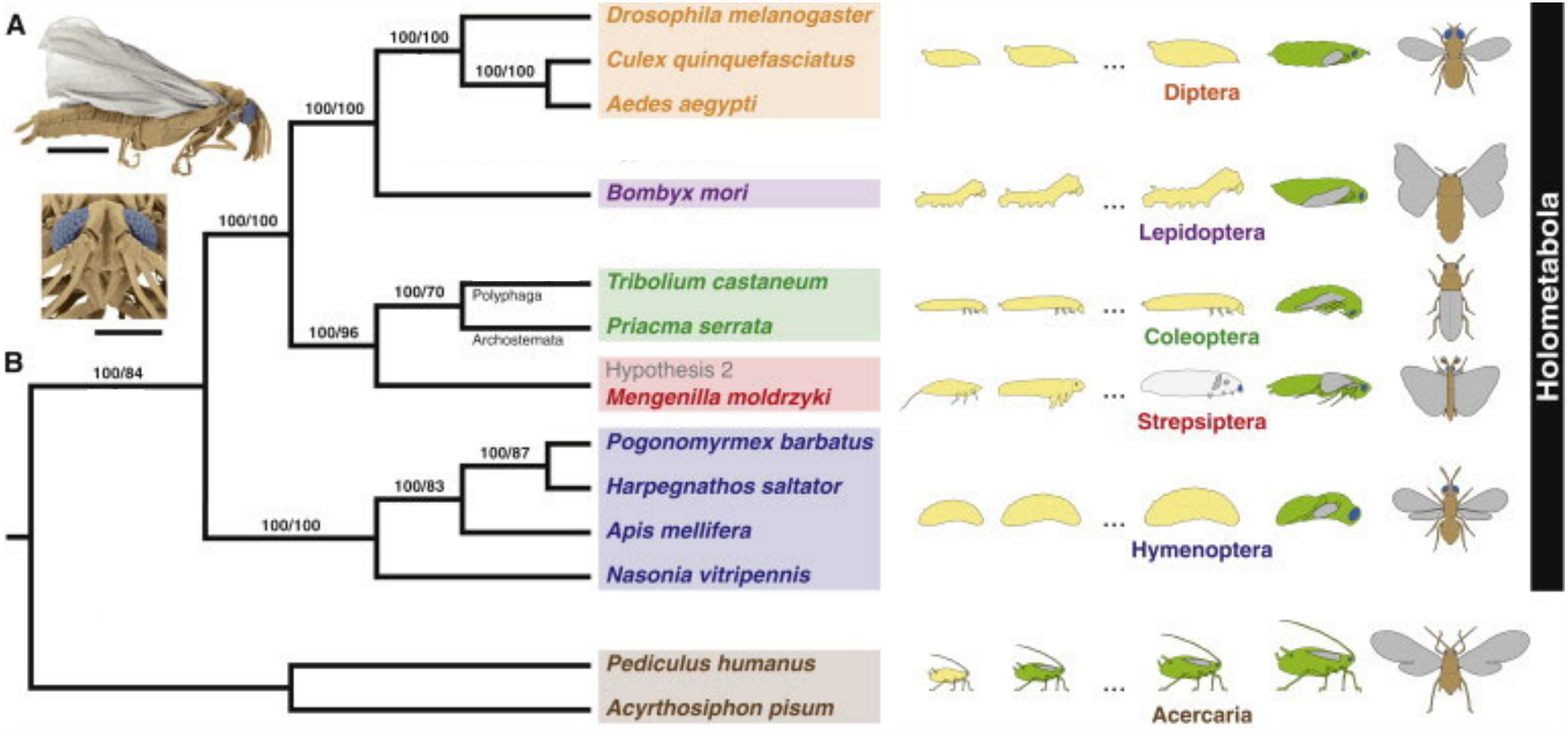


HKY85



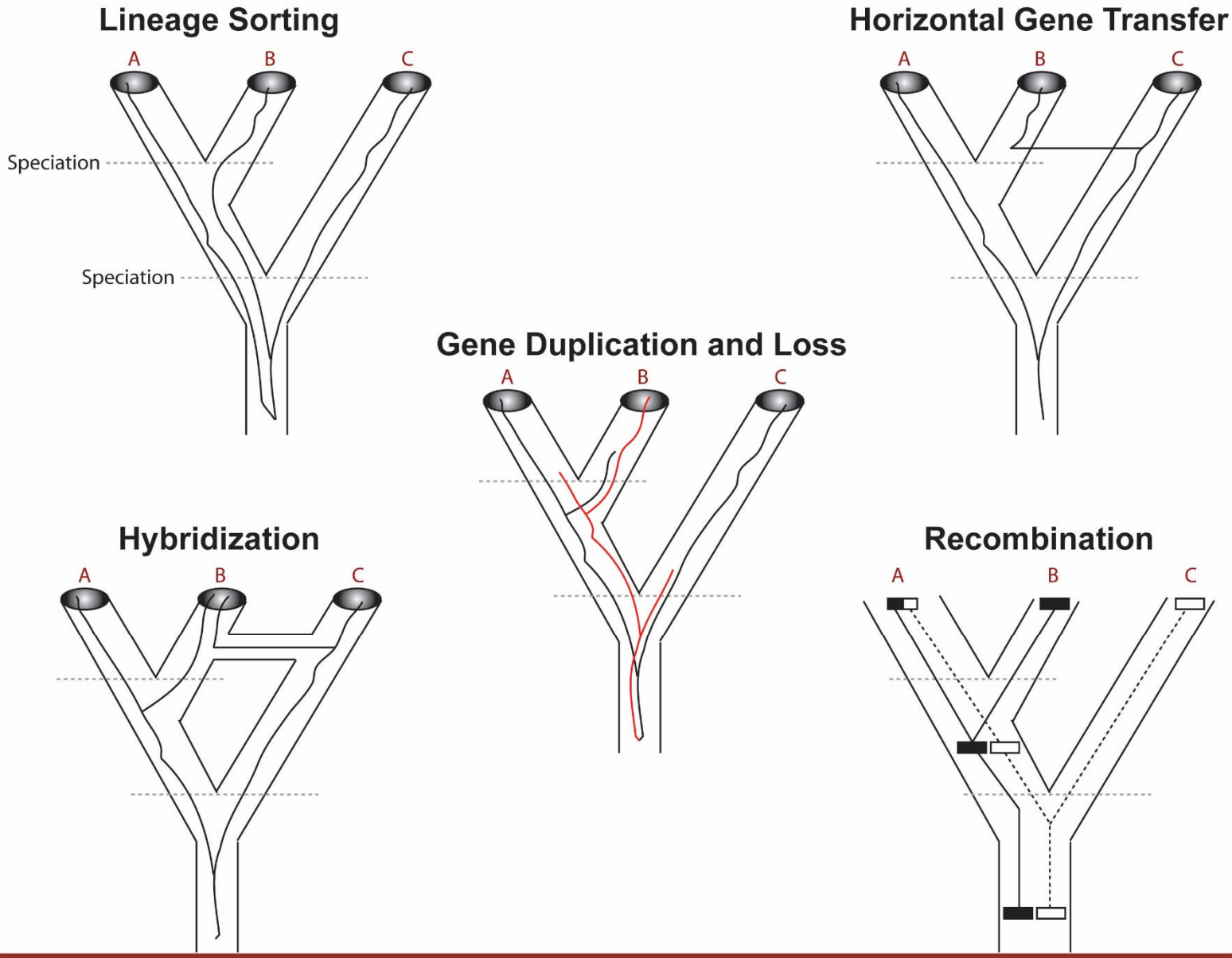
HKY85+GAMMA

Solving the Strepsiptera Problem with More Genes and Better Models



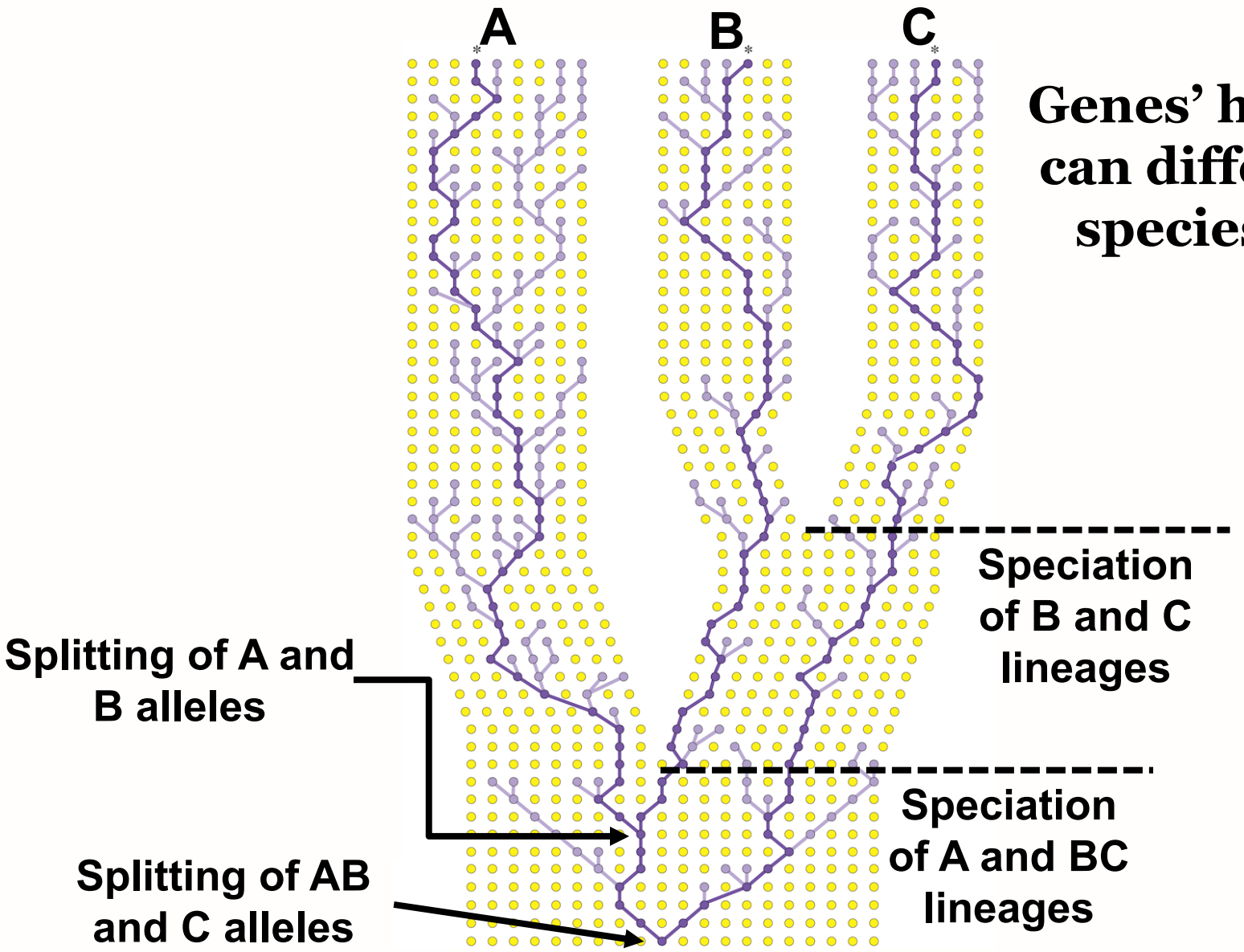
Niehuis et al. (2012) Curr. Biol.

Biological Factors



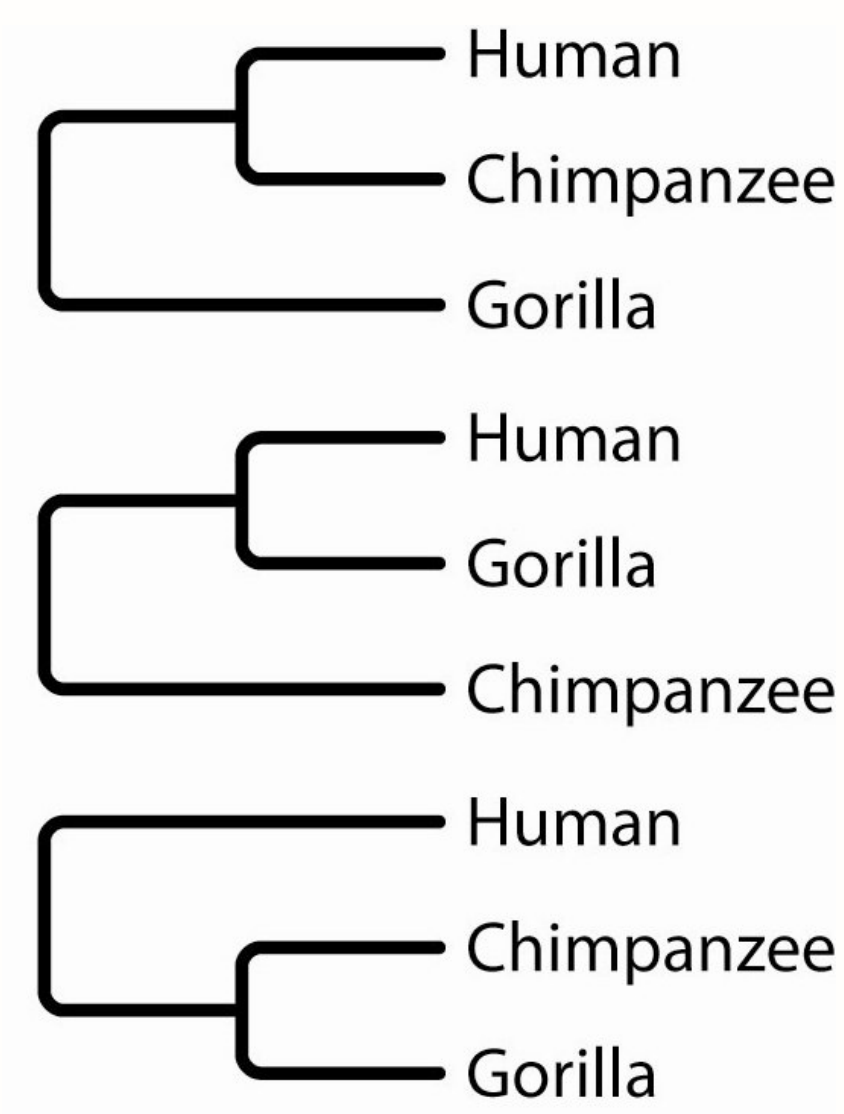
Degnan & Rosenberg (2009) Trends Ecol. Evol.

Stochastic Lineage Sorting of Ancestral Polymorphisms



Nei (1987) Mol. Evol. Genet.; Maddison (1997) Syst. Biol.

Lineage Sorting in Primates



Informative Sites

8,561 / 11,293

(~76%)

1,302 / 11,293

(~11.5%)

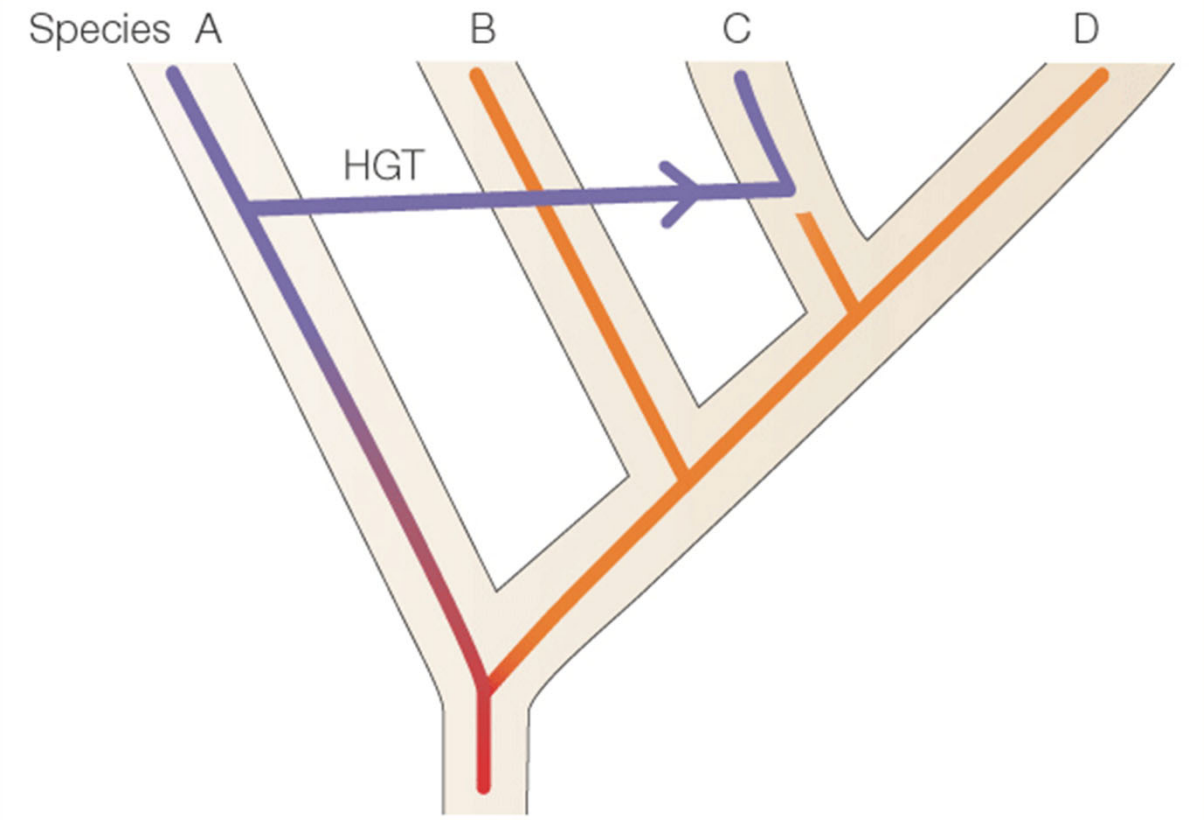
1,430 / 11,293

(~12.5%)

Patterson et al. (2006) Nature

Horizontal Transfer of Genes

Exchange of genes between organisms other than through reproduction



Gogarten & Townsend (2005) Nature Rev. Genet.

Horizontal Gene Transfer in Fungi

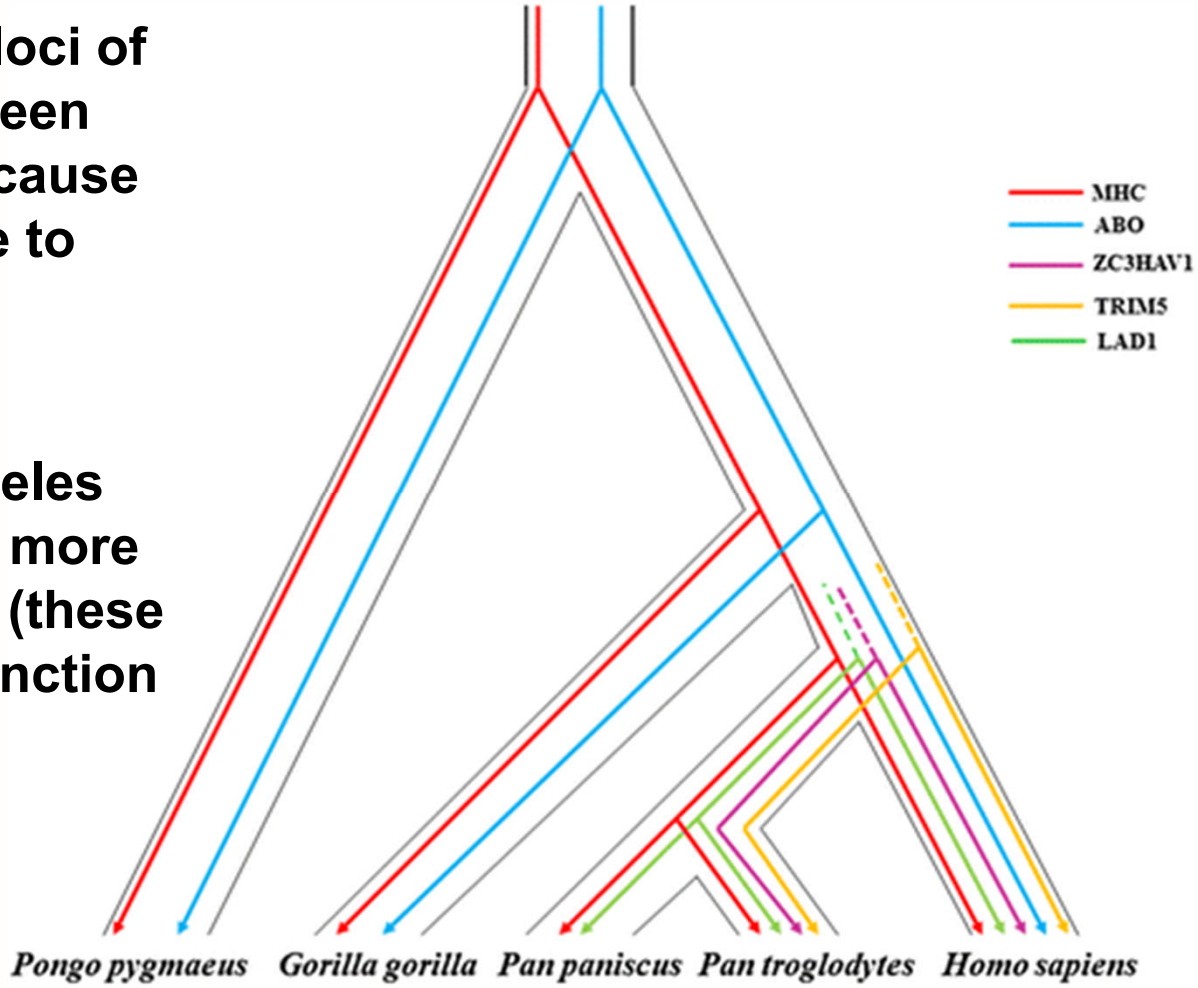
Slot & Rokas (2010) PNAS

Balancing Selection

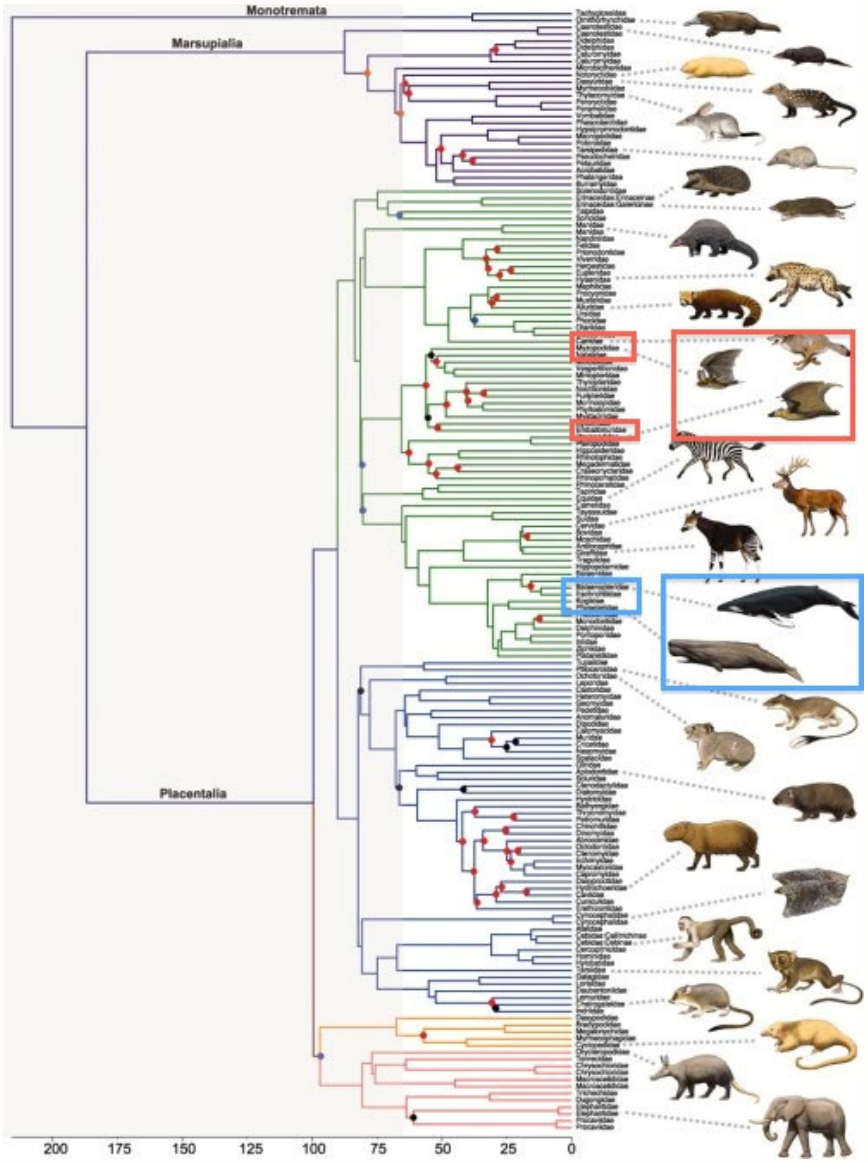
Balancing selection can maintain "trans-species polymorph-isms", in which the alleles are more ancient than the species

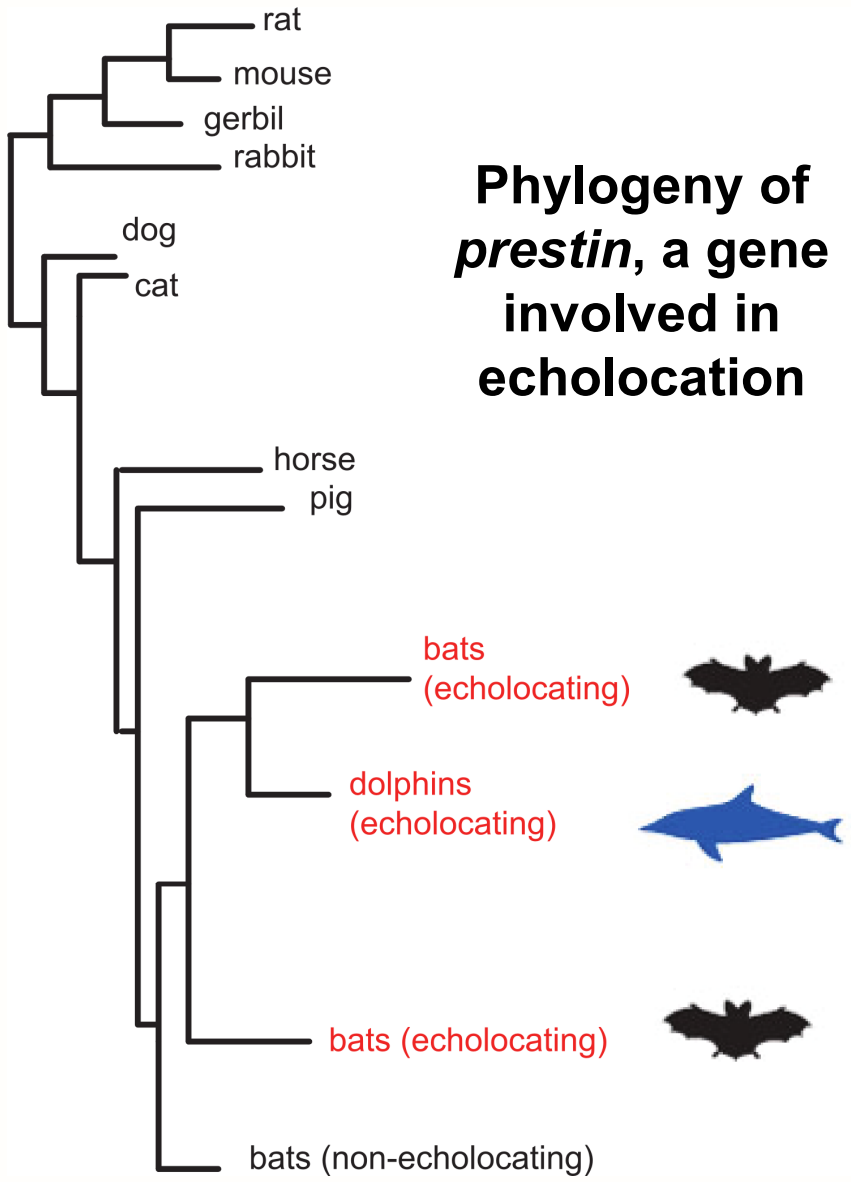
Best example: alleles at loci of the MHC – they have been retained by selection because they confer resistance to infection

Certain human MHC alleles appear to have diverged more than 65 million years ago (these alleles witnessed the extinction of dinosaurs!!!)



Positive Selection

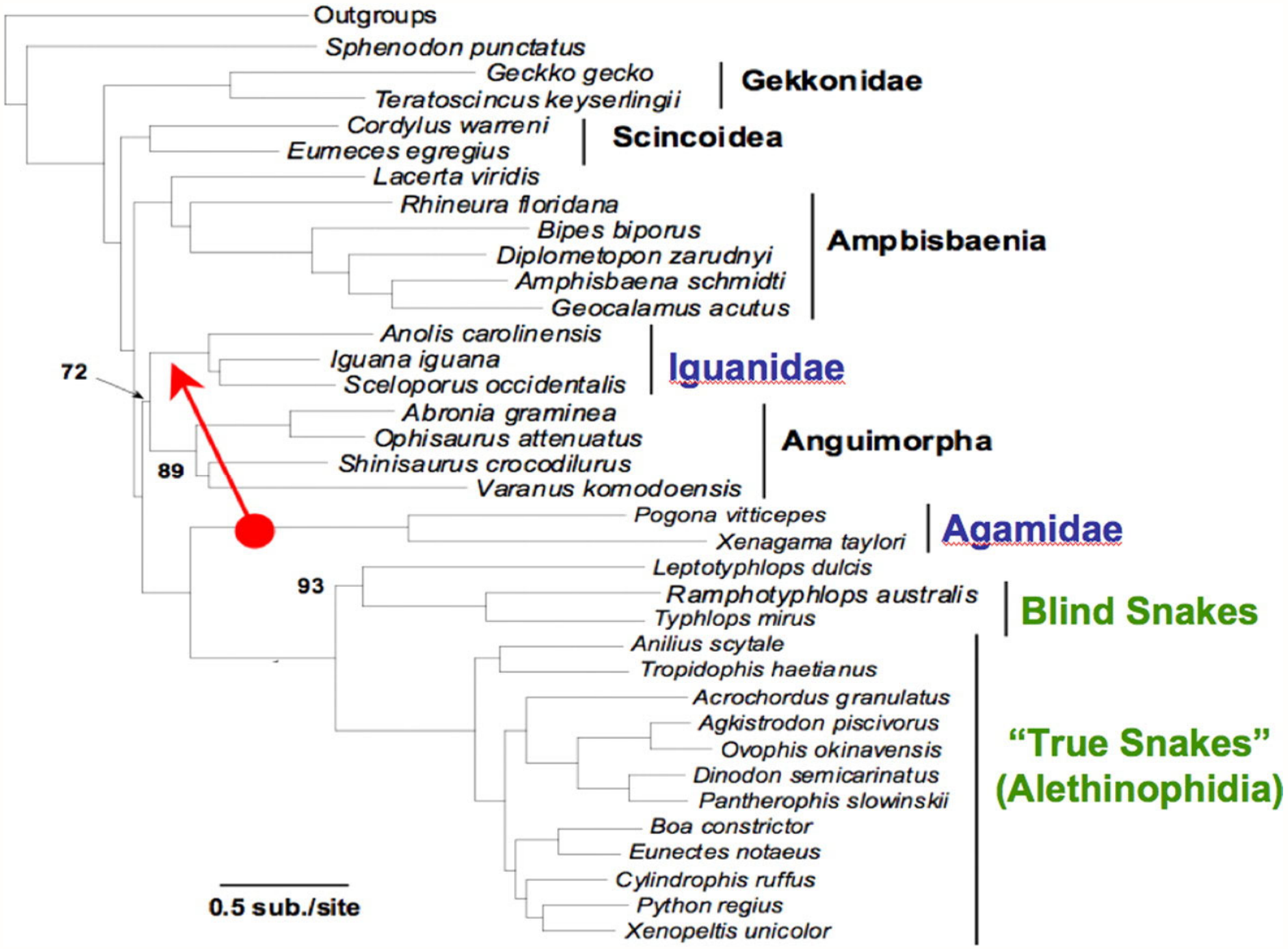




Phylogeny of *prestin*, a gene involved in echolocation

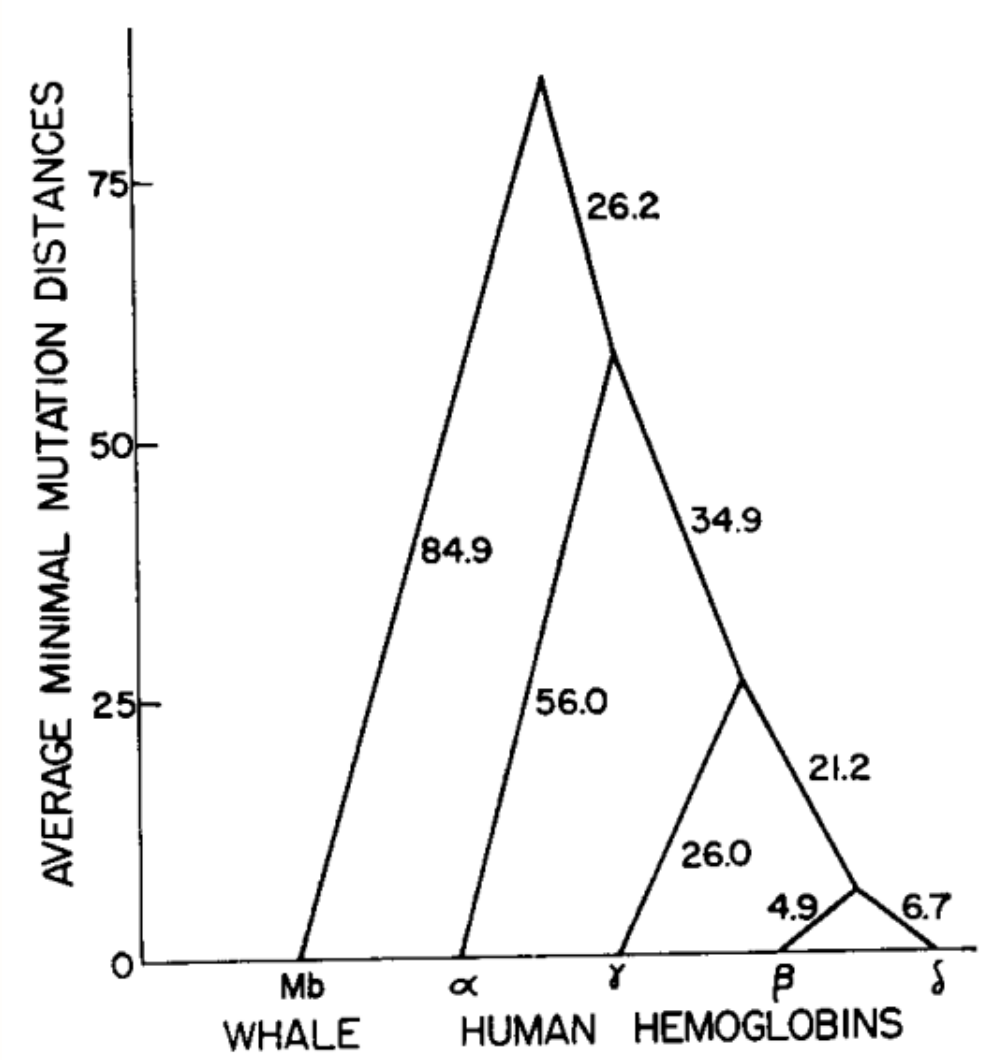
Li et al. (2010) Curr. Biol.

Positive Selection



Castoe et al. (2009) PNAS

Gene Duplication Can Confound Phylogeny



(*15*). A cautionary note may be derived from this. A wildly incorrect result could easily be obtained if the presence of multiple, homologous genes were not recognized and a phylogeny were constructed from sequences which were coded for, say, half by genes for alpha hemoglobin chains and half by genes for beta hemoglobin chains. This results from the speciation having occurred more recently than the gene duplication which permitted the separate evolution of the alpha and beta genes.

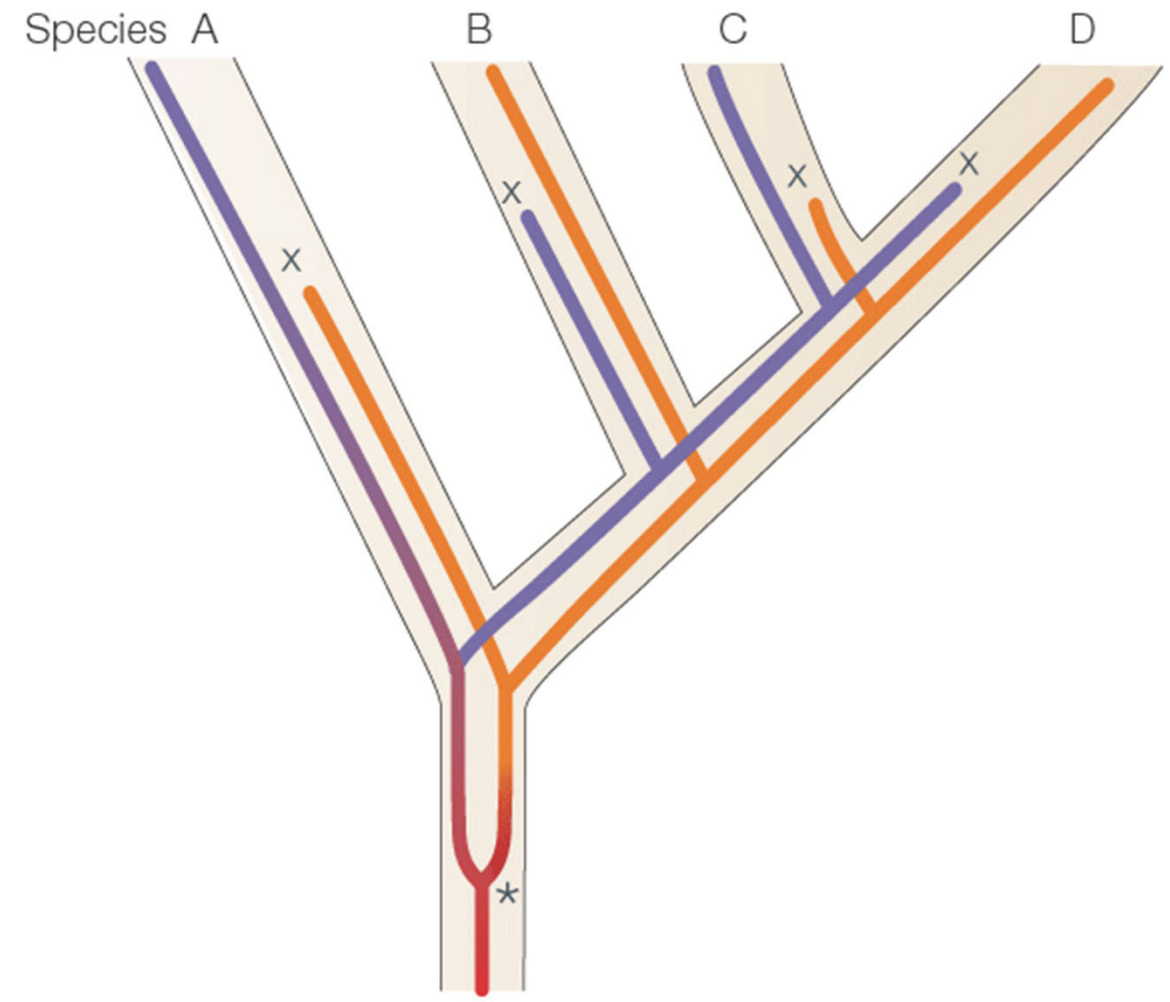
Fitch & Margoliash (1967) Science

Gene Duplication Can Confound Phylogeny

homologous. It has been pointed out before that a phylogeny of birds and mammals based upon a haphazard mixture of α and β hemoglobins would be biological nonsense since the initial dichotomy would be on the distinction between the α and β genes rather than between the birds and the mammals (Fitch and Margoliash, 1967). Therefore, there should be two subclasses of homology. Where the homology is the result of gene duplication so that both copies have descended side by side during the history of an organism, (for example, α and β hemoglobin) the genes should be called *paralogous* (para = in parallel). Where the homology is the result of speciation so that the history of the gene reflects the history of the species (for example α hemoglobin in man and mouse) the genes should be called *orthologous* (ortho = exact). Phylogenies require orthologous, not paralogous, genes. Note

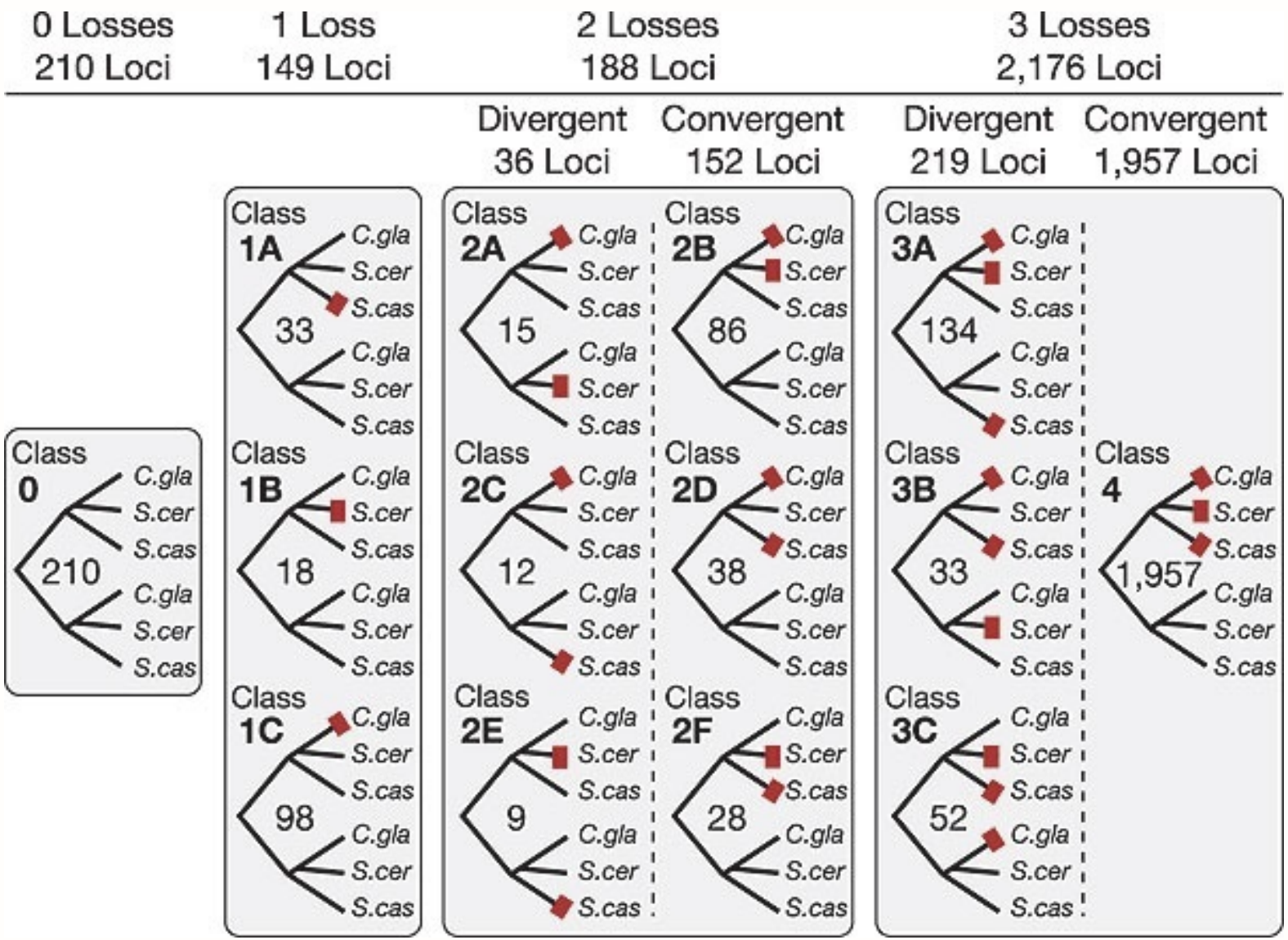
Fitch (1970) Syst. Zool.

Gene Duplication and Loss



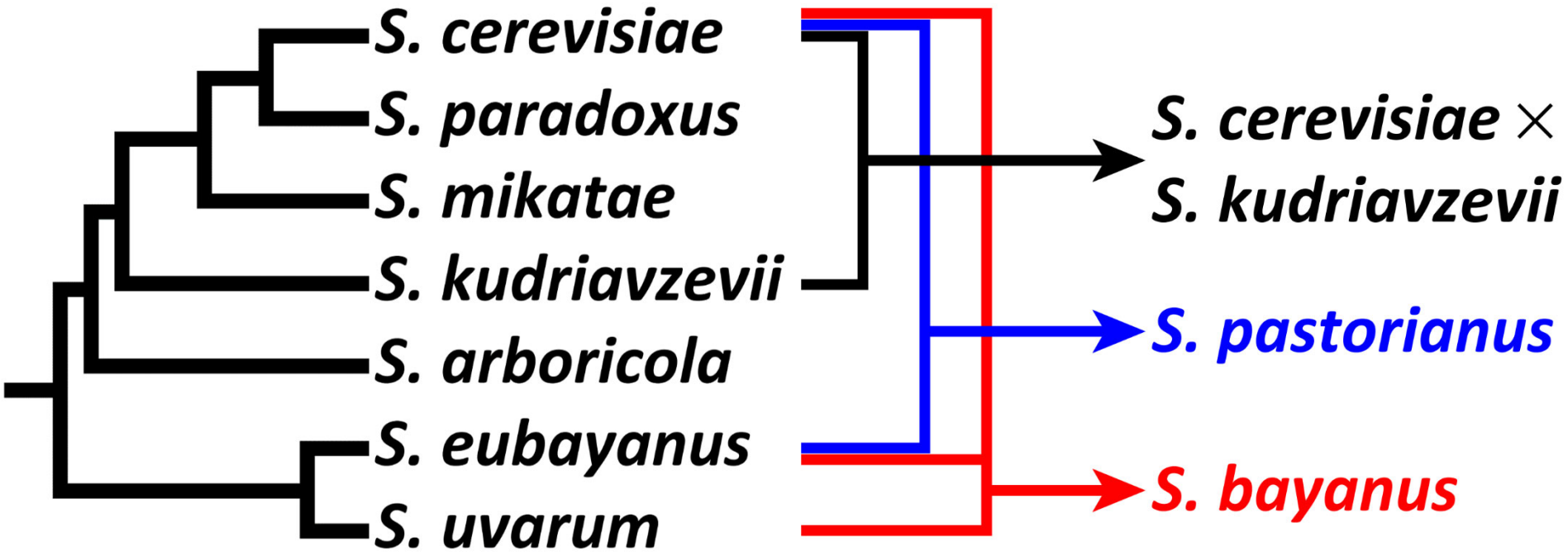
Gogarten & Townsend (2005) Nature Rev. Genet.

Gene Duplication and Loss



Scannell et al. (2006) Nature

Hybridization / Introgression



S. eubayanus **was discovered in 2011 – until then, *S. bayanus* was thought to be a "pure" species**

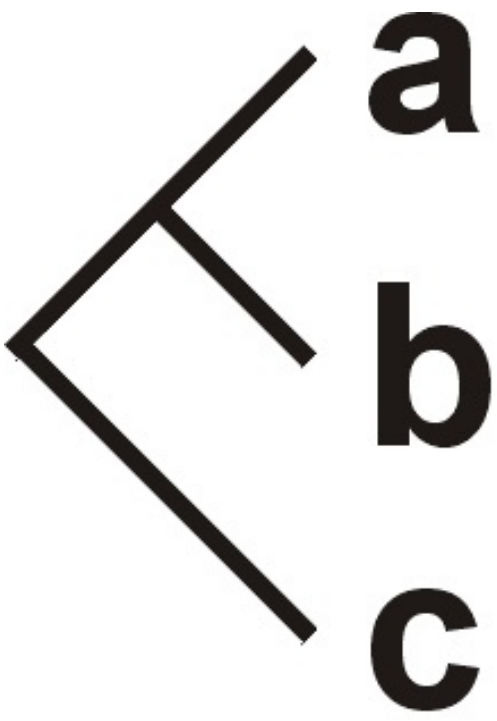
S. cerevisiae **– *S. paradoxus* divergence ≈ human – mouse divergence**
S. cerevisiae **– *S. uvarum* divergence ≈ human – chicken divergence**

OK, I now get why

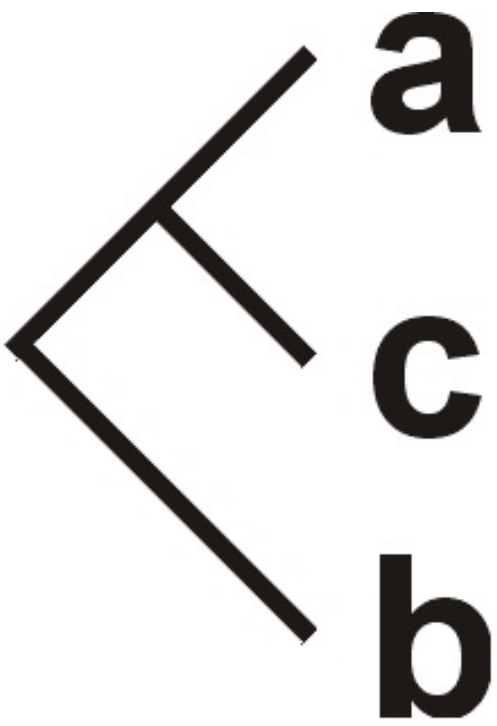
gene trees ≠ species trees

What does this have to do with phylogenomics?

All this Manifests Itself as Incongruence



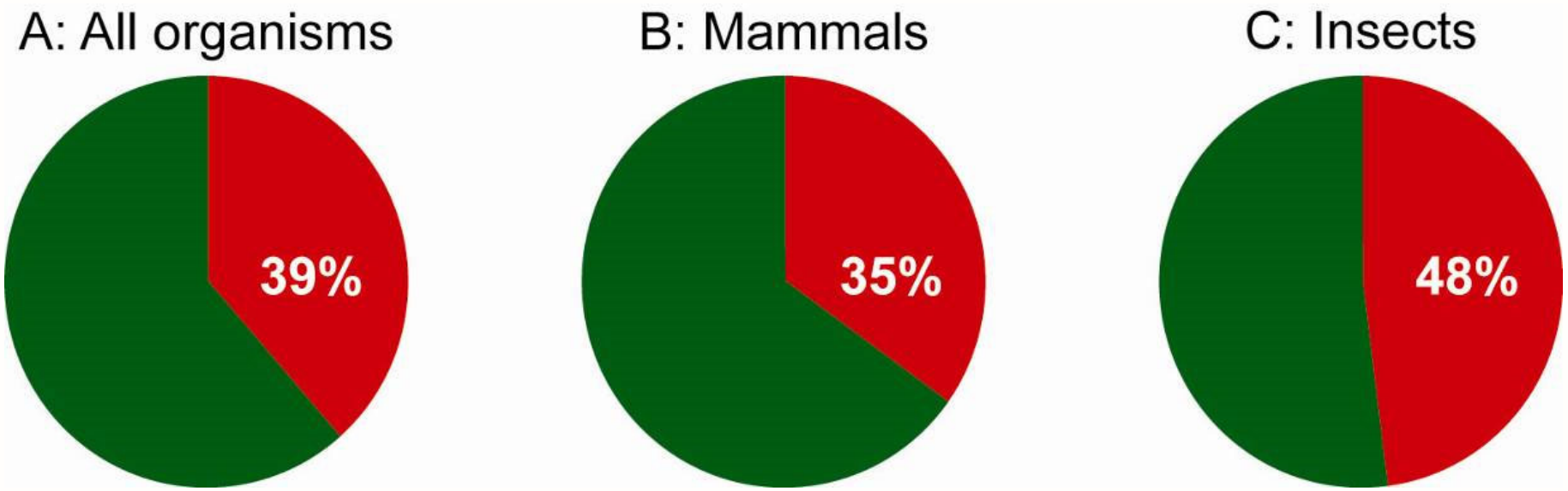
Gene X



Gene Y

Species phylogeny?

Incongruence is Pervasive in the Phylogenetics Literature



Rokas & Chatzimanolis (2008) in Phylogenomics (W. J. Murphy, Ed.)

A Systematic Evaluation of Single Gene Phylogenies

S. cerevisiae

S. bayanus

S. paradoxus

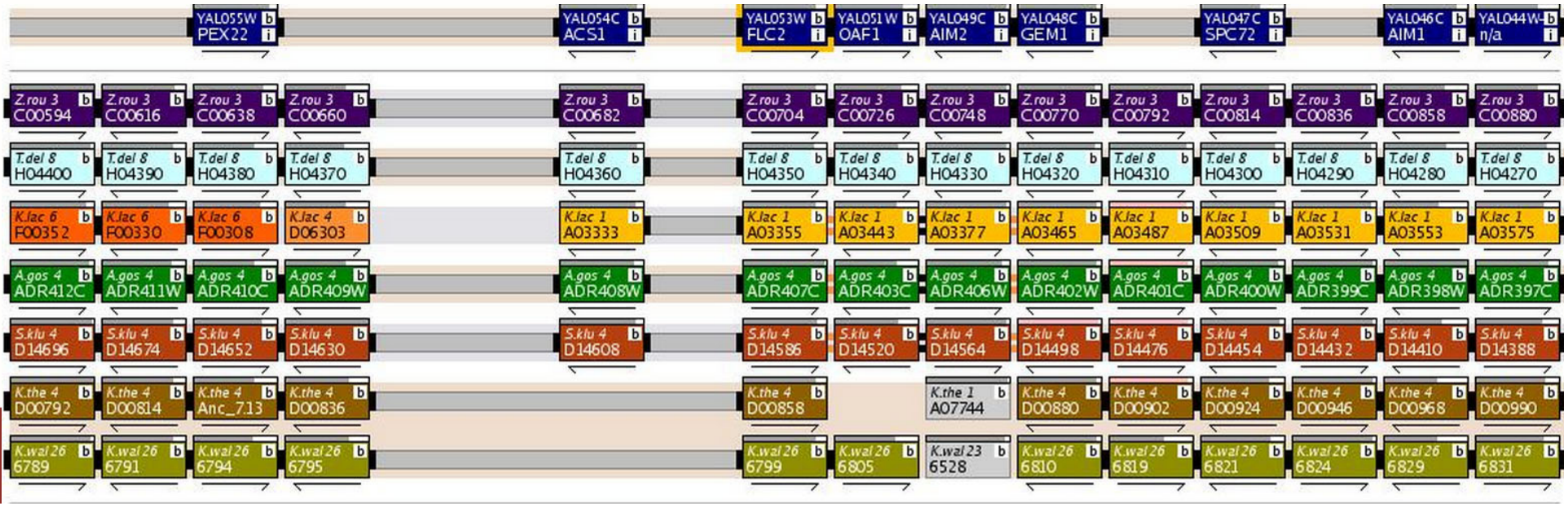
S. castellii

S. mikatae

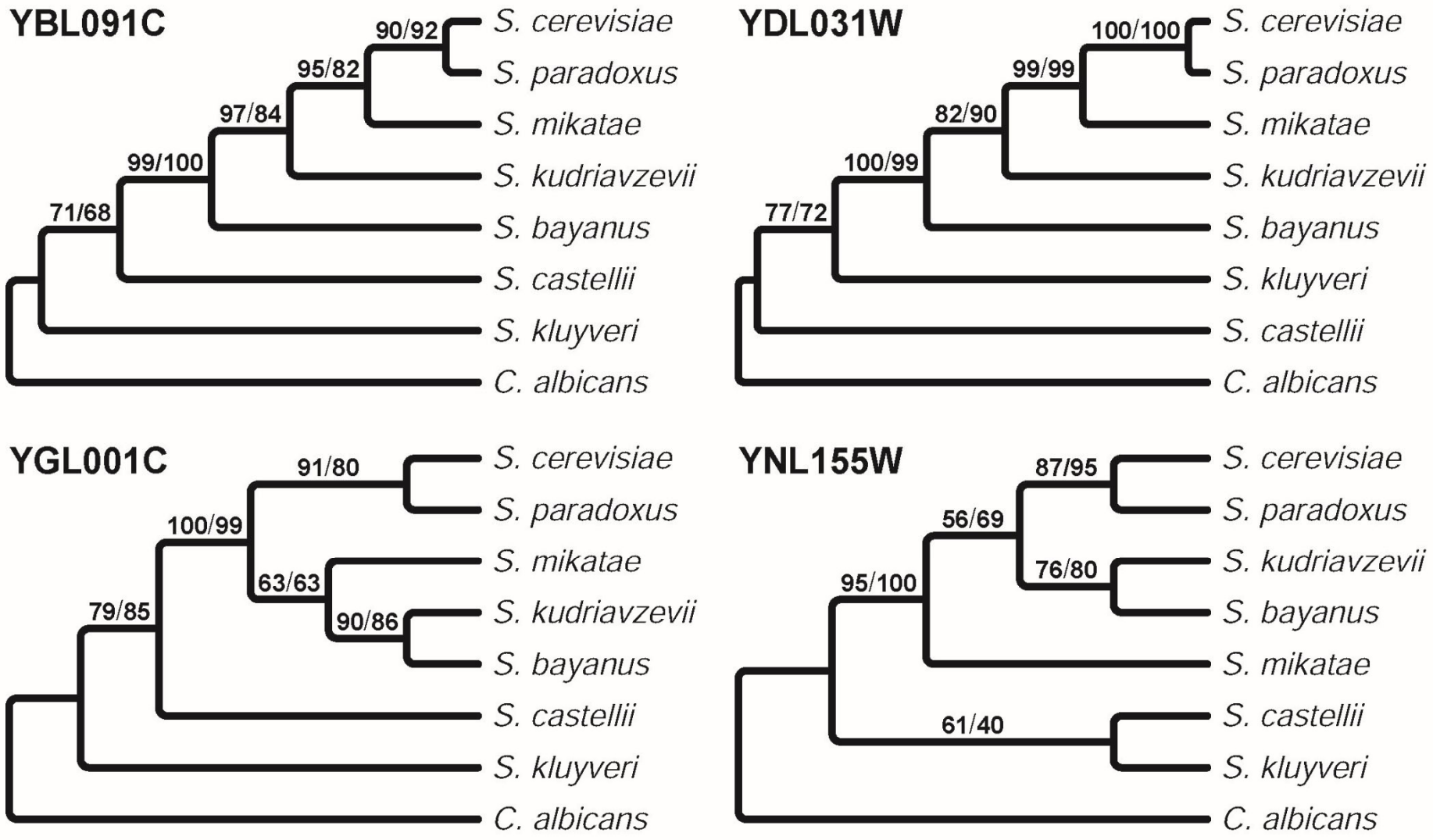
S. kluyveri

S. kudriavzevii

Candida glabrata

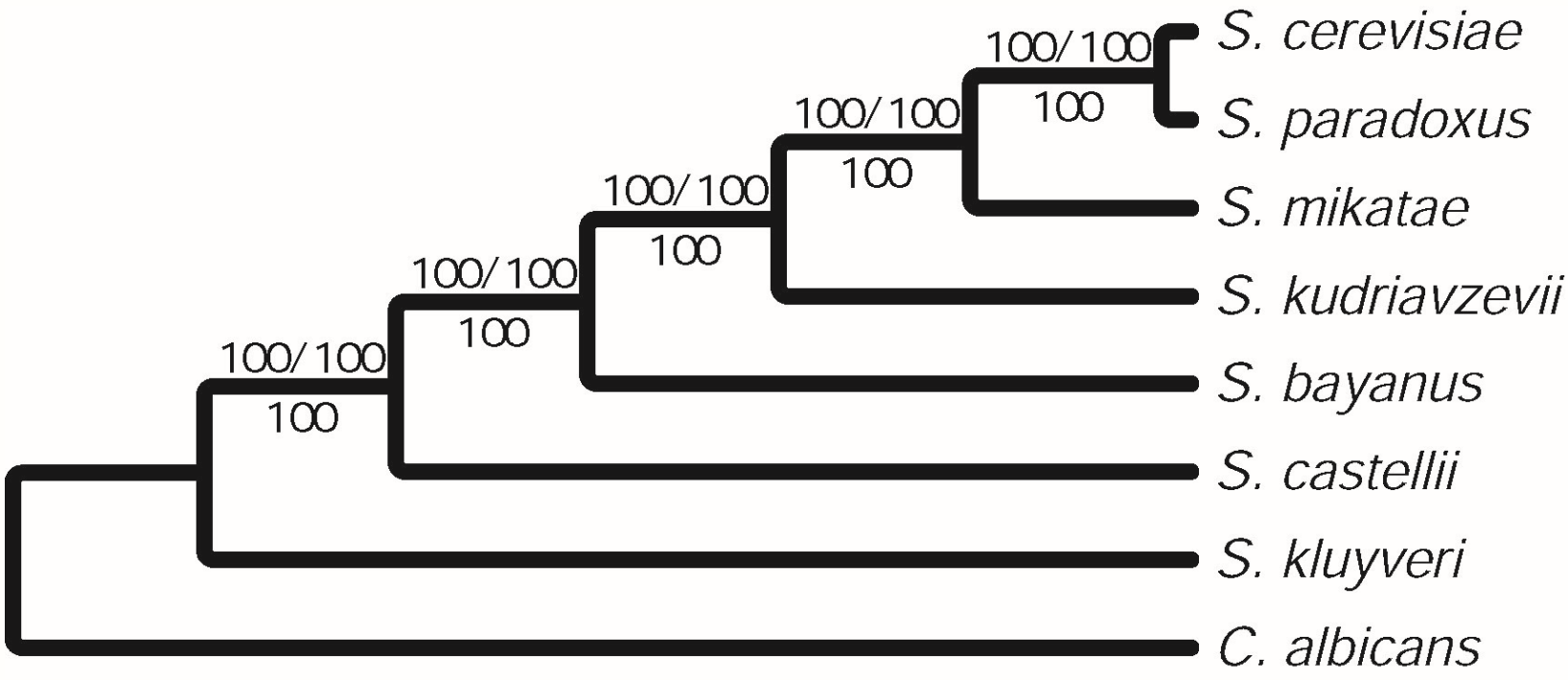


Incongruence at the Single Gene Level



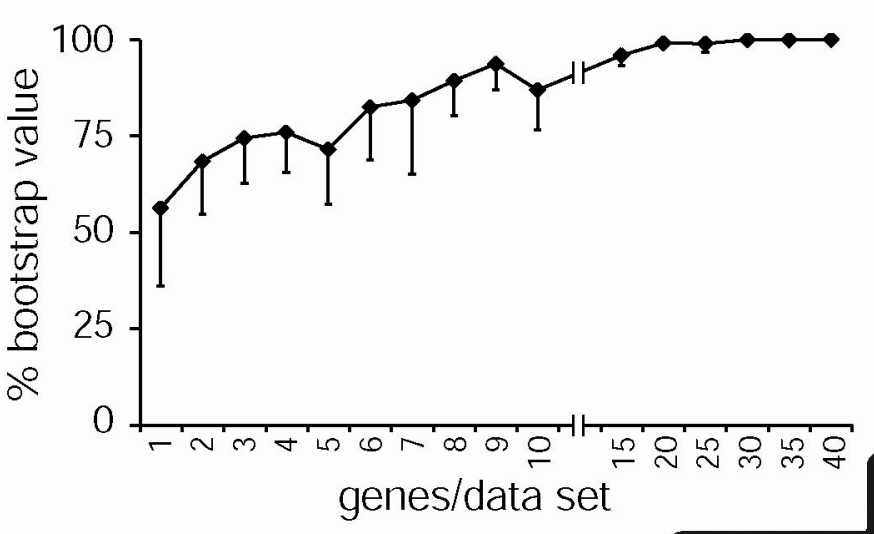
ML / MP *Anonymous Reviewer for Nature (2003)* *Rokas et al. (2003) Nature*

Concatenation of 106 Genes Yields a Single Yeast Phylogeny

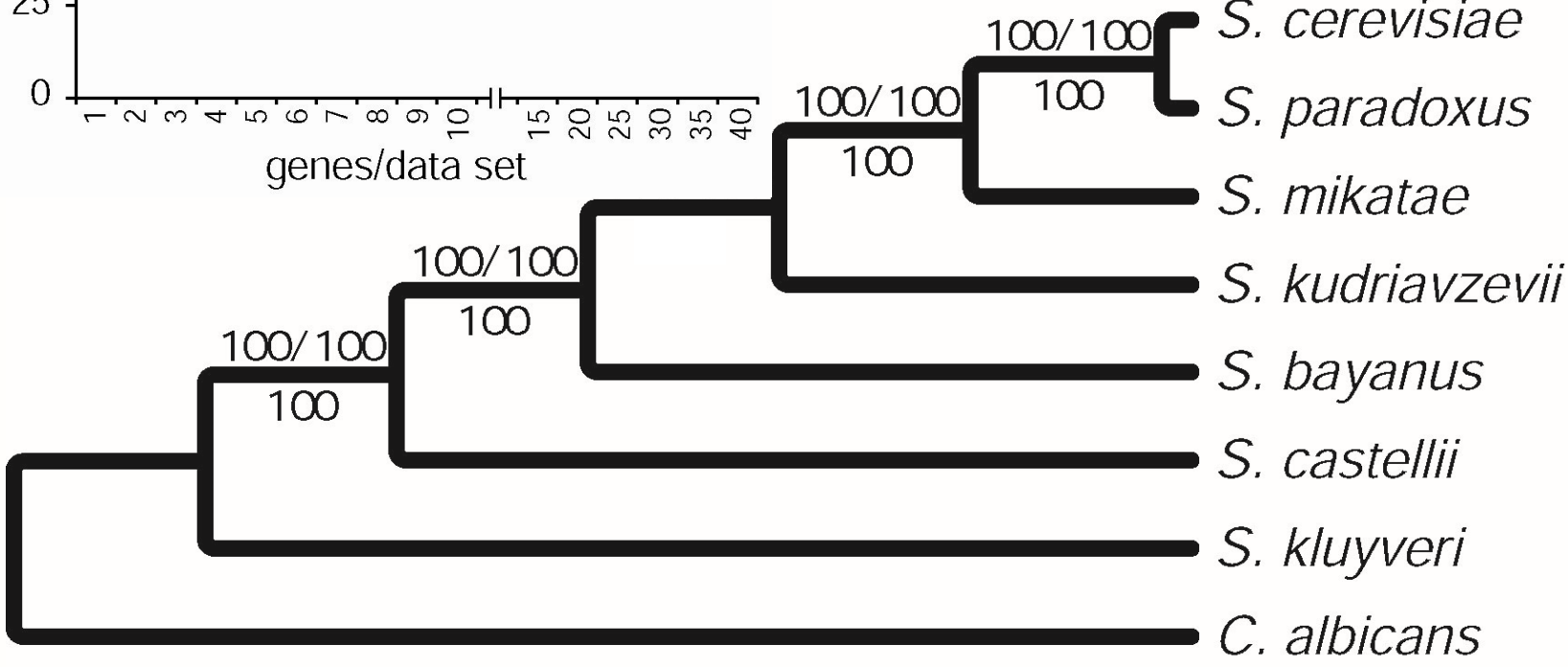


ML / MP on nt
MP on aa

The Use of Many Genes Eliminates Stochastic Error



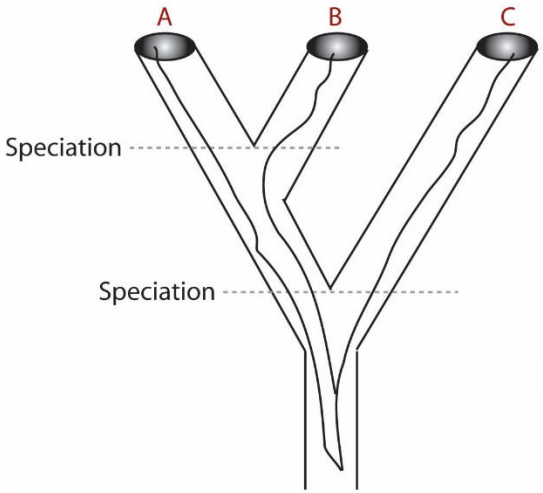
ML / MP on nt
MP on aa



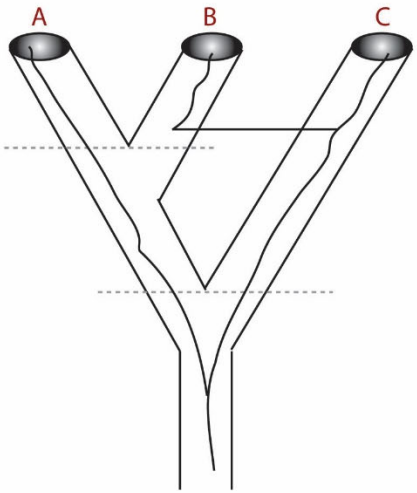
Rokas et al. (2003) Nature

Gene Trees Can Differ from Species Trees

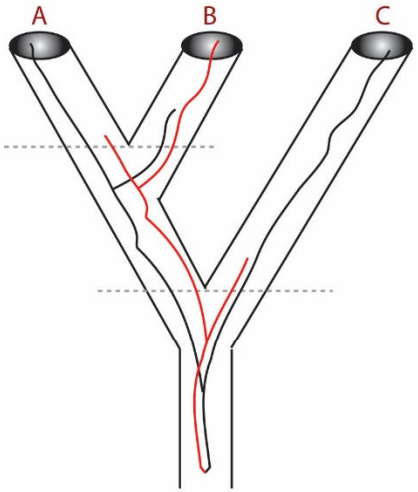
Lineage Sorting



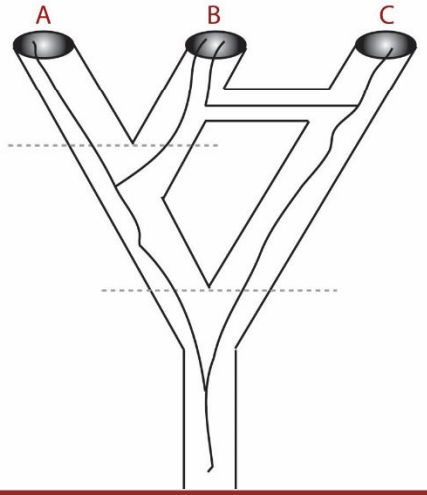
Horizontal Gene Transfer



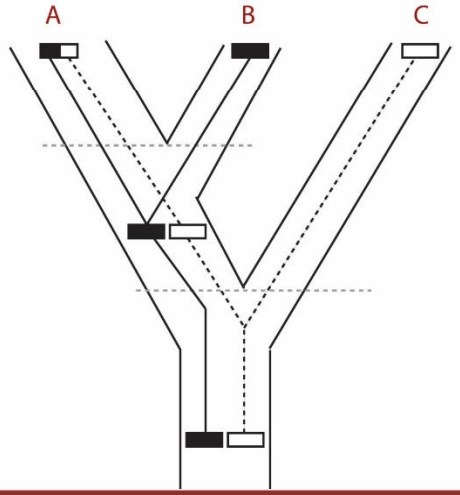
Gene Duplication and Loss



Hybridization



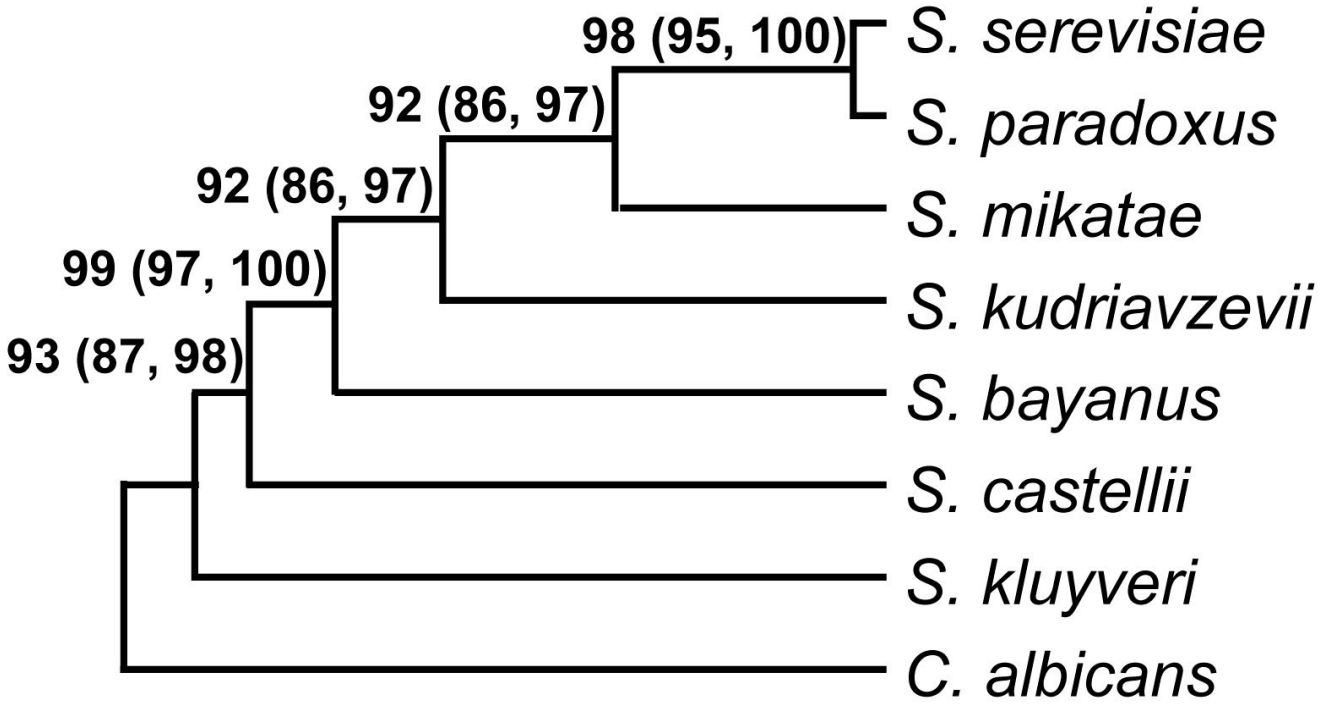
Recombination



Degnan & Rosenberg (2009) Trends Ecol. Evol.

Inferring the Species Tree from Individual Gene Histories

Concordance Factor: The proportion of the genome for which a clade is true



New methods to calculate concordance factors for phylogenomic datasets

Bui Quang Minh, Matthew Hahn, Robert Lanfear

doi: https://doi.org/10.1101/487801

This article is a preprint and has not been peer-reviewed [what does this mean?].

bioRxiv
THE PREPRINT SERVER FOR BIOLOGY

Ané et al. (2007) Mol. Biol. Evol.

The Phylogenomics Era – "Resolving" the Tree of Life

Syst. Biol. 61(1):150–164, 2012

DOI:10.1093/sysbio/syr089
Advance Access publication on September 7, 2011

LETT
LETT

Phylogenomic Analysis Resolves the Interordinal Relationships and Rapid Diversification of the Laurasiatherian Mammals

XUMING ZHOU, SHIXIA XU, JUNXIAO XU, BINGYAO CHEN, KAIYA ZHOU, AND GUANG YANG*

*Jiangsu Key Laboratory for Biodiversity and Biotechnology, College of Life Sciences, Nanjing Normal University, Nanjing 210046, China; *Correspondence to be sent to: Jiangsu Key Laboratory for Biodiversity and Biotechnology, College of Life Sciences, Nanjing Normal University, Nanjing 210046, China; E-mail: gyang@njnu.edu.cn.*

Resolving the evolutionary relationships of molluscs with phylogenomic tools

nature

LETTERS

Stephen A. Smith[1,2], Nerida G. Wilson[3,4], Freya
Gonzalo Giribet[5] & Casey W. Dunn[1]

Syst. Biol. 57(6):920–938, 2008

ISSN: 1063-5157 print / 1076-836X online
DOI: 10.1080/10635150802570791

Resolving Arthropod Phylogeny: Exploring Phylogenetic Signal within 41 kb of Protein-Coding Nuclear Gene Sequence

JEROME C. REGIER,[1] JEFFREY W. SHULTZ,[2] AUSTEN R. D. GANLEY,[3,6] APRIL HUSSEY,[1] DIANE SHI,[1] BERNARD BALL,[3] ANDREAS ZWICK,[1] JASON E. STAJICH,[3,7] MICHAEL P. CUMMINGS,[4] JOEL W. MARTIN,[5] AND CLIFFORD W. CUNNINGHAM[3]

Toward Resolving t
Tree: The Phylogen
of Jakobids and Cercozoans

Yeast

Prion-Like Proteins in the Fungal Kingdom

Edgar M. Medina · Gary W. Jones · David A. Fitzpatrick

An

OPEN ACCESS Free

Toward Resolving Priors

Towards

Renae C. Pratt, Gillian C. Gibb,* Mary Morgan-Richards,* Matthew J. Phillips,† Michael D. Hendy,* and David Penny**

*Allan Wilson Centre for Molecular Ecology and Evolution, Massey University, Palmerston North, New Zealand; and †Centre for Macroevolution and Macroecology, School of Botany and Zoology, Australian National University, Canberra ACT, Australia

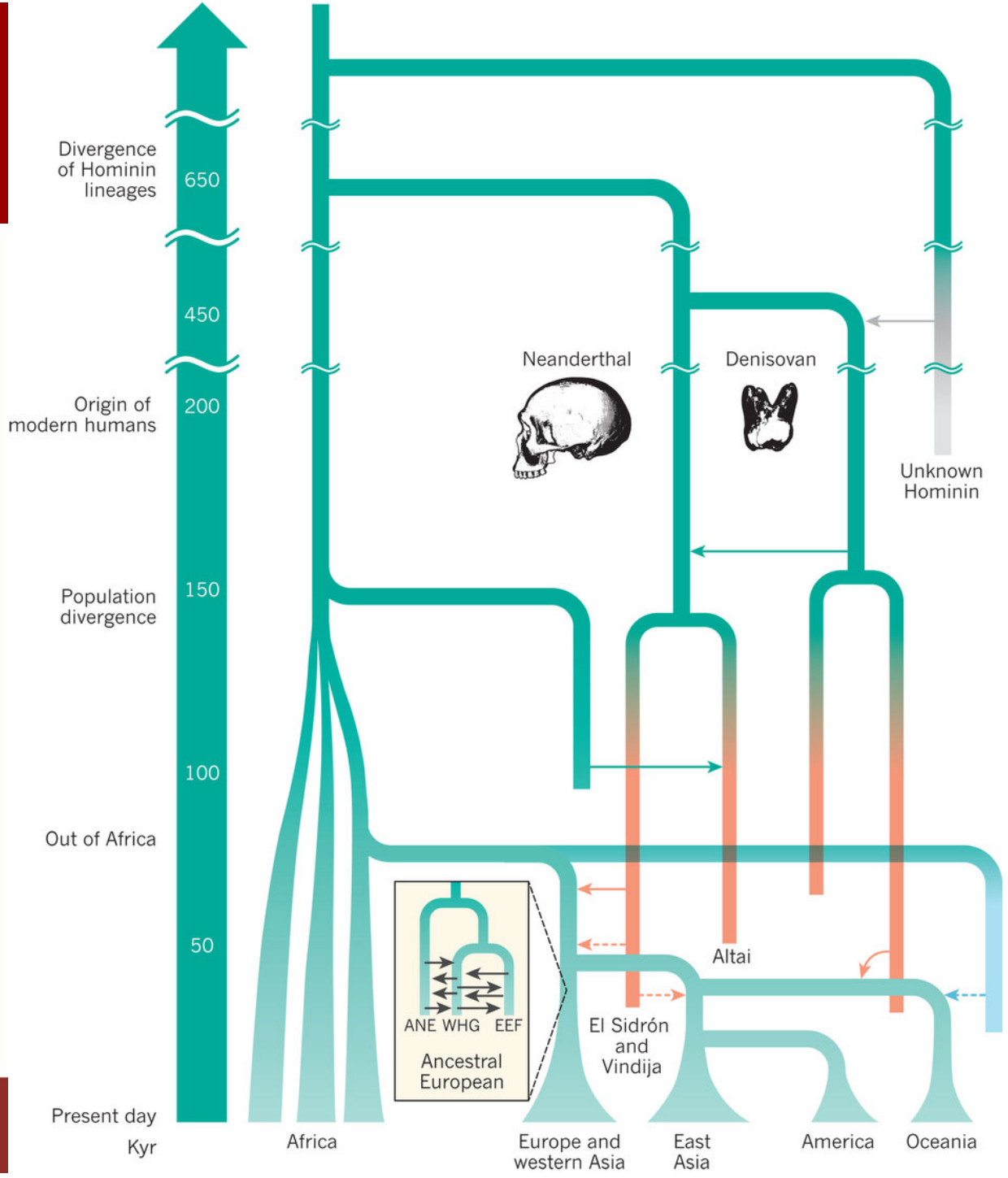
Samuli Lehtonen

Department of Biology, U

Have we eliminated incongruence?

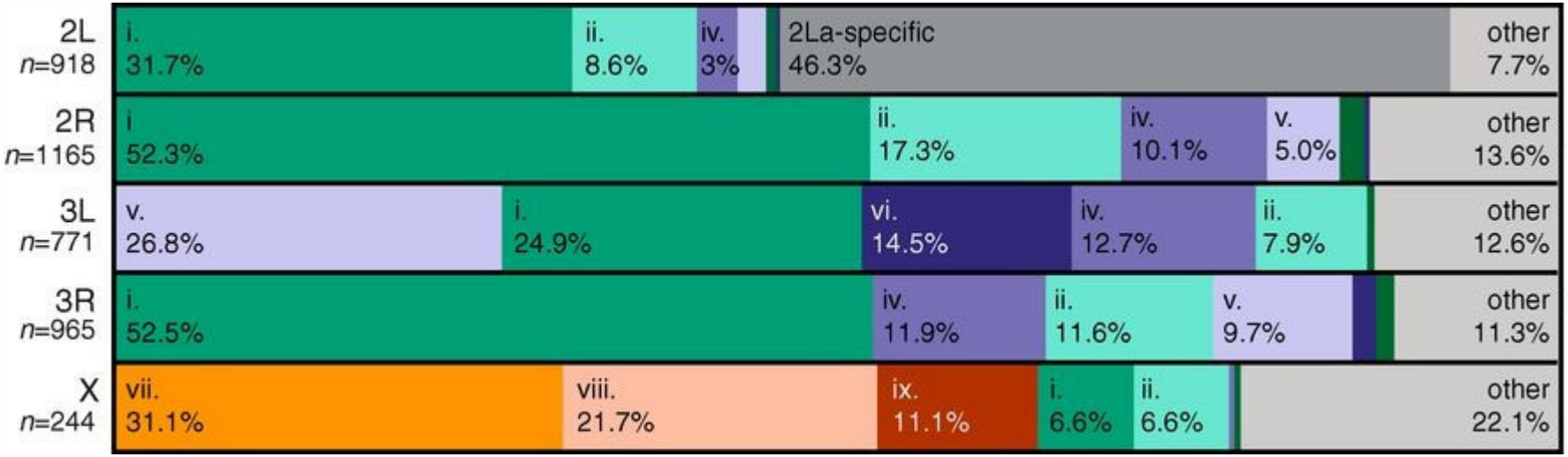
Figuring out what's going on is easier at shallow depths, b/c systematic error is usually absent

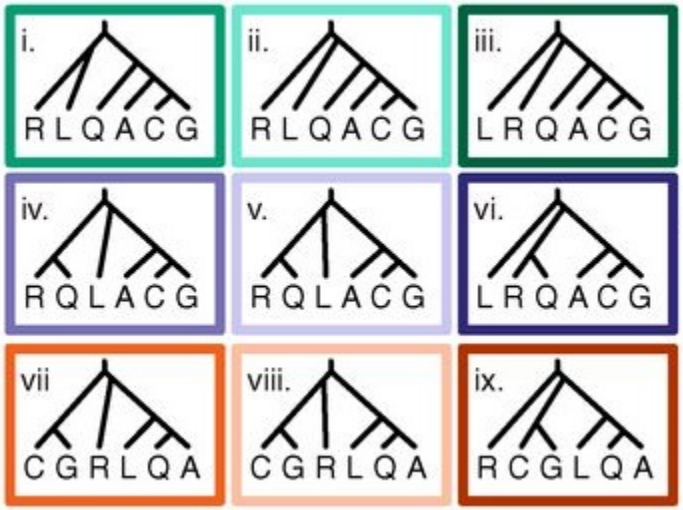
The Evolution of Human Populations



Nielsen et al. (2017) Nature

"Easier" Doesn't Mean "Easy"!





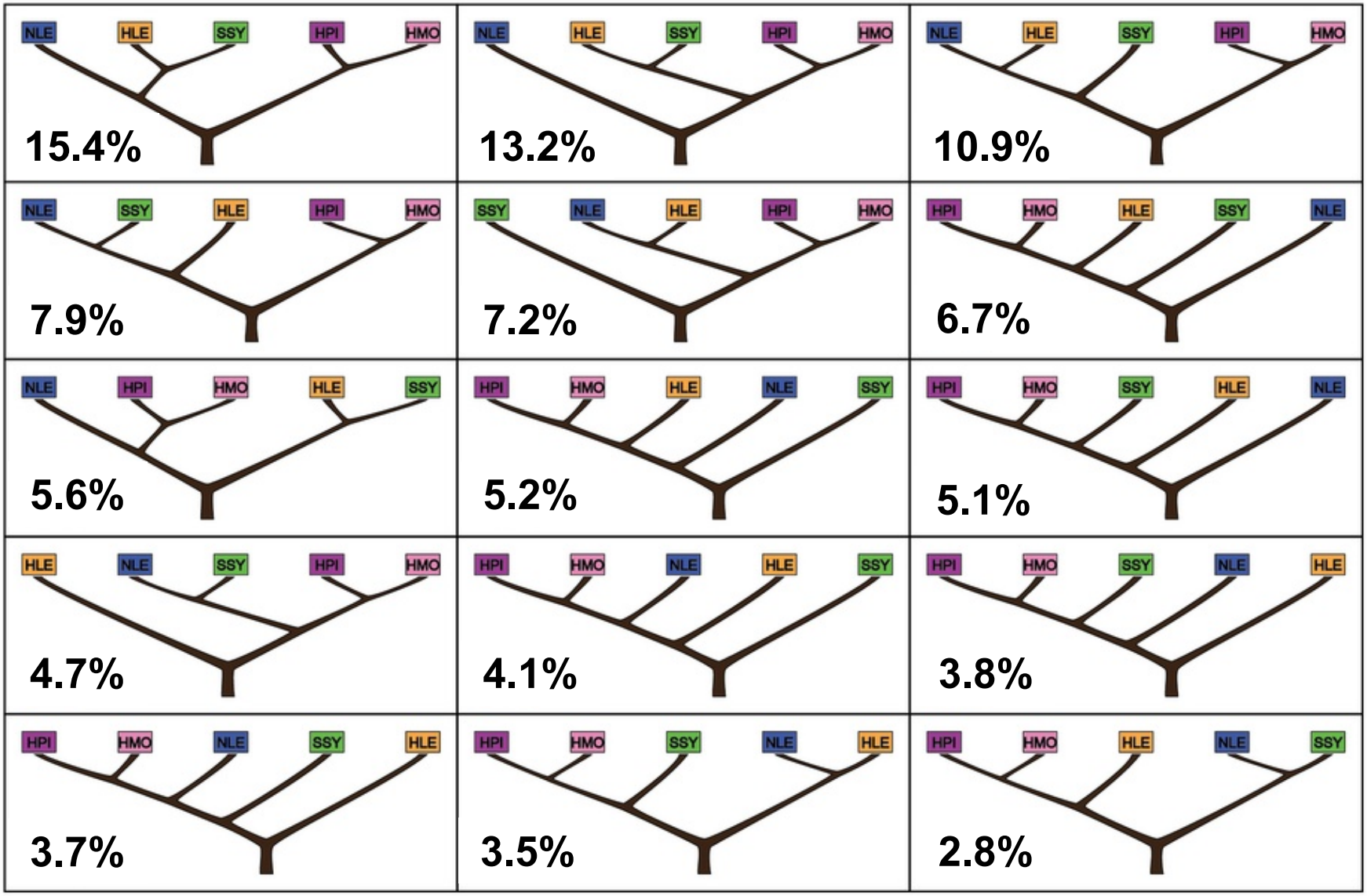
Fontaine *et al.* adhere to a classical view that there is a "true species tree" […]. But given that the bulk of the genome has a network of relationships that is different from this true species tree, perhaps we should dispense with the tree and acknowledge that these genomes are best described by a network, and that they undergo rampant reticulate evolution

Fontaine et al. (2015) Science; Clark & Messer (2015) Science

The Phylogeny of Primate Genera

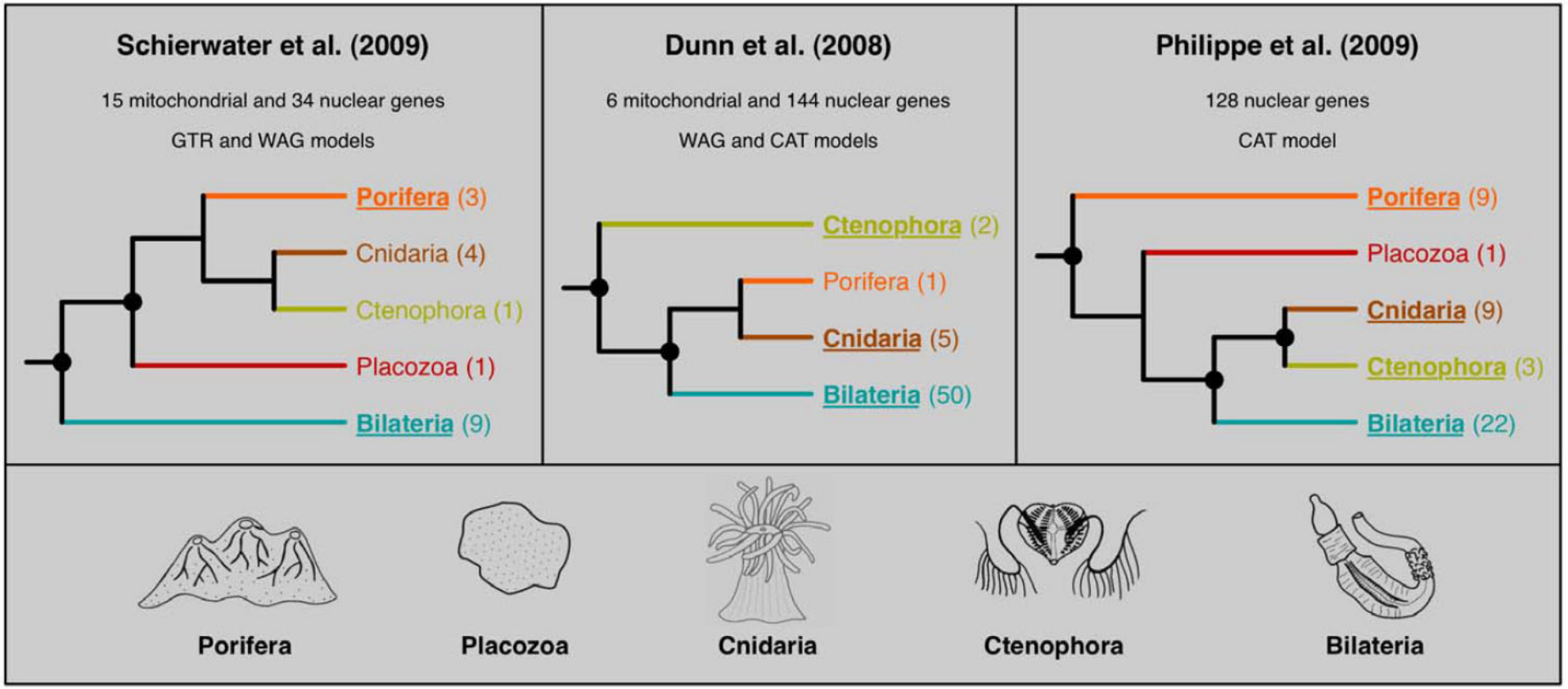
Nomascus leucogenys	*Hoolock leuconedys*	*Symphalangus syndactylus*	*Hylobates pileatus*	*Hylobates moloch*
				
NLE	HLE	SSY	HPI	HMO

"Easier" Doesn't Mean "Easy"!



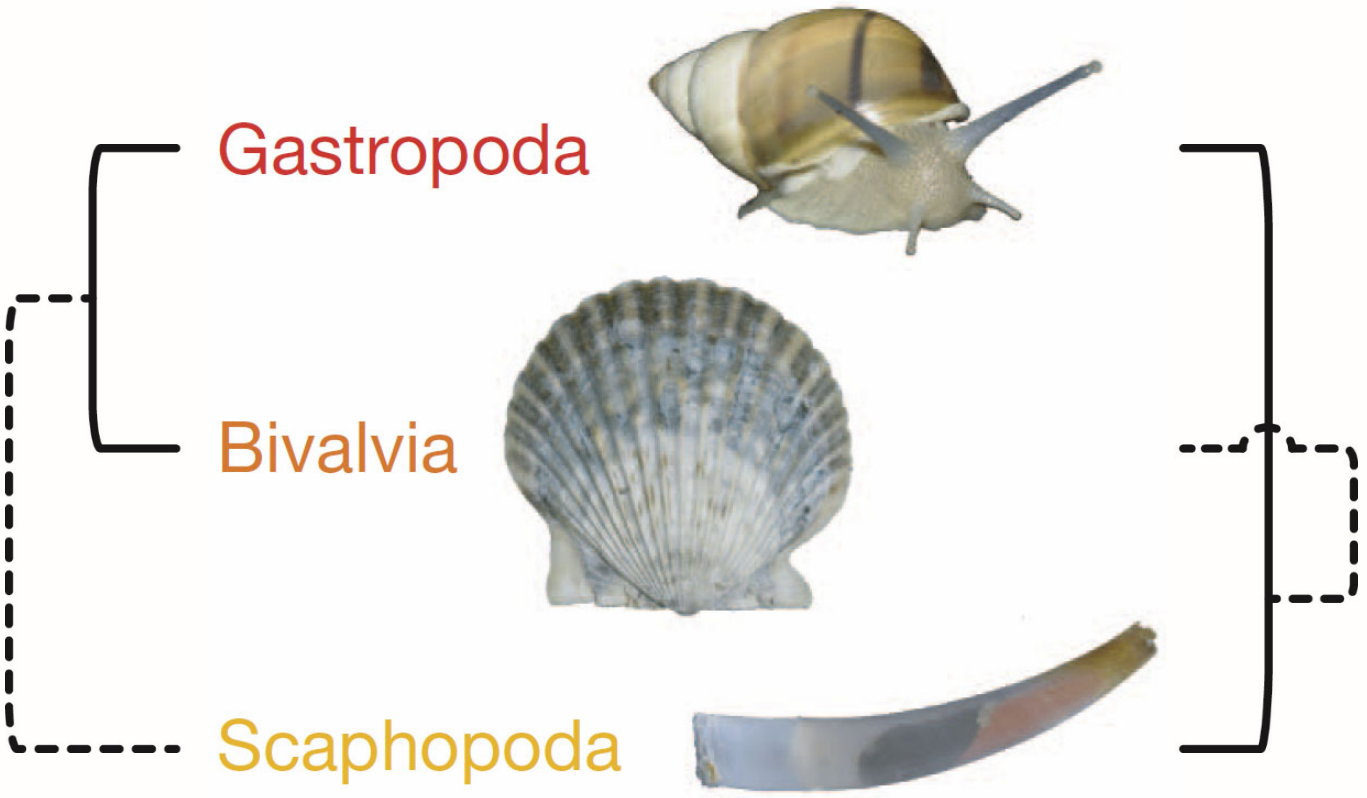
Carbone et al. (2014) Nature

Incongruence in Deep Time is More Challenging



Philippe et al. (2011) PLoS Biol.

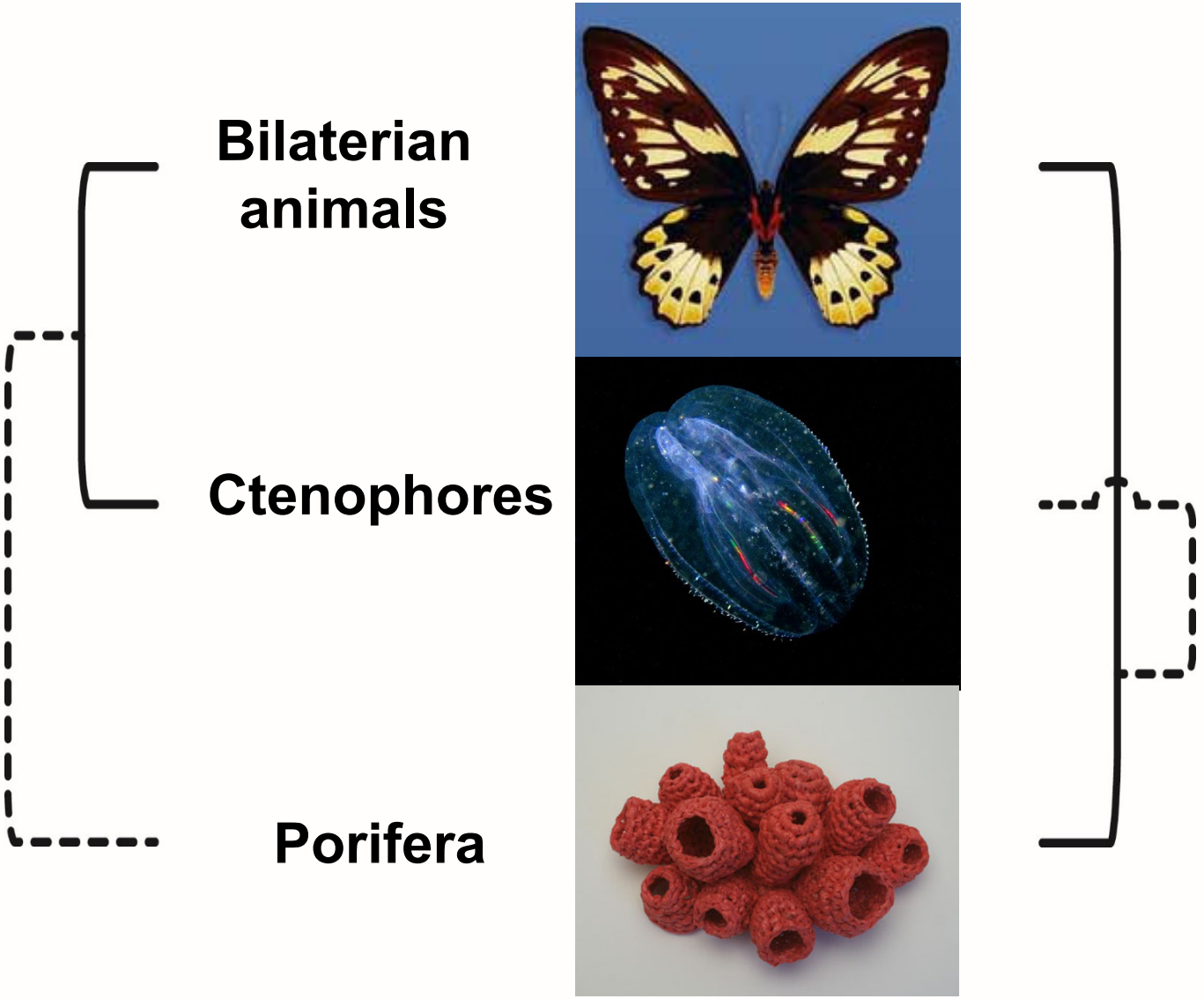
Incongruence in Deep Time is More Challenging



Kocot et al. (2011) Nature

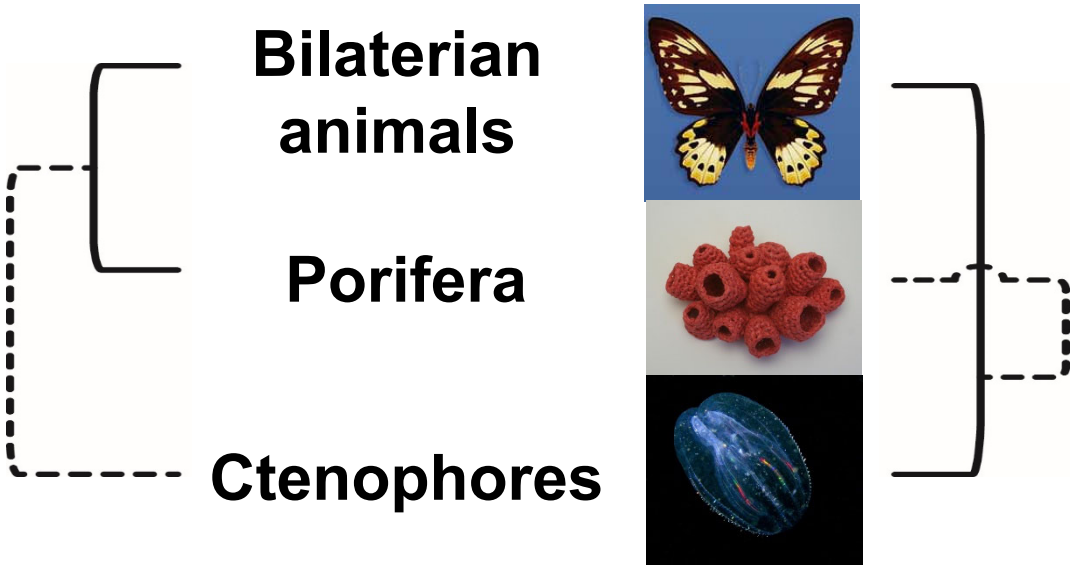
Smith et al. (2011) Nature

Incongruence in Deep Time is More Challenging



Why the disconnect?

Coffee Break

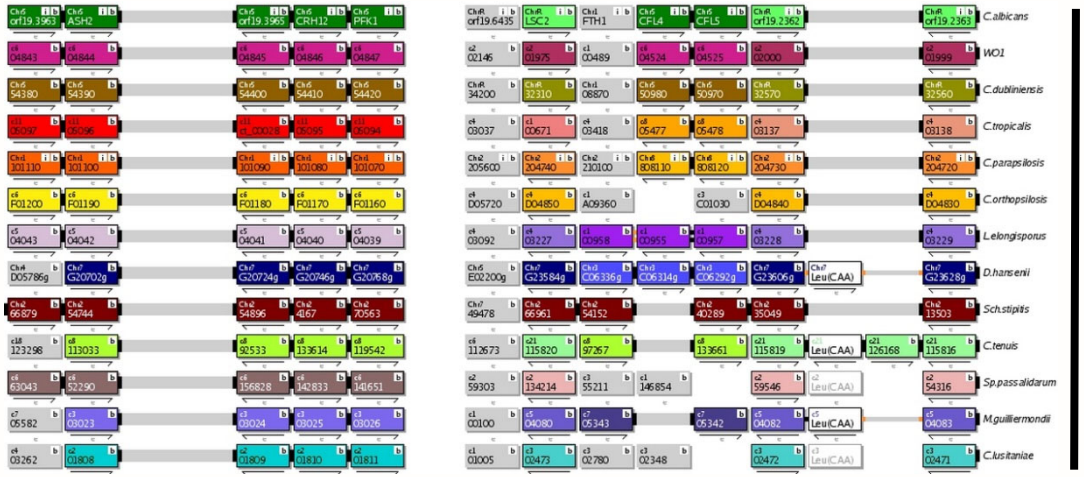


Why are deep branches incongruent? (How) can we resolve them?

An Expanded Yeast Data Matrix

Yeast Gene Order Browser (YGOB)

Candida Gene Order Browser (CGOB)



Saccharomyces **lineage**

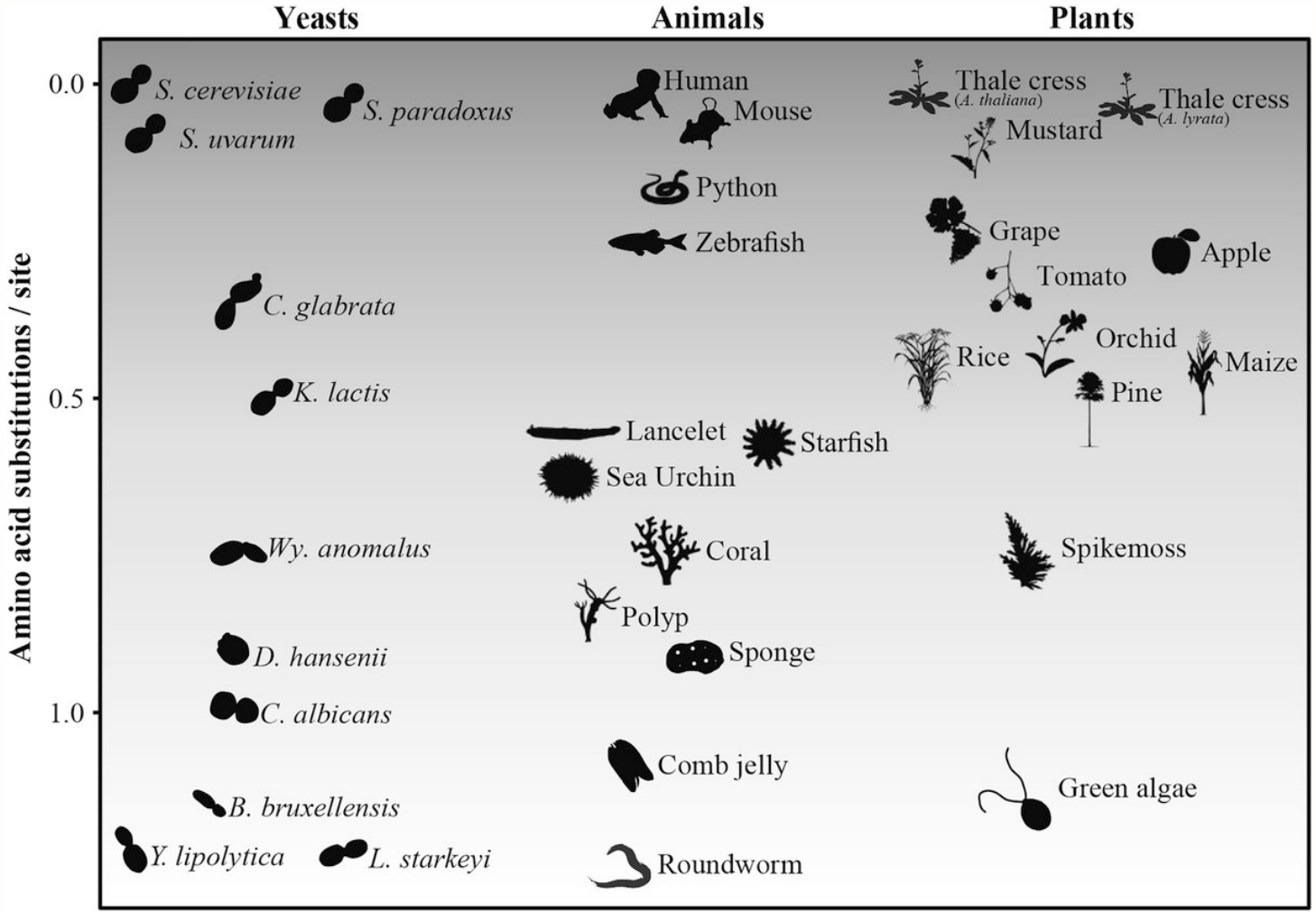
Candida **lineage**

1,070 genes
23 taxa
no missing data

Byrne & Wolfe (2005) Genome Res.

Fitzpatrick et al. (2010) BMC Genom.

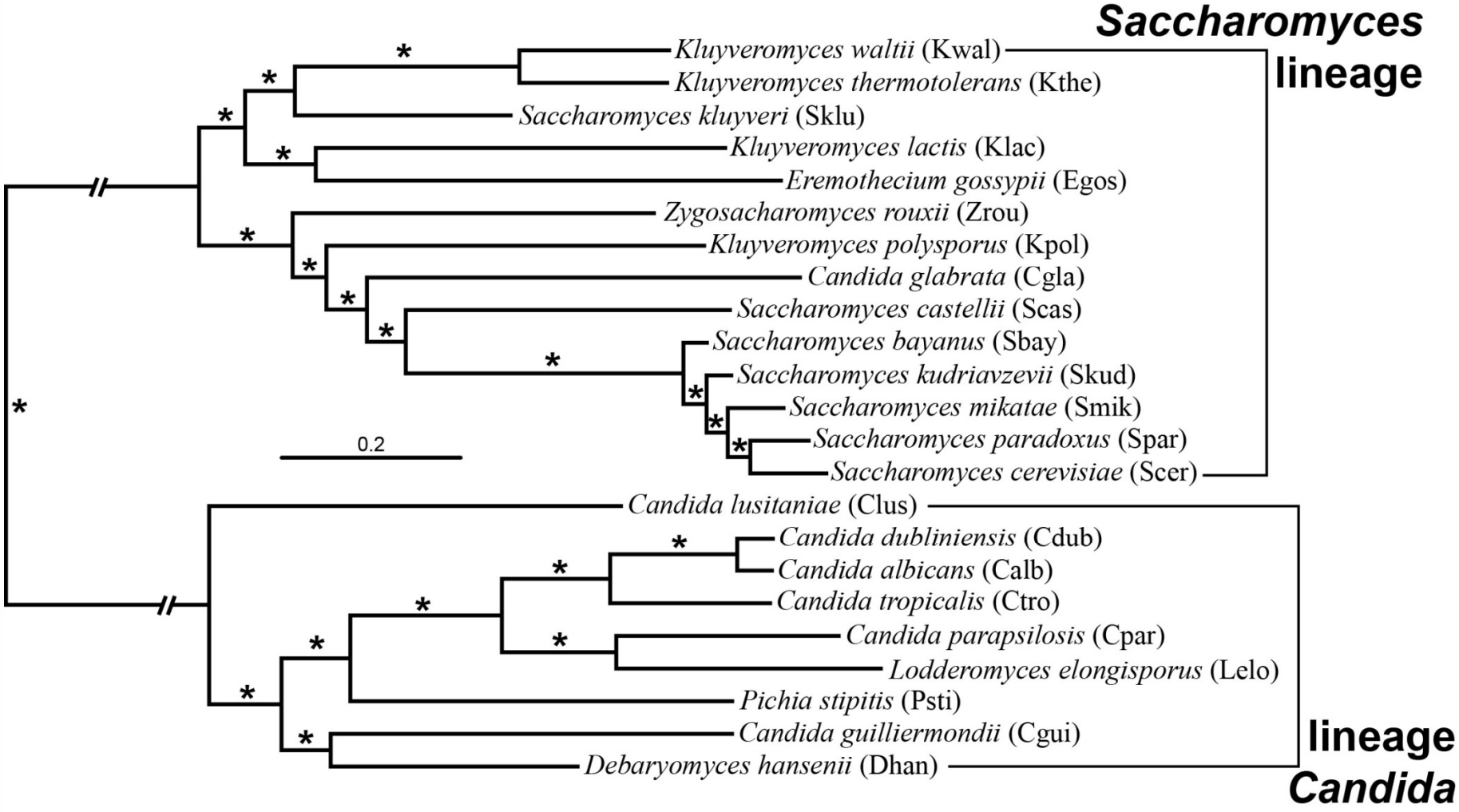
Budding Yeasts Exhibit Striking Genomic Diversity



***Saccharomyces*, *Candida*, *Kluyveromyces*, etc. are all polyphyletic genera**

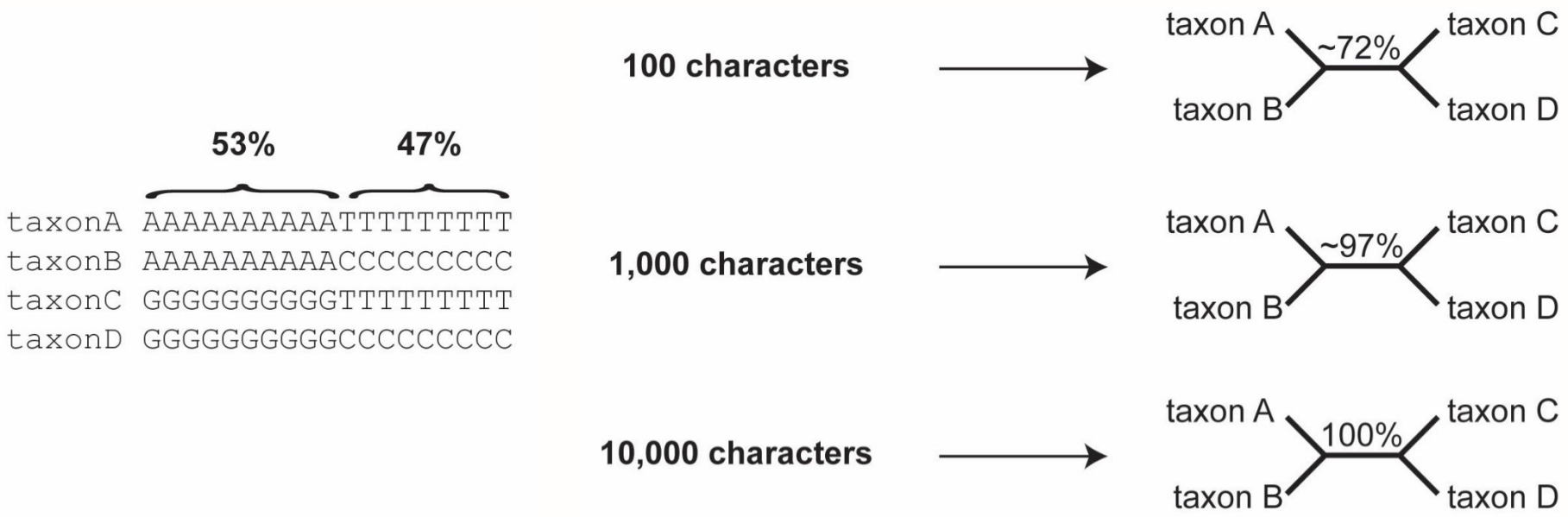
Shen, Opulente, Kominek, Zhou et al. (2018) Cell

Concatenation Yields an Absolutely Supported Phylogeny

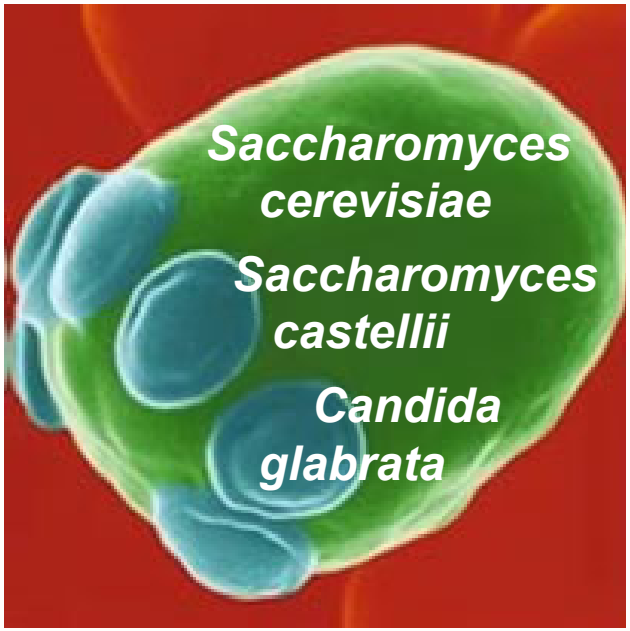


Salichos & Rokas (2013) Nature

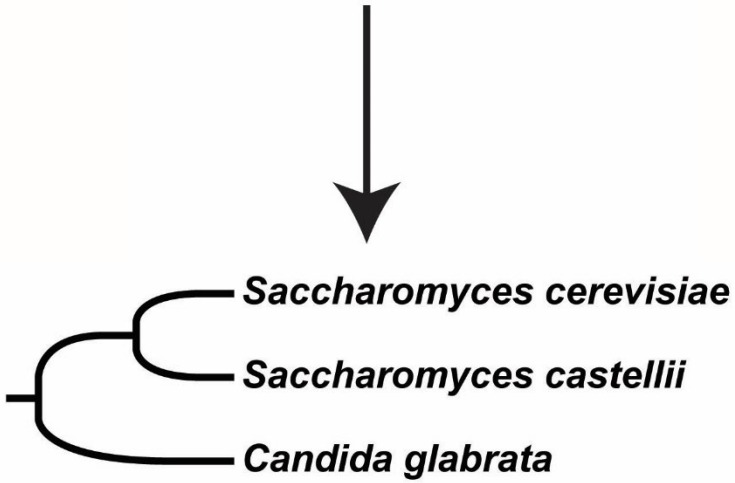
Bootstrap Support is Misleading When Used in Large Datasets



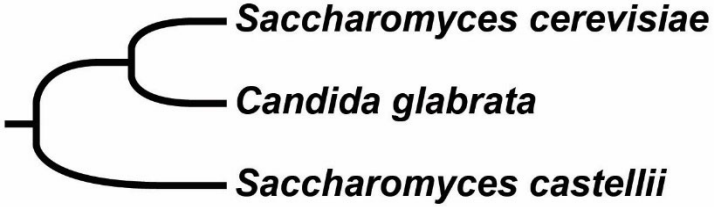
The Concatenation Phylogeny is at Least Partly Wrong



Linear Sequence Data

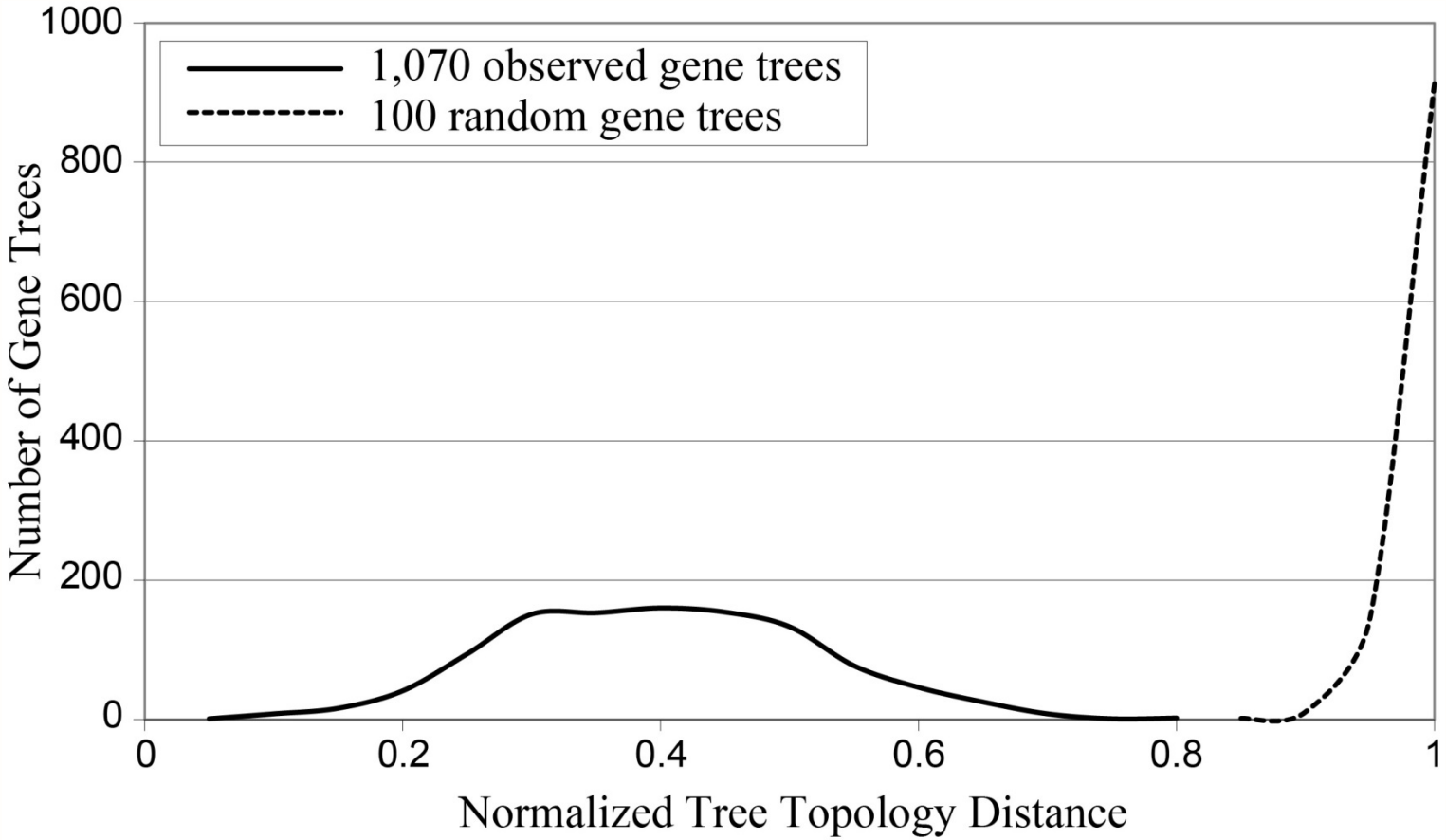


Rare Genomic Changes

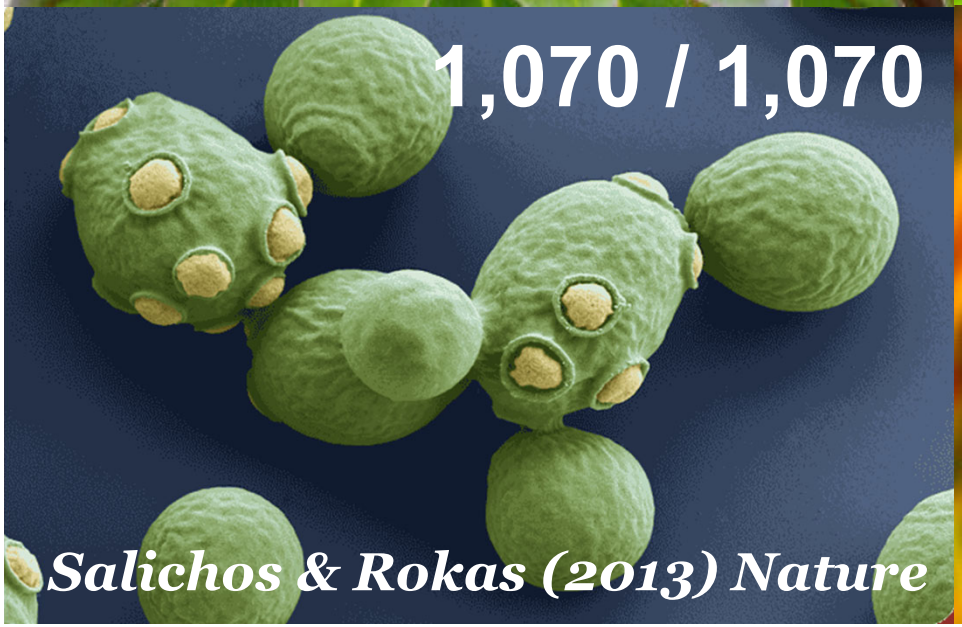


- 5 genomic rearrangements that are uniquely shared by *S. cerevisiae* and *C. glabrata*
- Much higher proportion of shared gene losses in *S. cerevisiae* and *C. glabrata*
- Bias in the placement of *C. glabrata* as an outgroup of *S. cerevisiae* and *S. castellii*

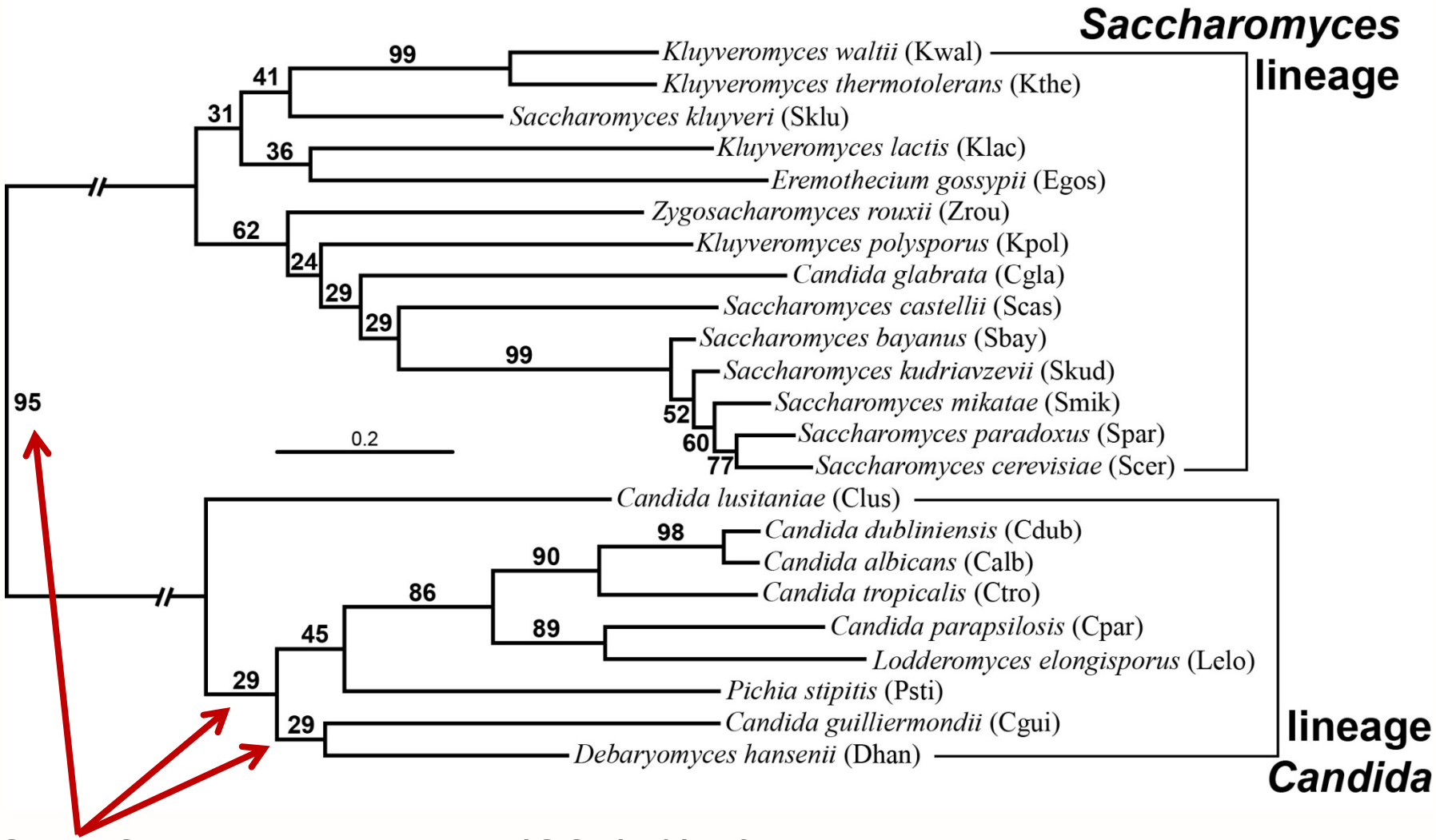
All Gene Trees Differ from the Concatenation Phylogeny



Gene Trees are Incongruent in Most Datasets



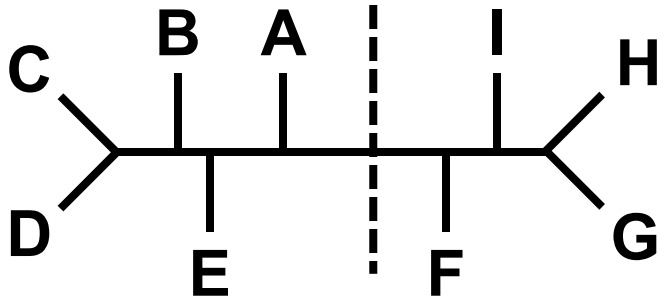
The Yeast Phylogeny Inferred by Majority-Rule Consensus



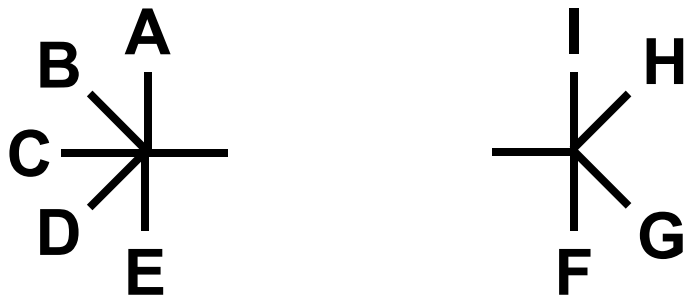
Gene Support Frequency (GSF): % of single gene trees supporting a given internode

Phylogenetic Trees are Sets of Splits / Bipartitions

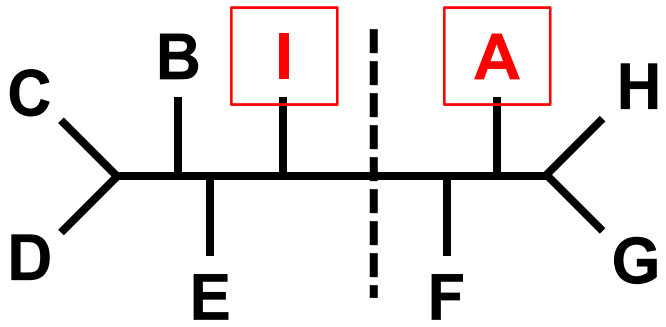
Division

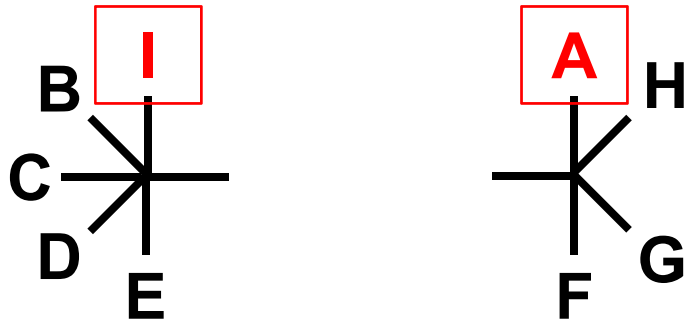


Splits / Bipartitions



Set of splits in reference tree: {A, B, C, D, E} **{F, G, H, I}**





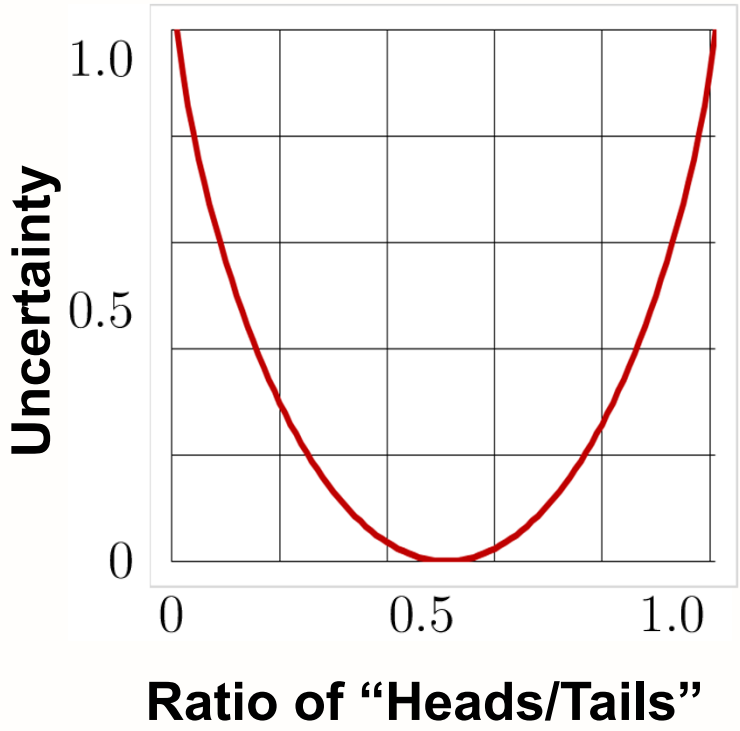
Conflicting **set of splits: {I, B, C, D, E}** **{F, G, H, A}**

Quantifying Incongruence

Internode Certainty (IC): a measure of the support for a given internode by considering its frequency in a given set of trees jointly with that of the most prevalent conflicting internode in the same set of trees

Tree Certainty (TC): the sum of IC across all internodes

IC and TC are implemented in the latest versions of RAxML



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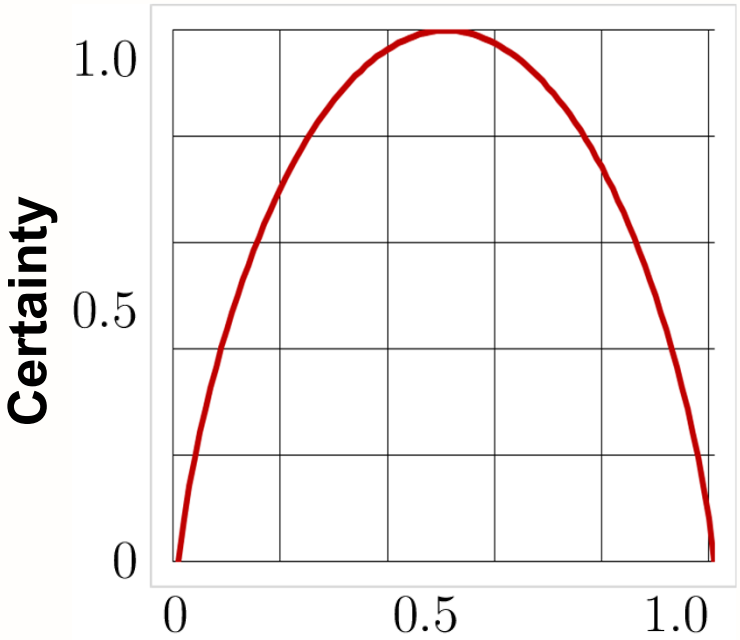
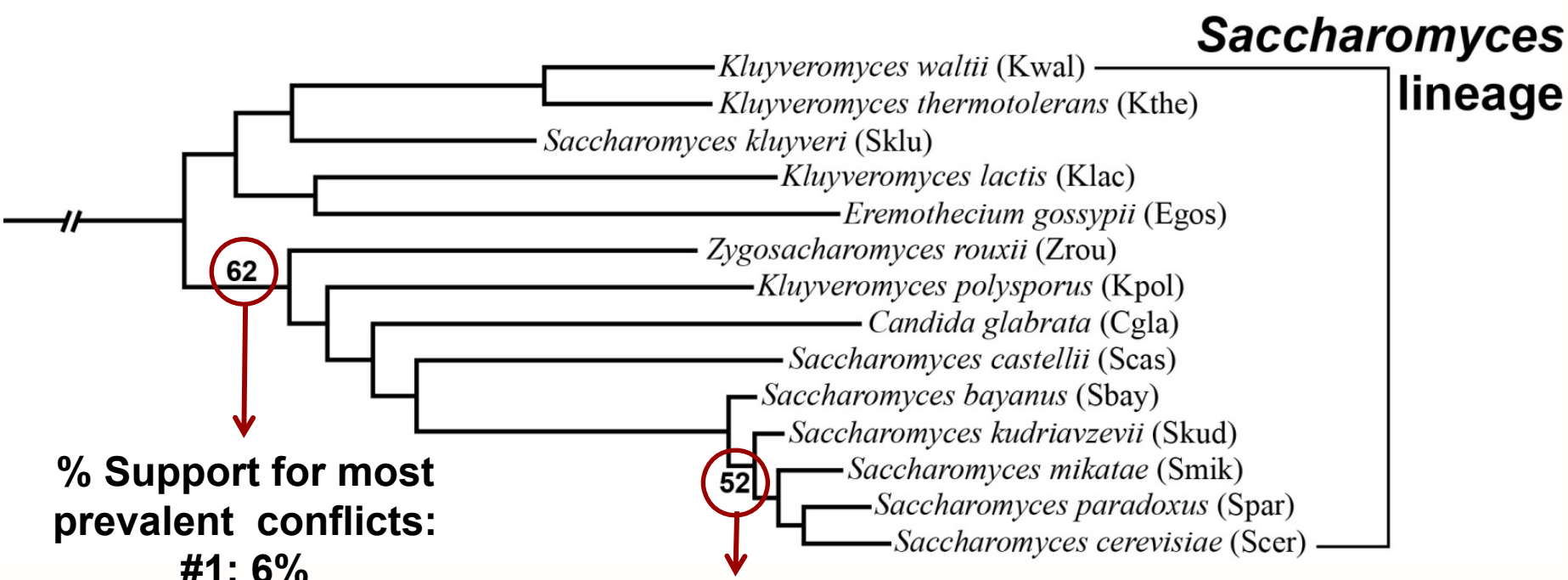


Ratio of Support for Two Conflicting Internodes

IC Can Be More Informative Measure of Internode Support



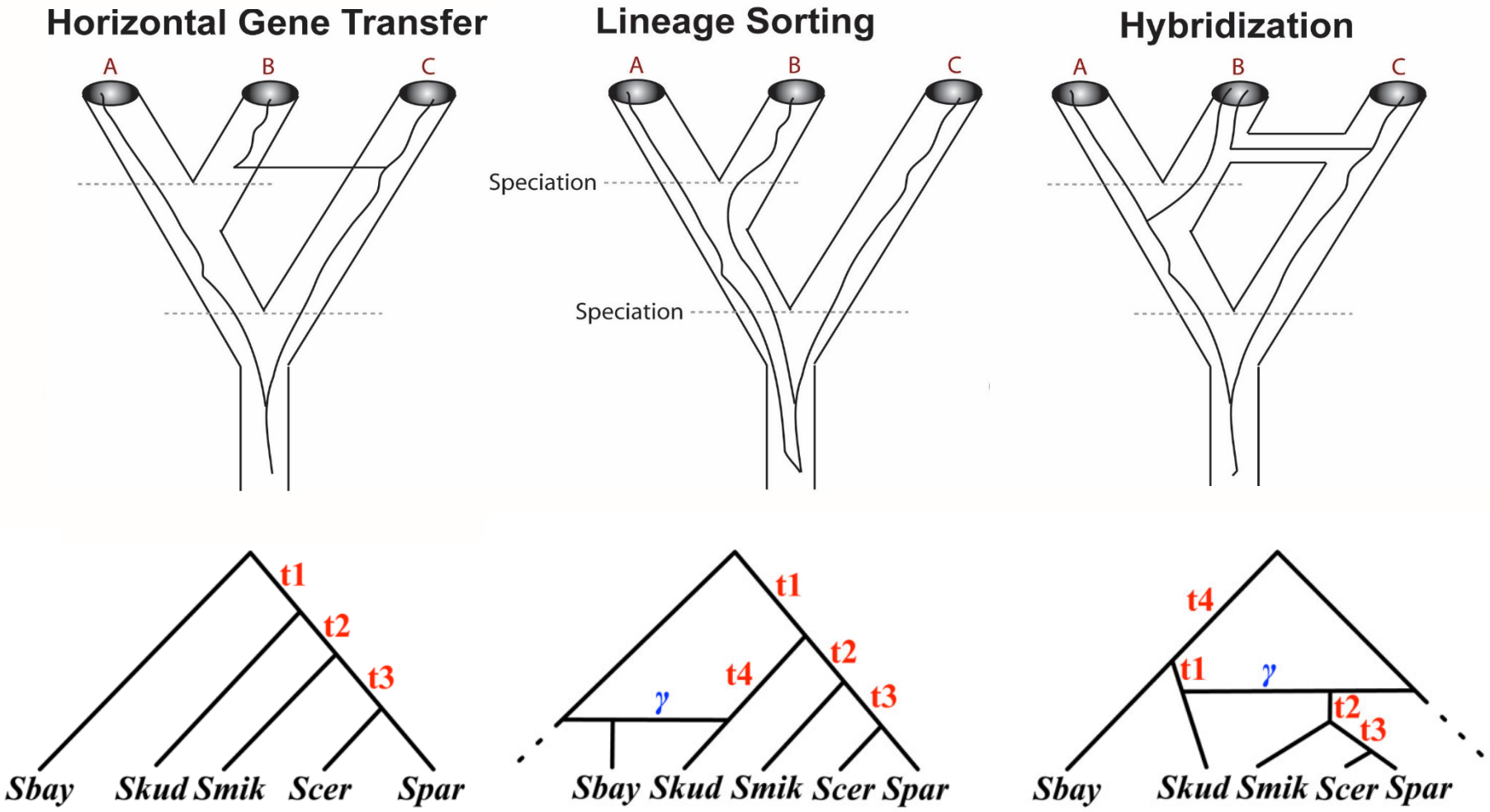
% Support for most prevalent conflicts: #1: 6%

IC value: 0.59

% Support for most prevalent conflicts: #1: 29%

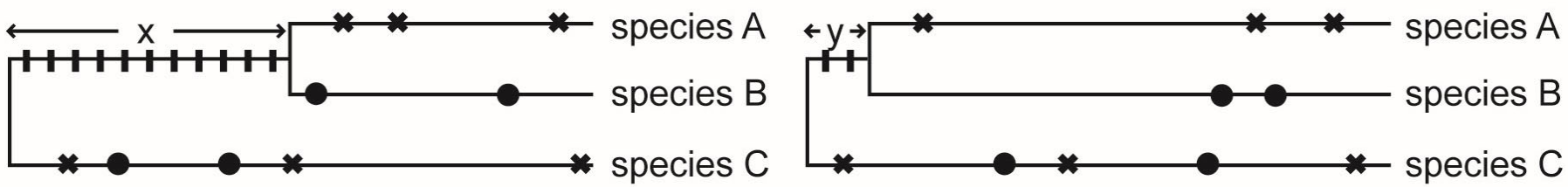
IC value: 0.06

Why So Much Incongruence? Biological Factors

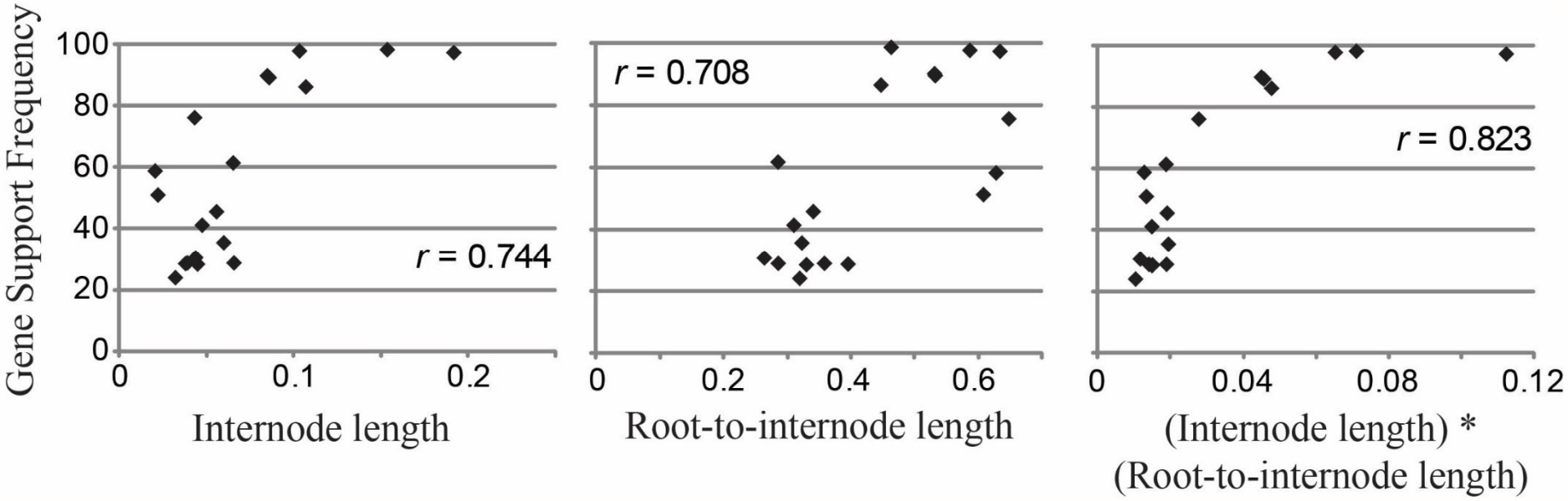


Yu et al. (2012) PLOS Genet.

Why So Much Incongruence? Analytical Factors



Internode length: influences amount of phylogenetic signal (I)
Homoplasy: independent evolution of identical characters (* , ●)



Standard Recipes for Handling Incongruence Didn't Help

Treatment	Tree Certainty	# of Internodes where IC increased | decreased
Default analysis	**8.35**	**n/a**
Removing sites containing gaps		
All sites with gaps excluded	**7.91**	**0 | 7**
Removing fast-evolving or unstable species		
C. lusitaniae	**8.15**	**1 | 2**
C. glabrata	**8.30**	**2 | 2**
E. gossypii*, *C. glabrata*, *K. lactis	**7.88**	**1 | 3**
Selecting genes that recover specific clades		
[*C. tropicalis*, *C. dubliniensis*, *C. albicans*]	**8.62**	**0 | 0**
Selecting the most slow-evolving genes		
100 slowest-evolving genes	**6.76**	**2 | 9**

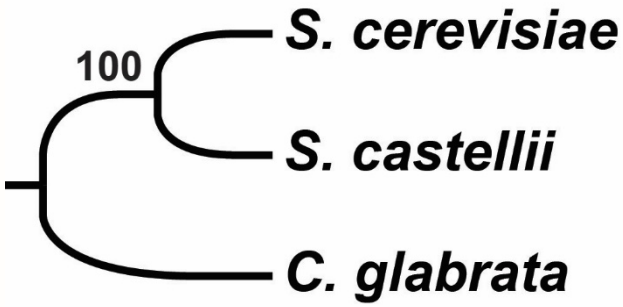
Salichos & Rokas (2013) Nature


WHAT CAN I DO?!!

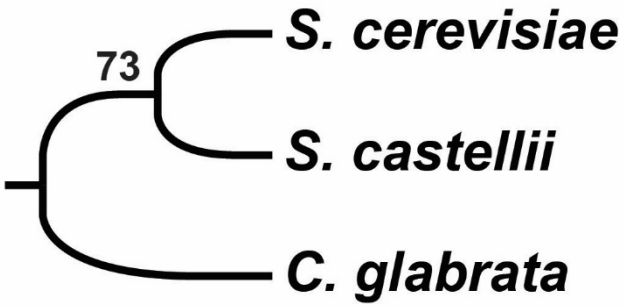
What Do We Do Then?

Treatment	Tree Certainty	# of Internodes where IC increased | decreased
Default analysis	**8.35**	**n/a**
Selecting genes whose bootstrap consensus trees have high average support		
All genes with average BS ≥ 60%	**8.59**	**4 | 0**
All genes with average BS ≥ 70%	**9.18**	**14 | 0**
All genes with average BS ≥ 80%	**9.92**	**15 | 0**

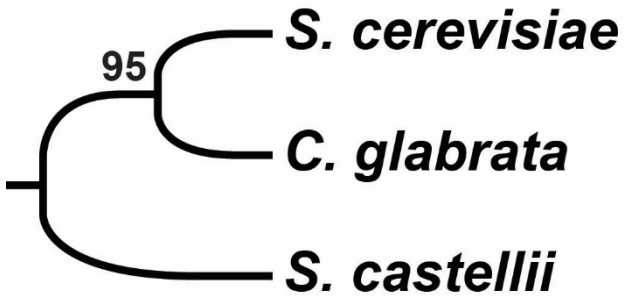
average BS ≥60%



average BS ≥70%

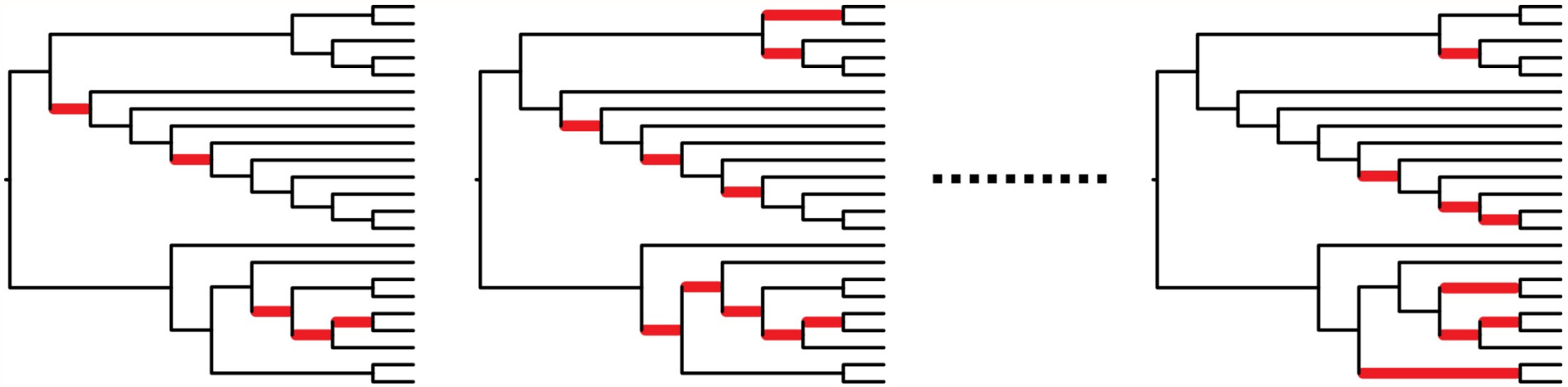


average BS ≥80%



Salichos & Rokas (2013) Nature

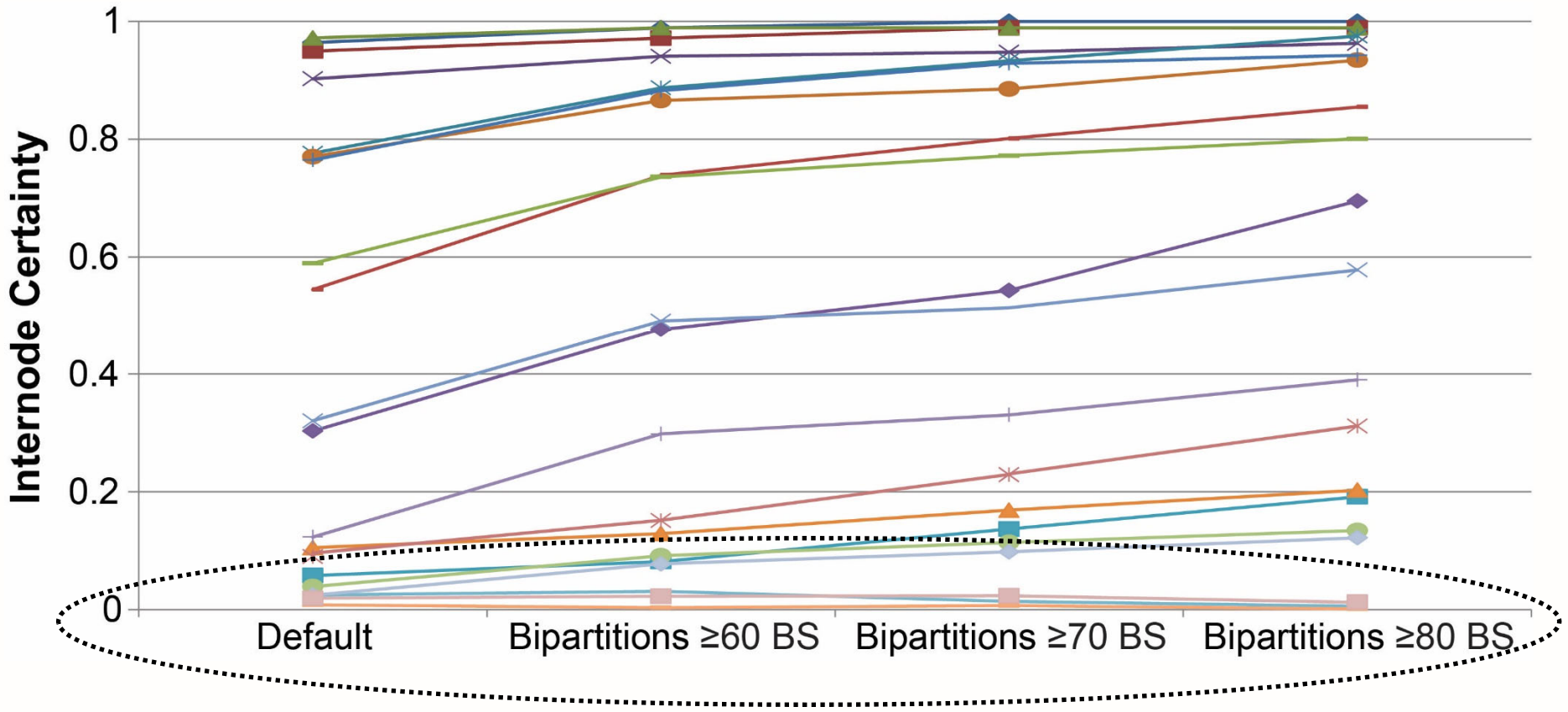
Selecting Specific Bipartitions Dramatically Improves Phylogeny



Treatment	Tree Certainty	# of Internodes where IC increased | decreased
Default analysis	**8.35**	**n/a**
Selecting genes whose bootstrap consensus trees have high average support		
All bipartitions with BS ≥ 60%	**10.11**	**14 | 0**
All bipartitions with BS ≥ 70%	**10.70**	**16 | 0**
All bipartitions with BS ≥ 80%	**11.32**	**15 | 0**

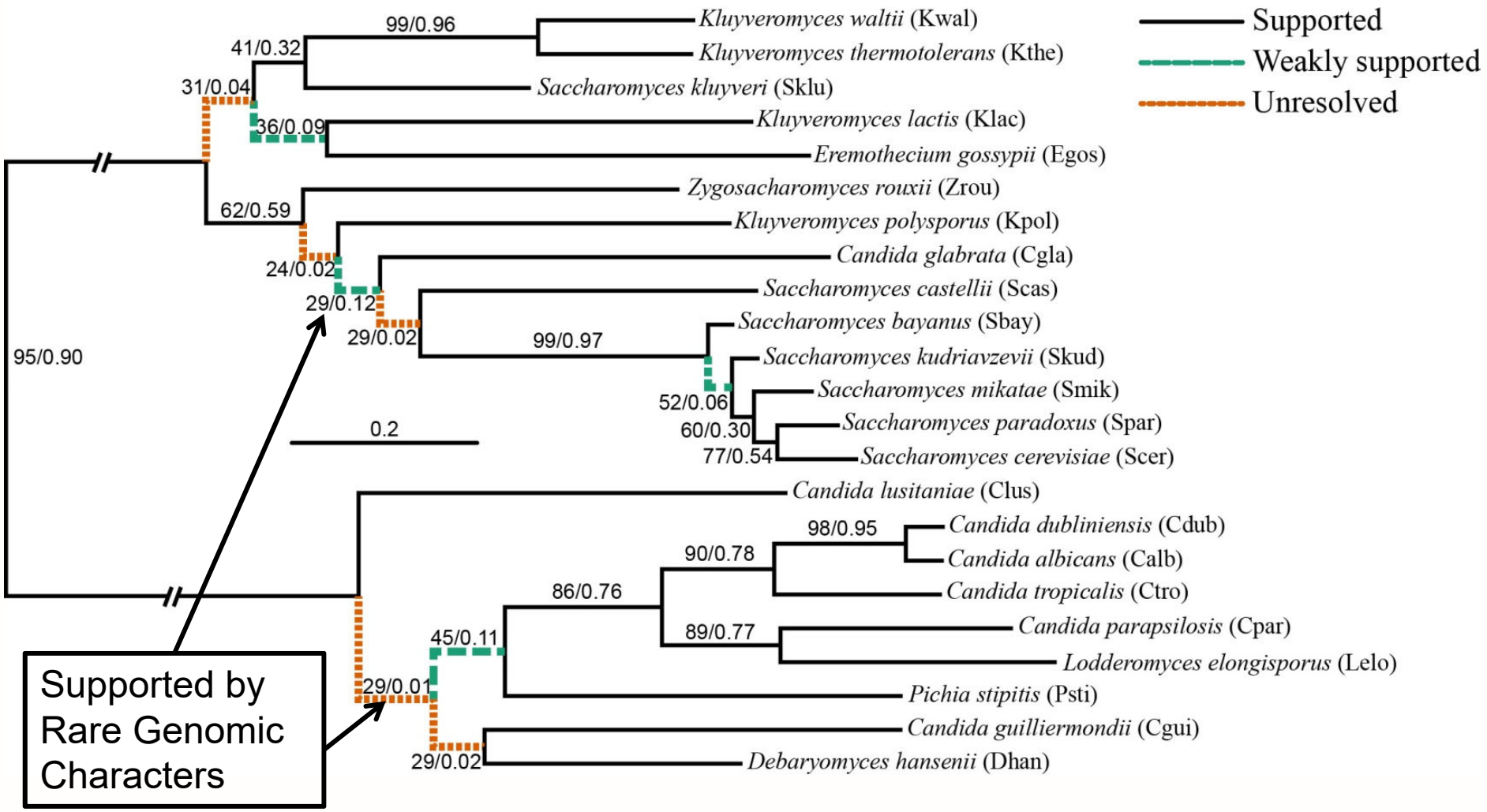
Salichos & Rokas (2013) Nature

Least Supported Internodes Harbor the Most Conflict



Salichos & Rokas (2013) Nature

The Status of the Yeast Phylogeny



Gene Support Frequency / Internode Certainty

Similar Results in Other Lineages

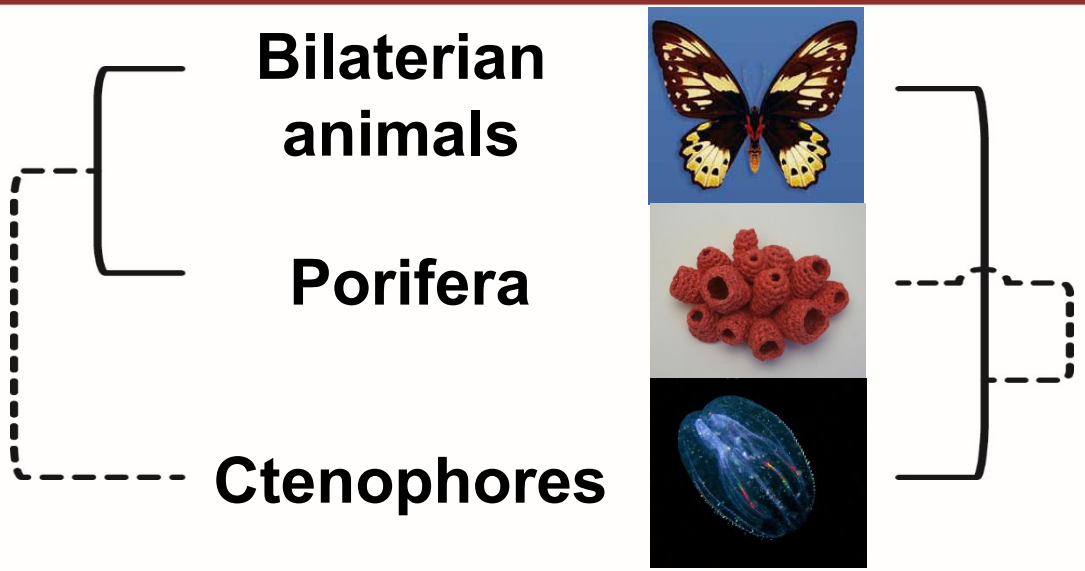
Vertebrates
(1,086 genes, 18 taxa)

Animals
(225 genes, 21 taxa)

Mosquitoes
(2,007 genes, 20 taxa)

Salichos & Rokas (2013) Nature; Wang et al. (2015) Genome Biol. Evol.

Incongruence in Phylogenomic Datasets



These debates concern internodes that are poorly supported by individual gene trees

What is the phylogenetic signal in branches of the tree of life that are challenging to resolve?

Definitions of Phylogenetic Signal

A measure of the statistical dependence among species' trait values due to their phylogenetic relationships / the tendency of related species to resemble each other more than species drawn at random from the same tree

Revell et al. (2008) Syst. Biol.
Münkemüller et al. (2012) Methods Ecol. Evol.

The amount of support for a particular topology, e.g., the relative number of resolved internodes in a consensus tree

Sanderson (2008) Science

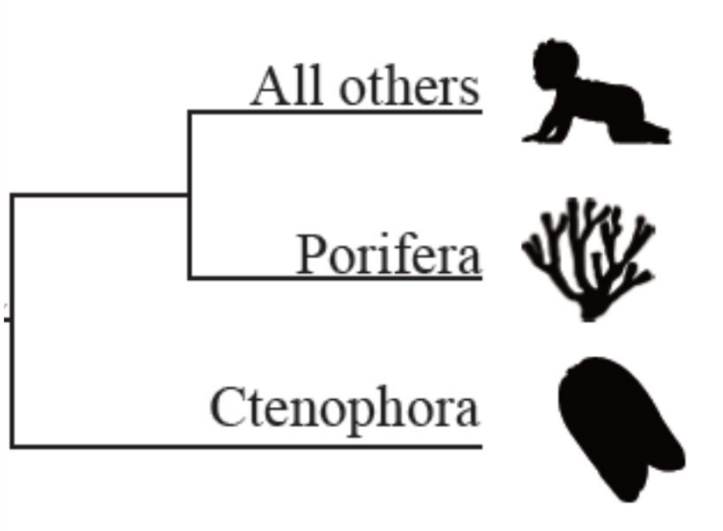
A measure of the substitutions occurring along a given branch of the evolutionary tree. In parsimony methods, the signal is encoded in shared derived characters. In probabilistic methods, the amount of phylogenetic signal actually extracted from a given dataset depends on the model and is expected to increase with the fit of the model to the data

Philippe et al. (2011) PLoS Biol.
Townsend et al. (2012) Syst. Biol.

Our Definition

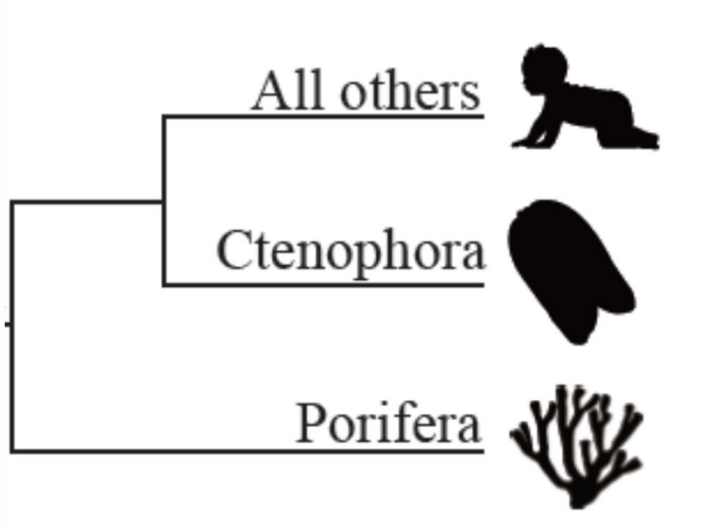
Maximum Likelihood tree

(T1)



Conflicting tree

(T2)

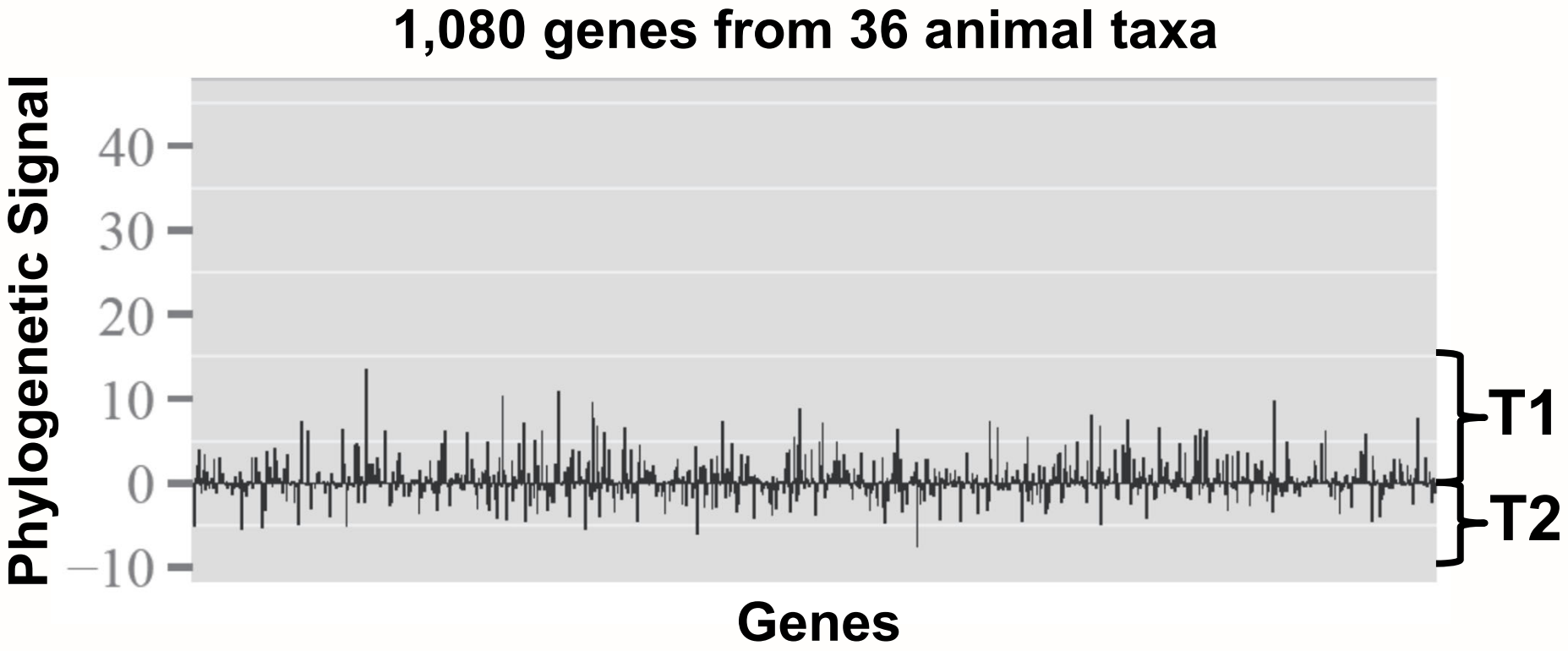


$$\ln(T_1|X_i) = -100$$

$$\ln(T_2|X_i) = -150$$

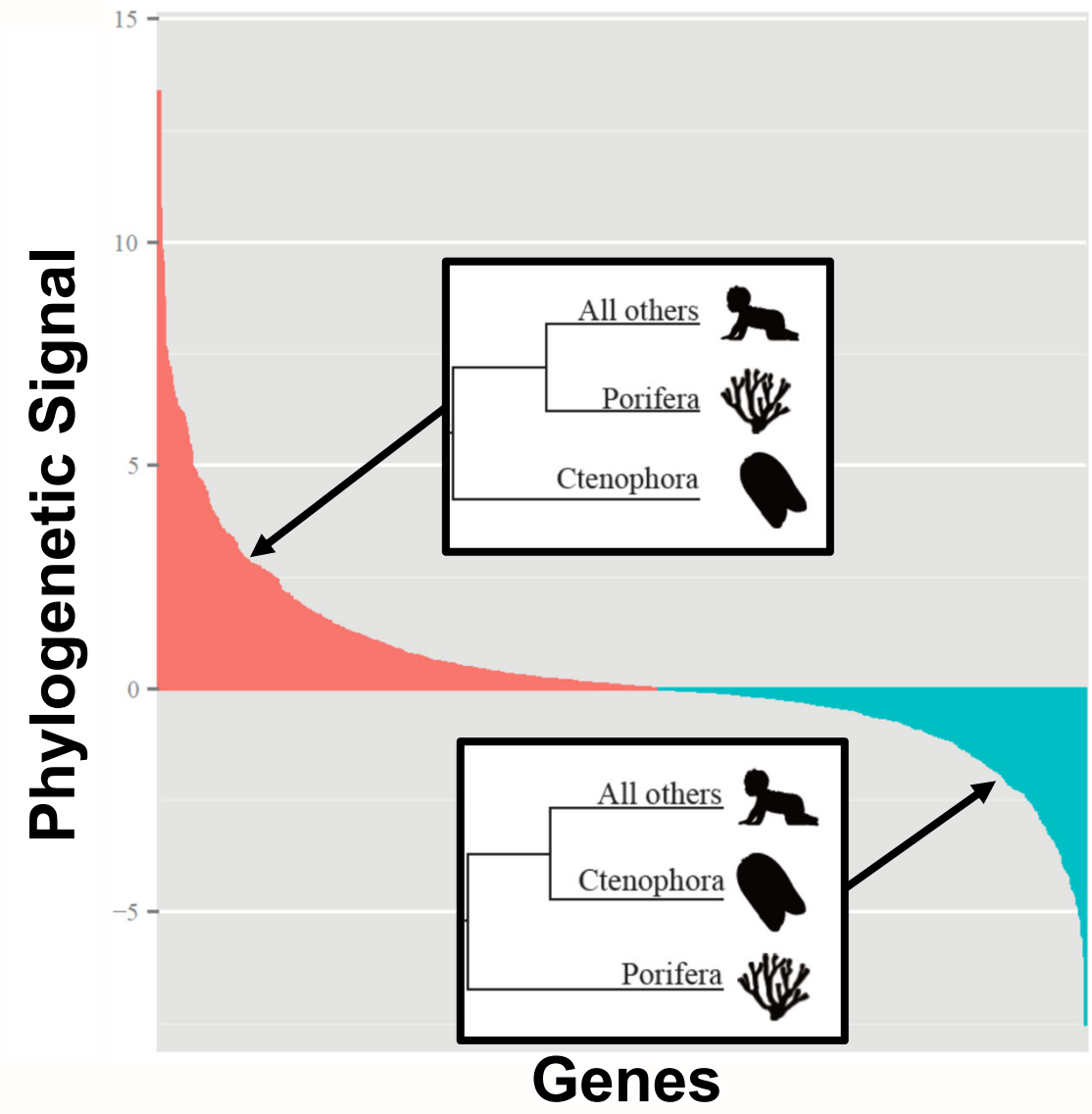
$$Phylogenetic\ Signal = -(\ln(T_1|X_i) - \ln(T_2|X_i))$$

Signal of the Genes in a Phylogenomic Data Matrix



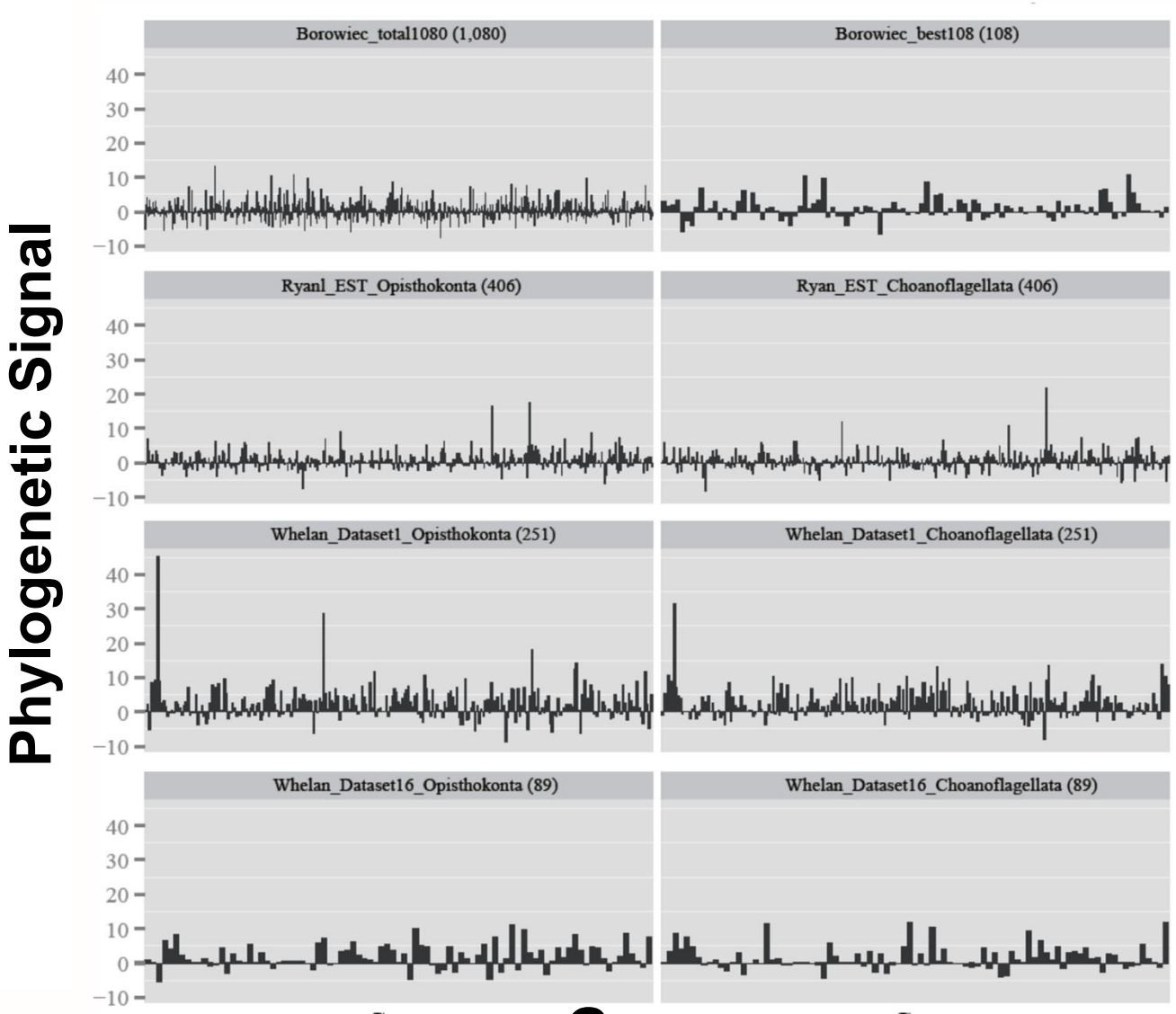
Shen et al. (2017) Nature Ecol. Evol.; data from Borowiec et al. (2015) BMC Genomics

Signal of the Genes in a Phylogenomic Data Matrix



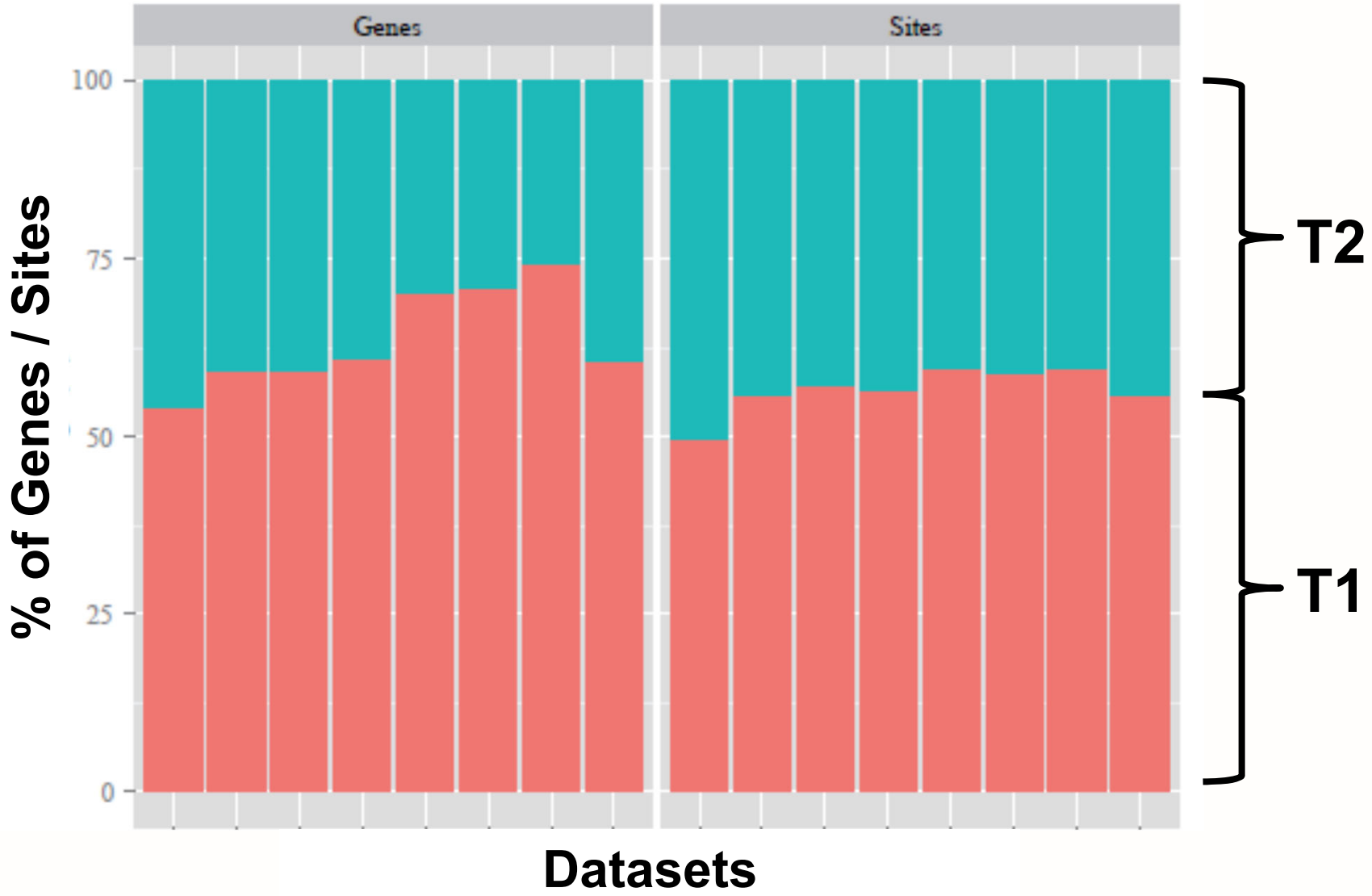
Shen et al. (2017) Nature Ecol. Evol.

Signal of the Genes in Multiple Phylogenomic Data Matrices



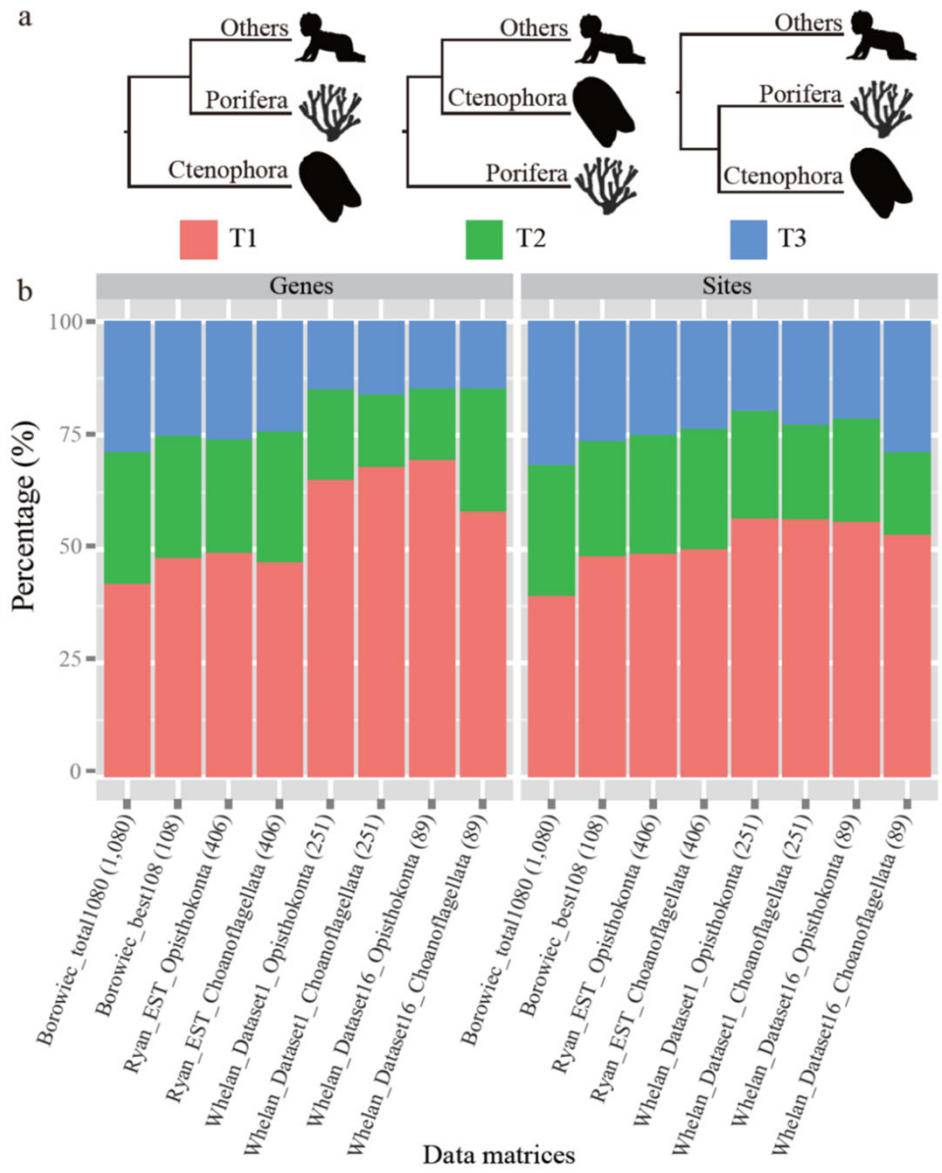
Shen et al. (2017) Nature Ecol. Evol.

Summarizing Phylogenetic Signal Across Genes and Sites



Shen et al. (2017) Nature Ecol. Evol.

Summarizing the Signal Across All 3 Possible Topologies



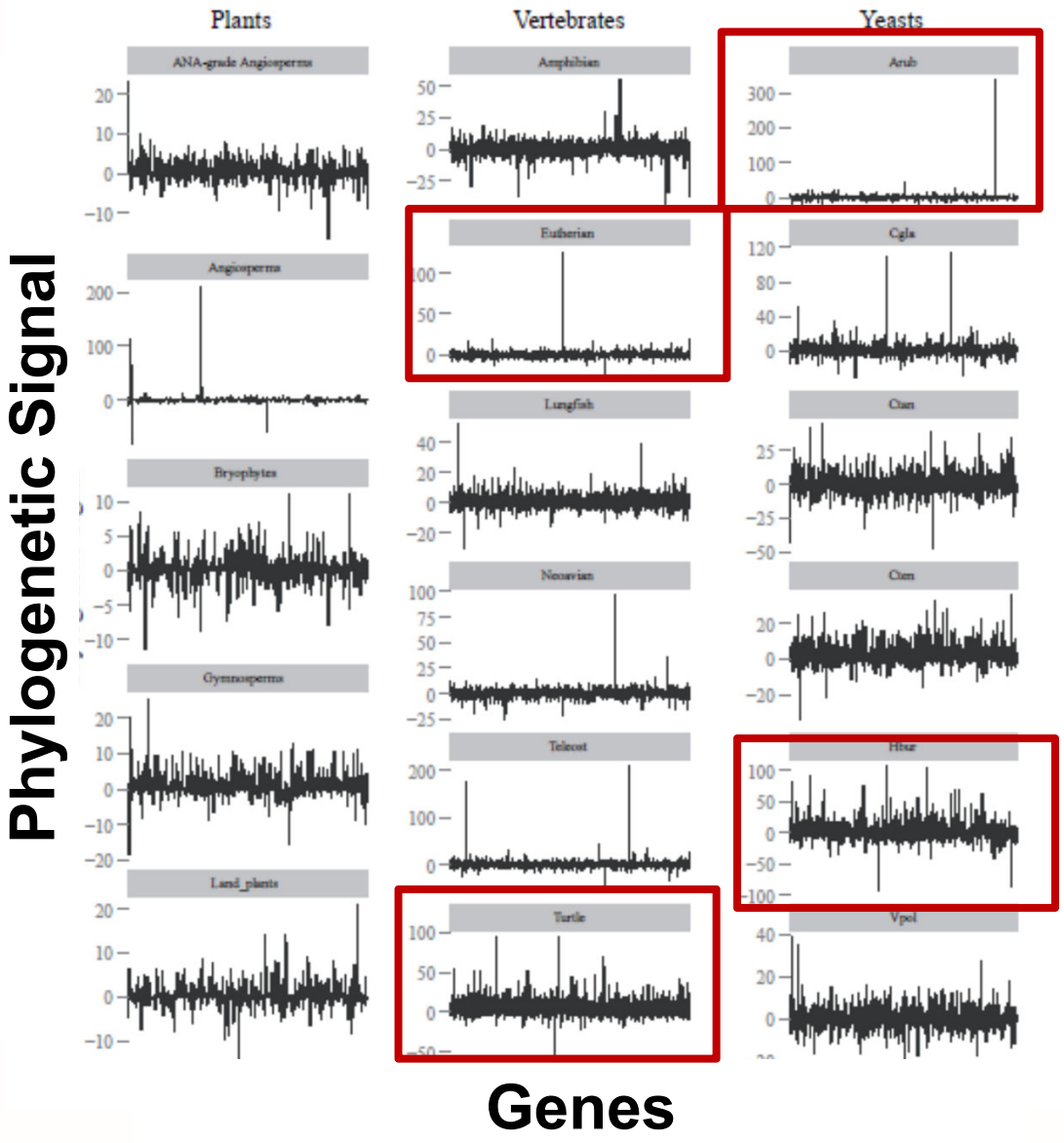
Shen et al. (2017) Nature Ecol. Evol.

Testing Several Contentious Branches of the Tree of Life

Clade	ML Tree (T1)	Conflicting Tree (T2)
Plants	*Amborella* as sister to all other flowering plants	*Amborella* + *Nuphar* as sister to all other flowering plants
	Magnoliids as sister to Eudicots + Chloranthales	Eudicots as sister to Magnoliids + Chloranthales
	Hornworts as sister to all other land plants, followed by a mosses + liverworts clade	Hornworts as sister to a mosses + liverworts clade
	Gnetales as sister to the Pinaceae, nested within the Coniferales	Gnetales as sister to the Coniferales
	Zygnematophyceae as sister to all land plants	Charales as sister to all land plants
Vertebrates	Gymnophiona as sister to all other amphibians	Anura as sister to all other amphibians
	Atlantogenata (Afrotheria + Xenarthra) as sister to all other placental mammals	Afrotheria as sister to all other placental mammals
	Lungfishes as sister to all tetrapods	Lungfishes + coelacanths as sister to all tetrapods
	Pigeons as sister to all other Neoaves	Falcons as sister to all other Neoaves
	Elopomorpha + Osteoglossomorpha as sister to all other teleosts	Osteoglossomorpha alone as sister to all other teleosts
	Turtles as sister to archosaurs (birds + crocodiles)	Turtles as sister to crocodiles
Yeasts	Ascoideaceae as sister to Phaffomycetaceae + Saccharomycetaceae	Ascoideaceae as sister to a clade comprising Pichiaceae, Debaryomycetaceae, Phaffomycetaceae, and Saccharomycetaceae
	Candida glabrata rather than *Naumovozyma castellii* as sister to Saccharomyces sensu stricto yeasts	*Naumovozyma castellii* rather than *Candida glabrata* sister to Saccharomyces sensu stricto yeasts
	Hyphopichia burtonii as sister to *Candida auris* + *Metschnikowia bicuspidata*	*Hyphopichia burtonii* as sister to *Debaryomyces hansenii*
	Zygosaccharomyces rouxii as sister to all other yeasts with occurring whole-genome duplication event	*Vanderwaltozyma polyspora* as sister to all other yeast with occurring whole-genome duplication event
	Meyerozyma guilliermondii as sister to *Debaryomyces hansenii*	*Meyerozyma guilliermondii* as sister to *Hyphopichia burtonii* + *Candida auris*
	Candida tanzawaensis as sister to *Pichia stipiti* + *Candida maltosa*	*Pichia stipiti* as sister to *Candida tanzawaensis* + *Candida maltosa*

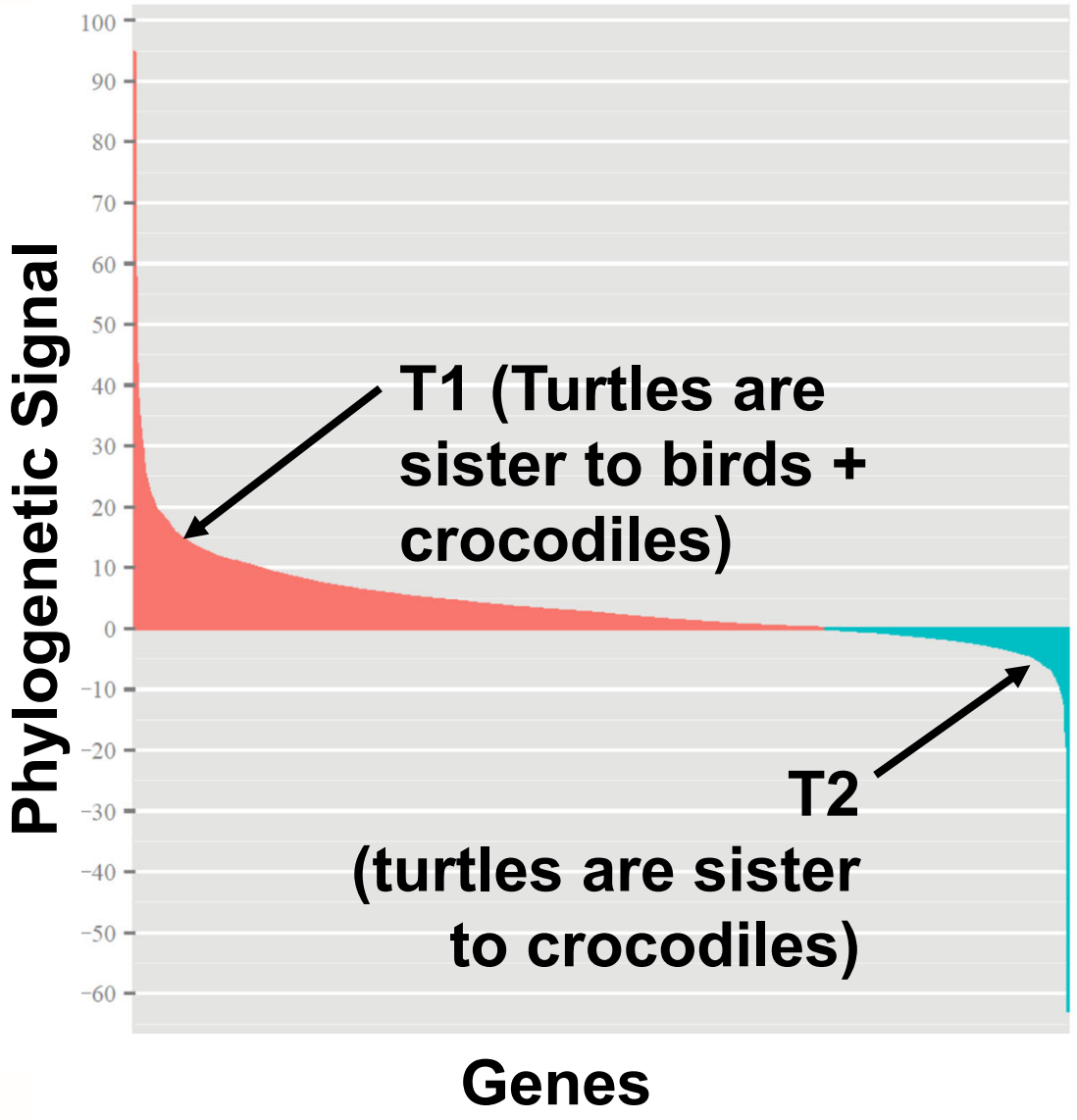
Shen et al. (2017) Nature Ecol. Evol.

Phylogenetic Signal in Contentious Branches of the ToL



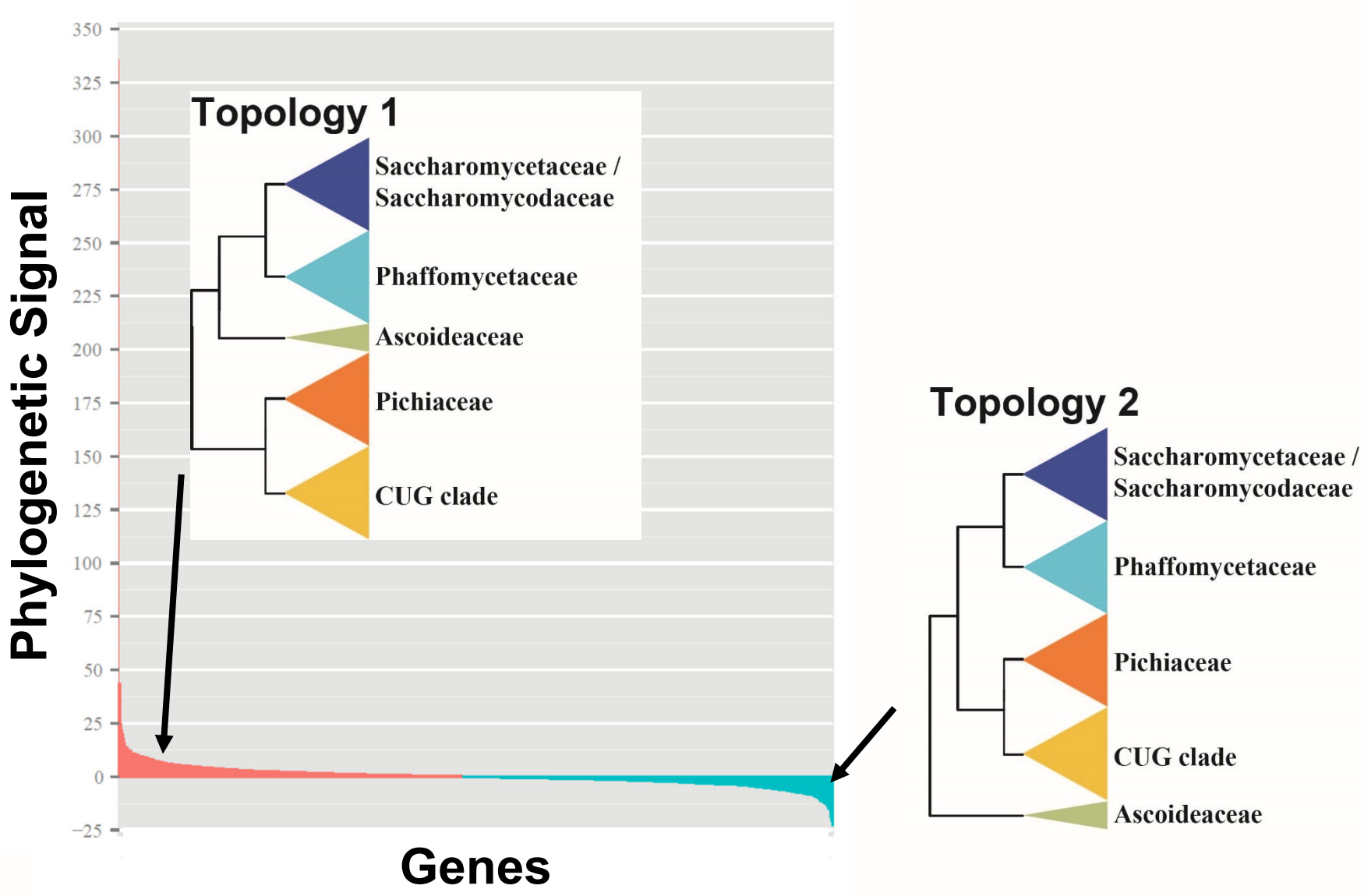
Shen et al. (2017) Nature Ecol. Evol.

The Signal in Some Branches is Very Strong…

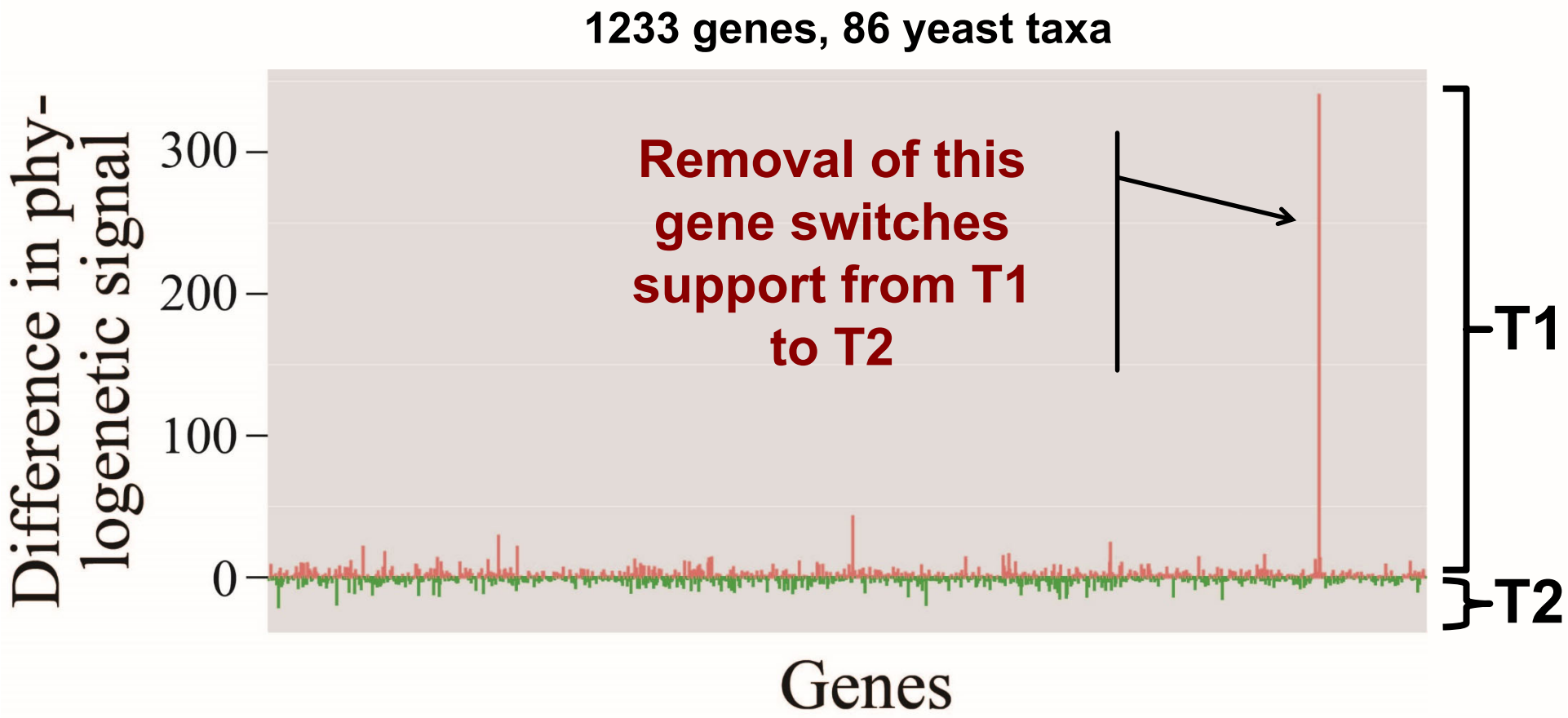


Shen et al. (2017) Nature Ecol. Evol.

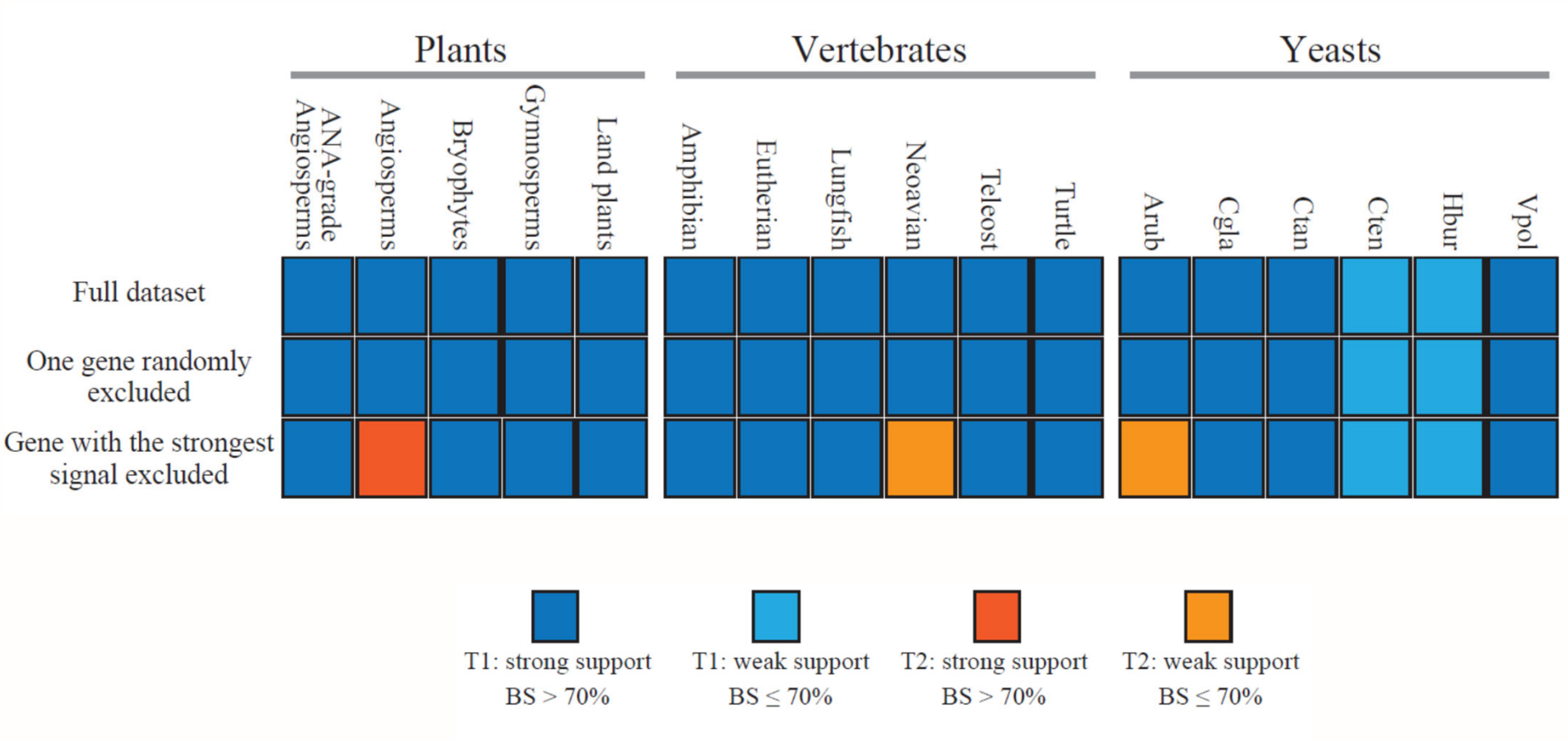
...But in Others It Stems from One or Two Genes



Phylogenetic Signal per Gene for the Two Hypotheses



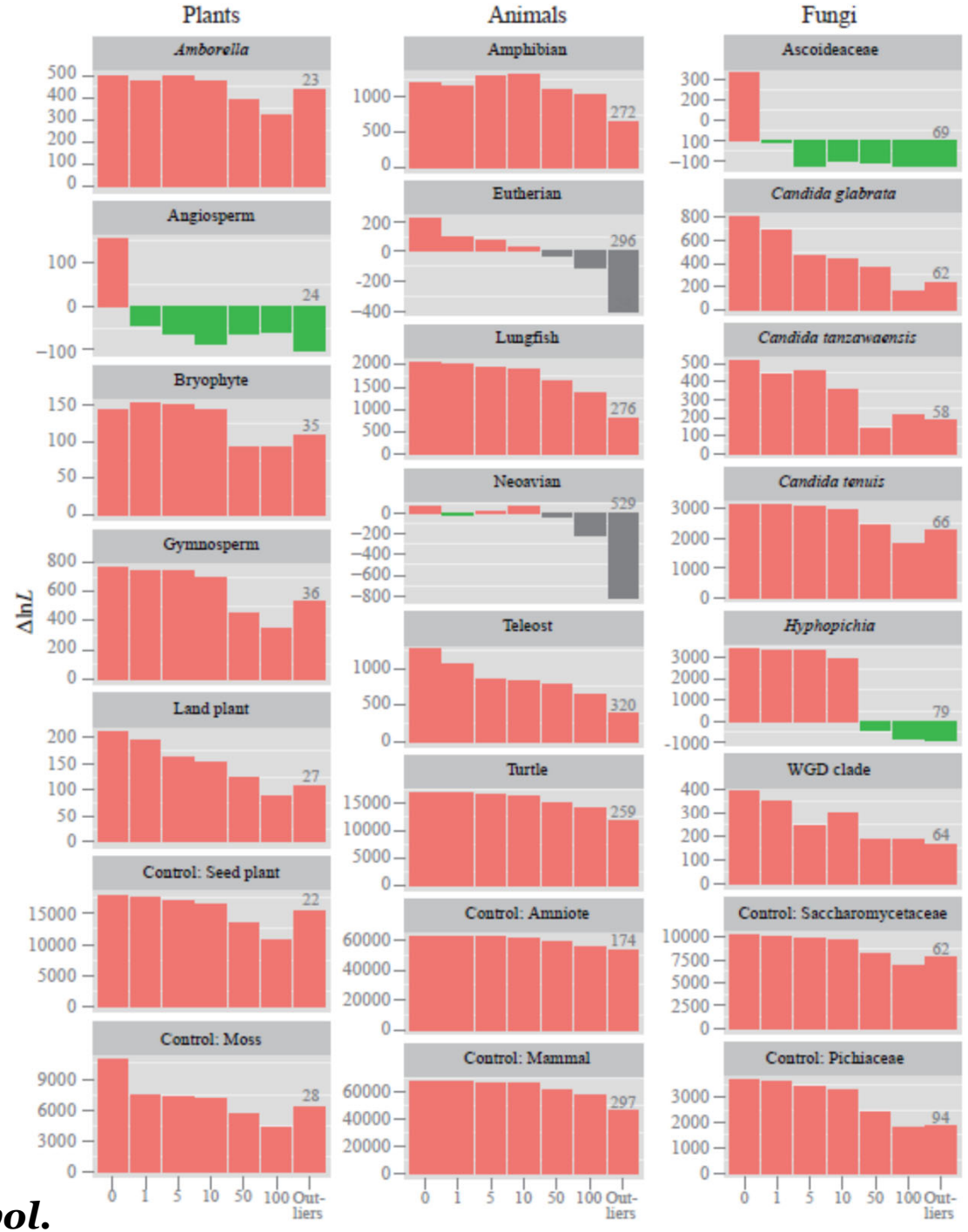
What Happens if we Remove That One Gene?



T1 T2 Others

# of genes excluded

Quantifying the Impact of Removing Opinionated Genes

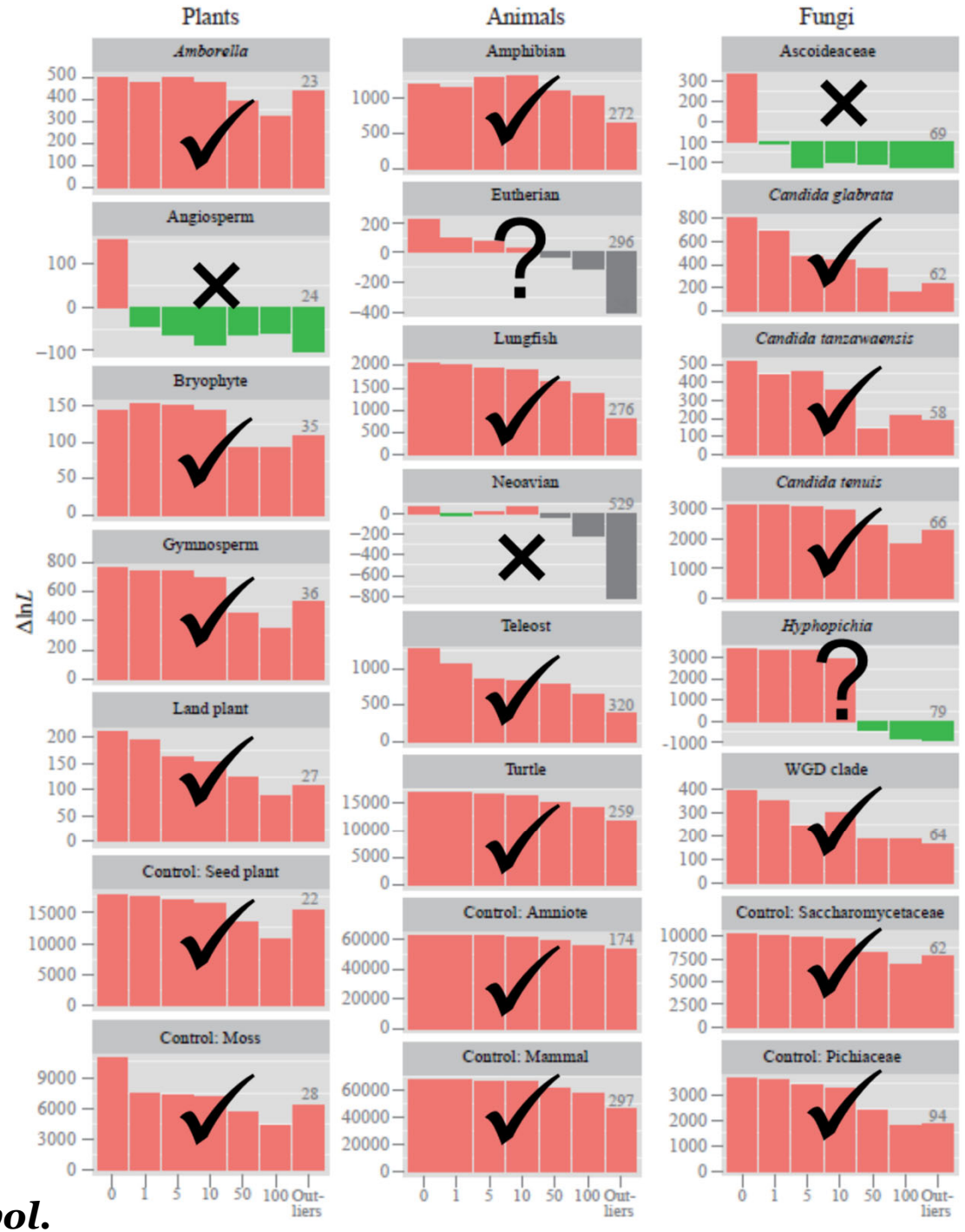


Shen et al. (2017) Nature Ecol. Evol.

T1 T2 Others

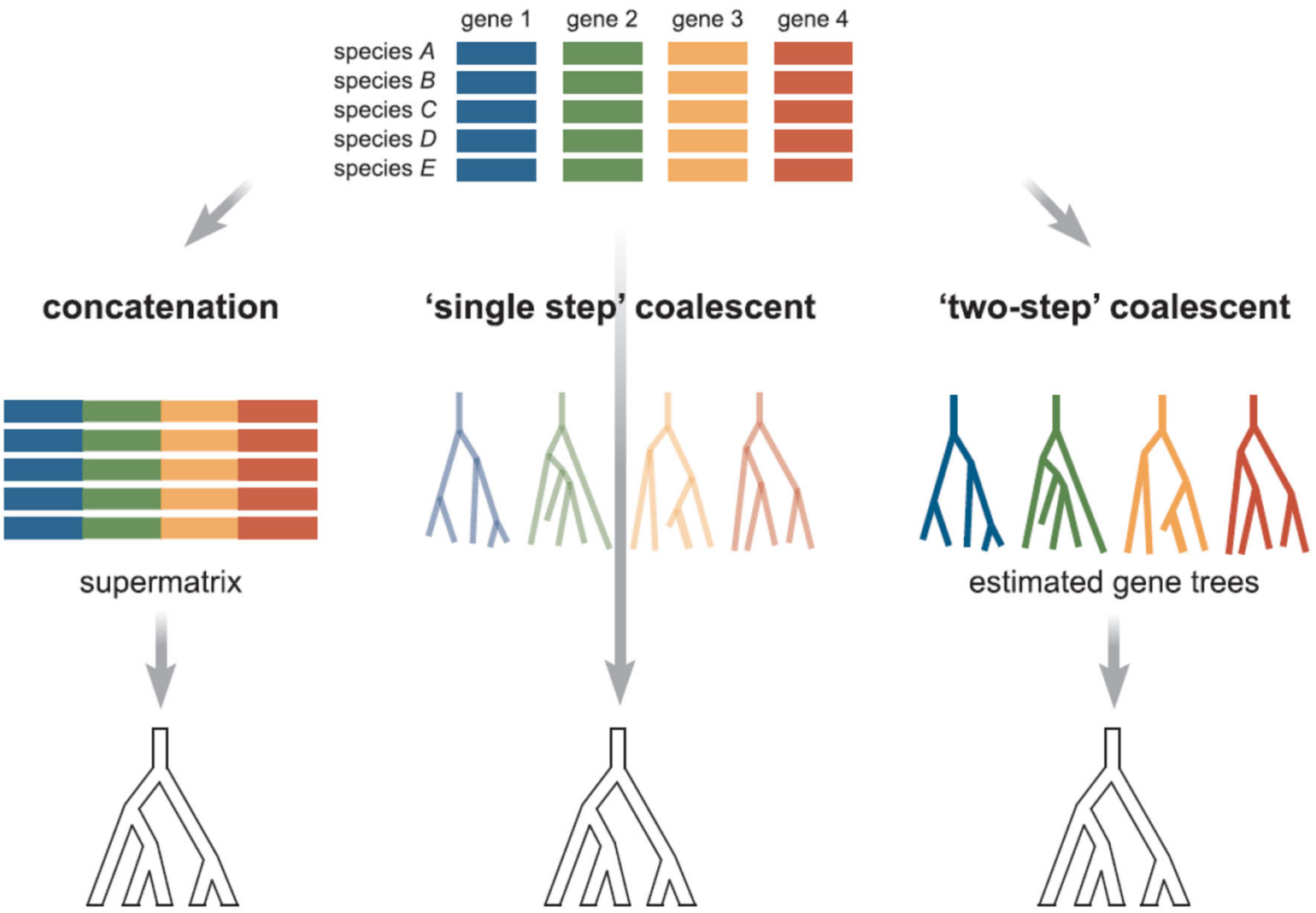
# of genes excluded

Which Branches are Resolved and Which are Unresolved?



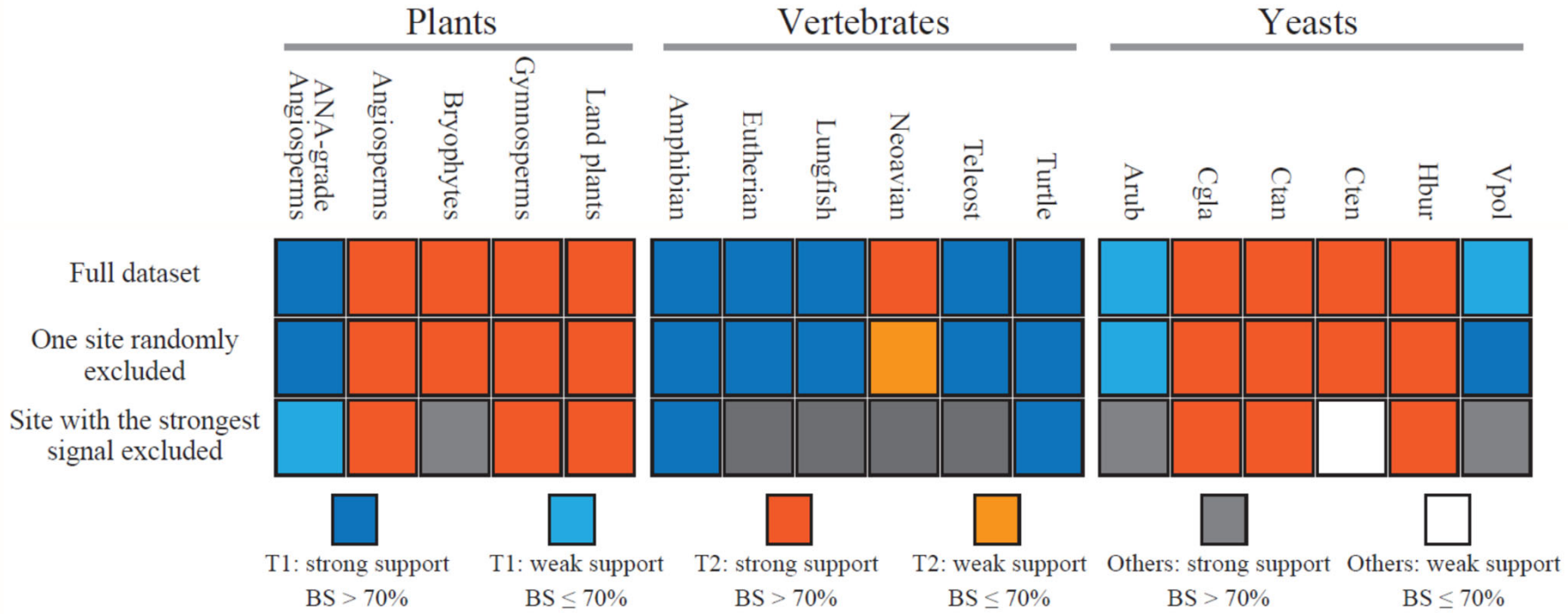
Shen et al. (2017) Nature Ecol. Evol.

Methods for Phylogenomic Inference

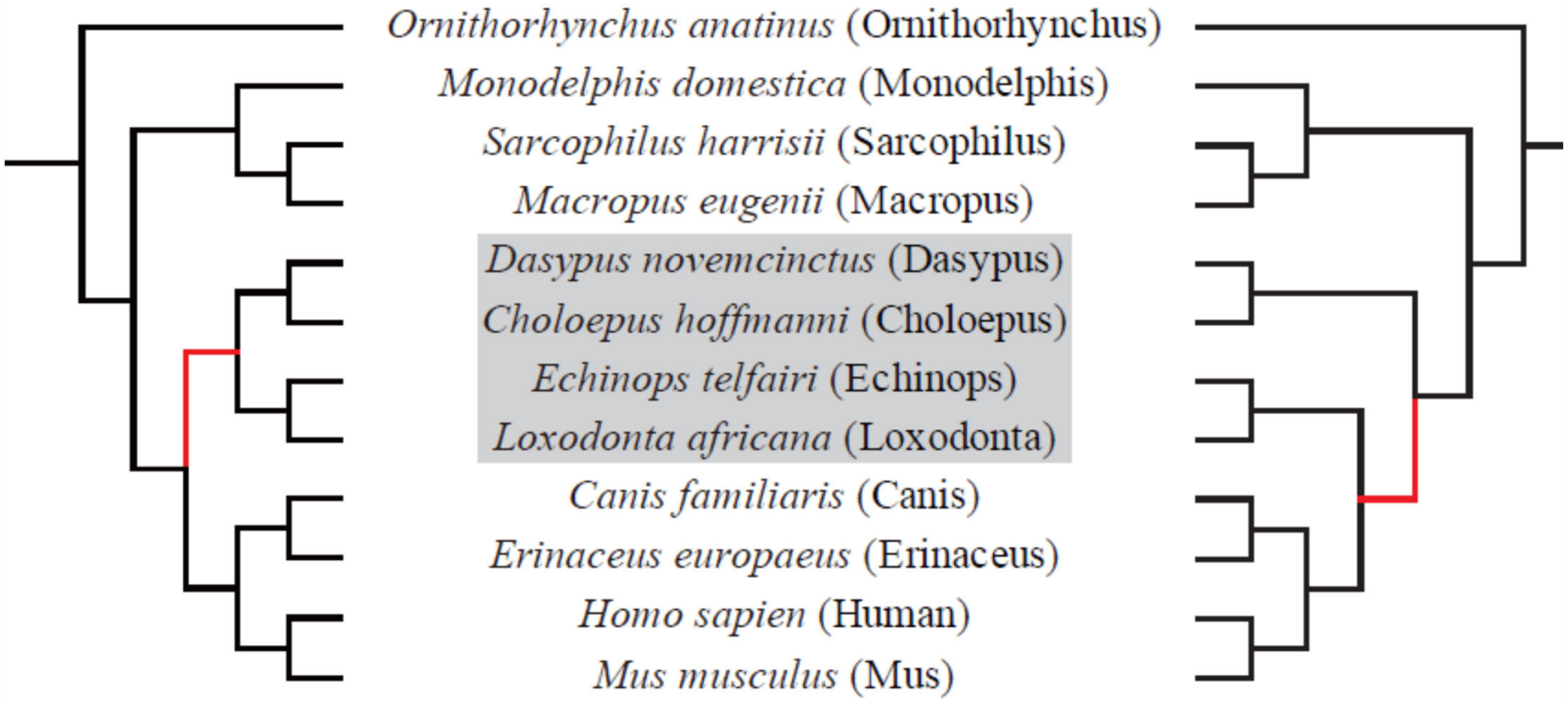


Liu et al. (2015) Ann. N. Y. Acad. Sci.

What Happens if we Remove One Site from Every Gene?



Removing 1 Site Alters the Topology



Shen et al. (2017) Nature Ecol. Evol.

What's Going On?

Explanation #1: Biological factors (parts of the tree of life are bush-like / network-like rather than tree-like)

Explanation #2: Analytical factors (systematic error due to the bad fit of our models to our data)

The Making of Biodiversity across the Yeast Subphylum

THE UNIVERSITY of WISCONSIN MADISON

USDA

VANDERBILT UNIVERSITY

Hittinger lab

Kurtzman lab

Rokas lab

The Making of Biodiversity across the Yeast Subphylum

- Sequence the genomes of all ~1,000+ known budding yeast species
- Construct their definitive phylogeny and timetree
- Examine the impact of metabolism on yeast diversification
- Revise their taxonomy

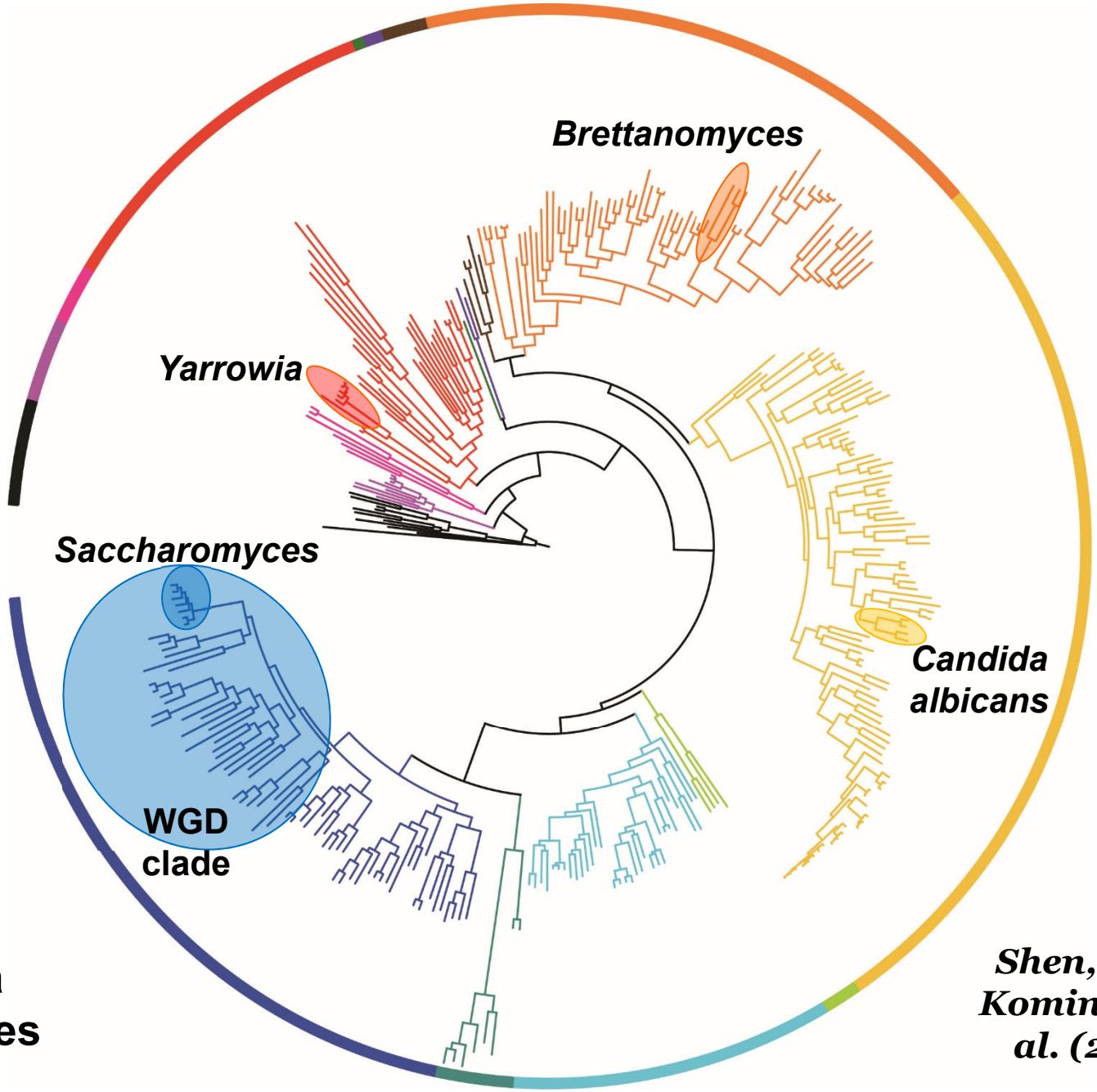
❖ **Sequenced the genomes of 220 species (196 Y1000+ species + 24 RIKEN genomes); most of them are from type strains**

❖ **+ 112 publicly available genomes -> 332 genomes**

❖ **Sampled taxa from 79 / 92 genera (~85%)**

Shen, Opulente, Kominek, Zhou et al. (2018) Cell

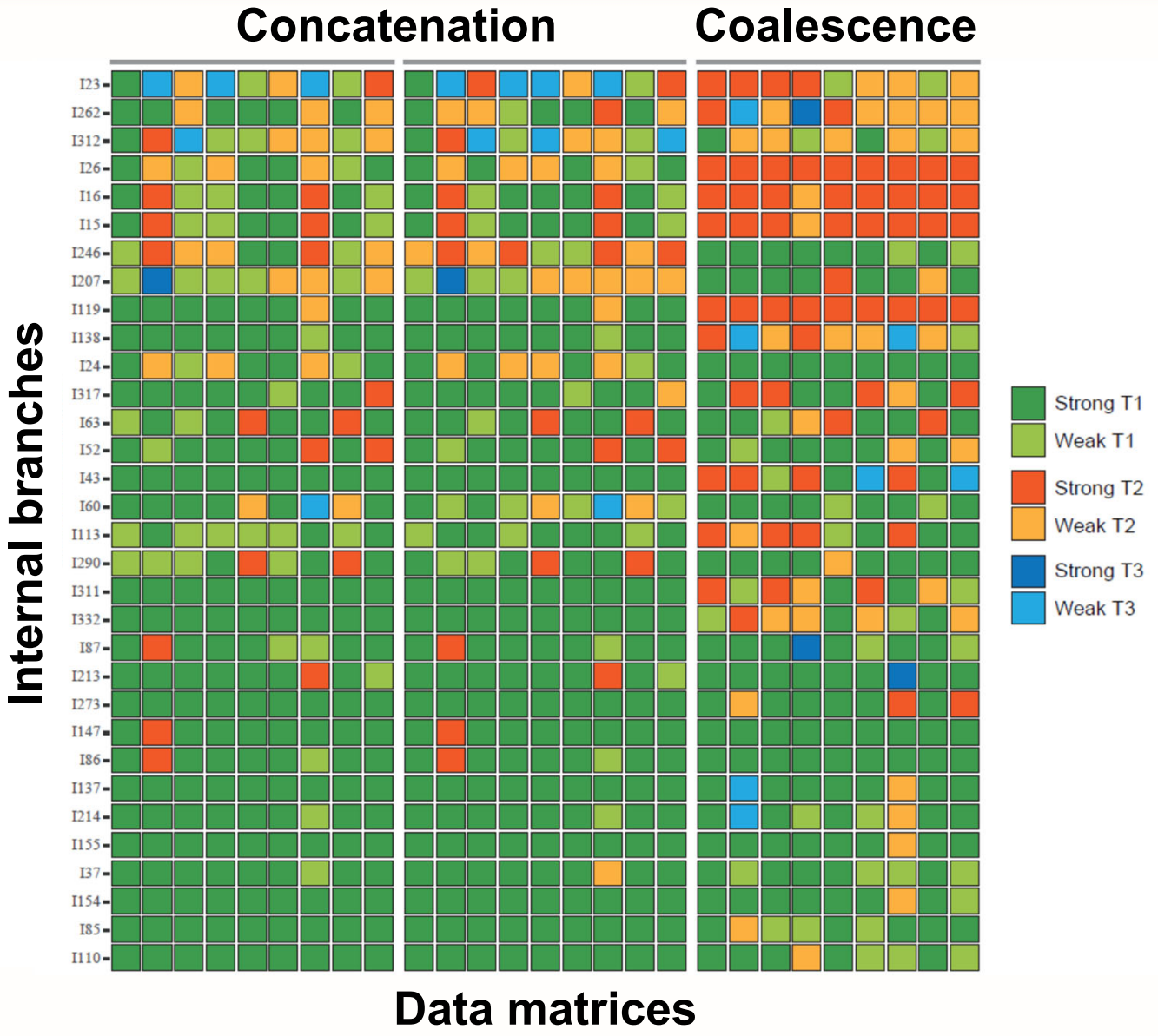
Genome-Scale Phylogeny of Budding Yeasts



332 taxa
2,408 genes

Shen, Opulente, Kominek, Zhou et al. (2018) Cell

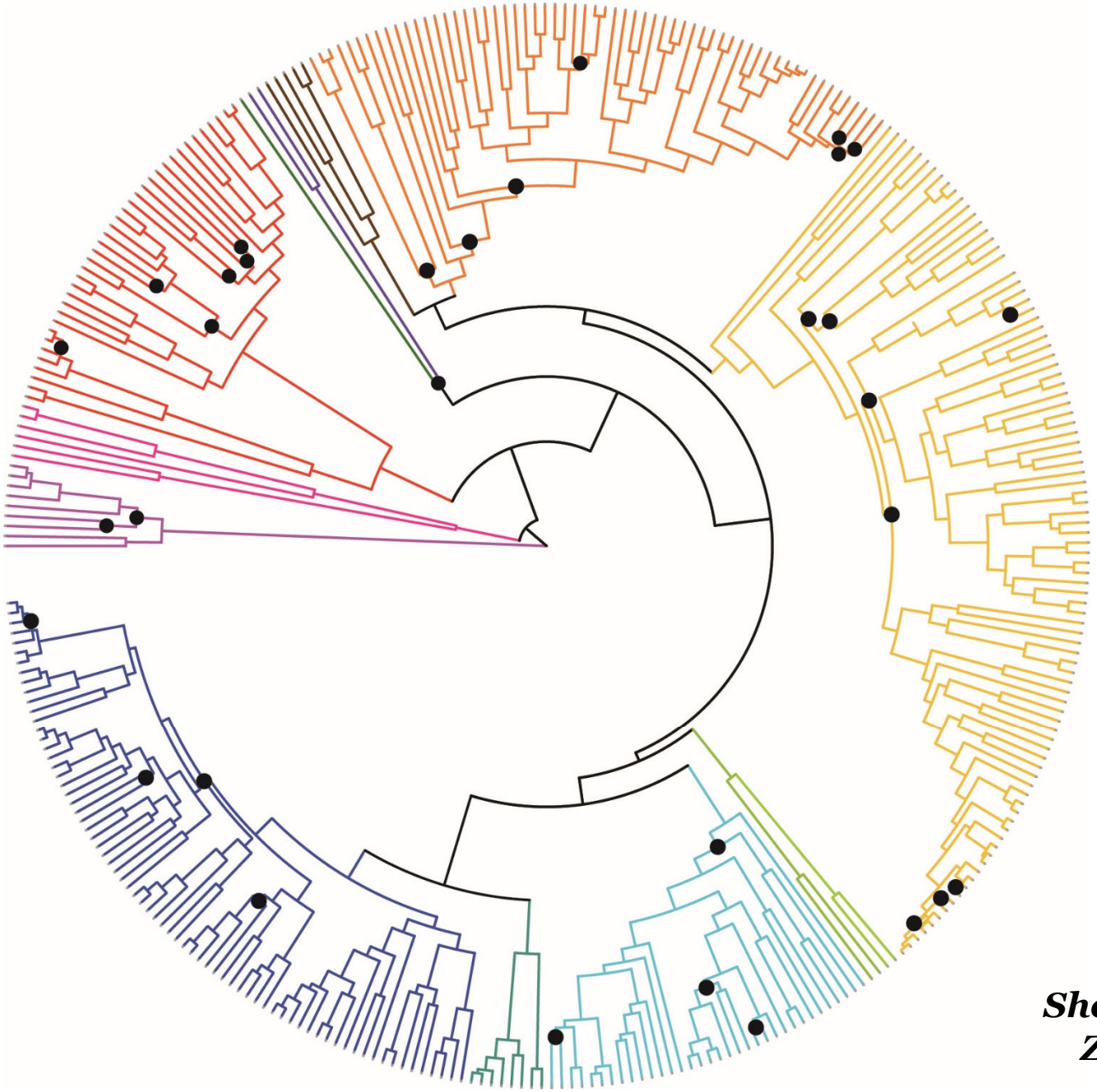
The 32 Conflicting Branches in the Yeast Phylogeny



~10% (32 / 331) of internal branches show conflict between analyses

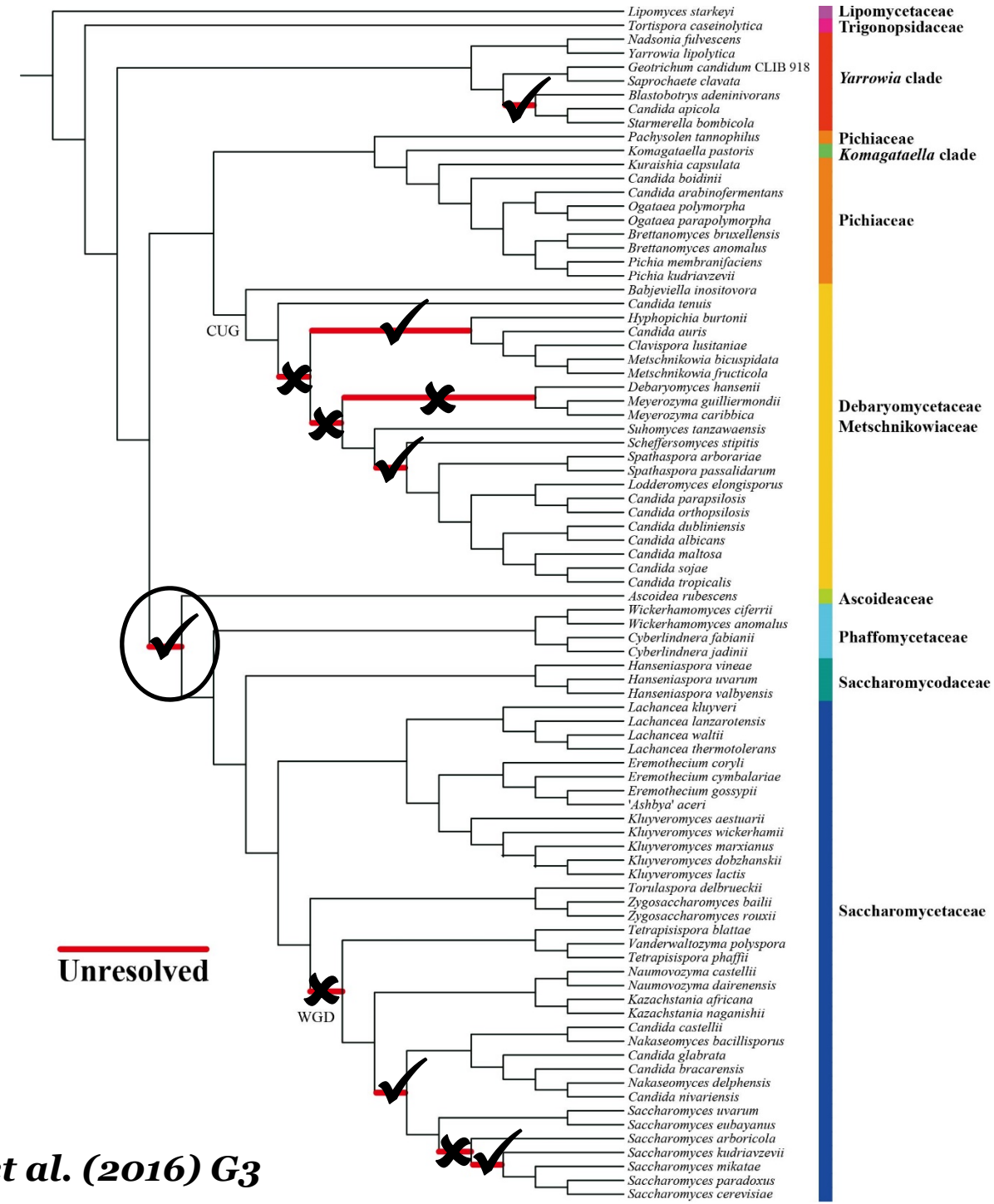
Shen, Opulente, Kominek, Zhou et al. (2018) Cell

Distribution of Conflict on the Yeast Phylogeny



- Lipomycetaceae
- Trigonopsidaceae
- Dipodascaceae / Trichomonascaceae
- Alloascoideaceae
- *Sporopachydermia* clade
- CUG-Ala clade
- Pichiaceae
- CUG-Ser1 clade
- CUG-Ser2 clade
- Phaffomycetaceae
- Saccharomycodaceae
- Saccharomycetaceae

Shen, Opulente, Kominek, Zhou et al. (2018) Cell



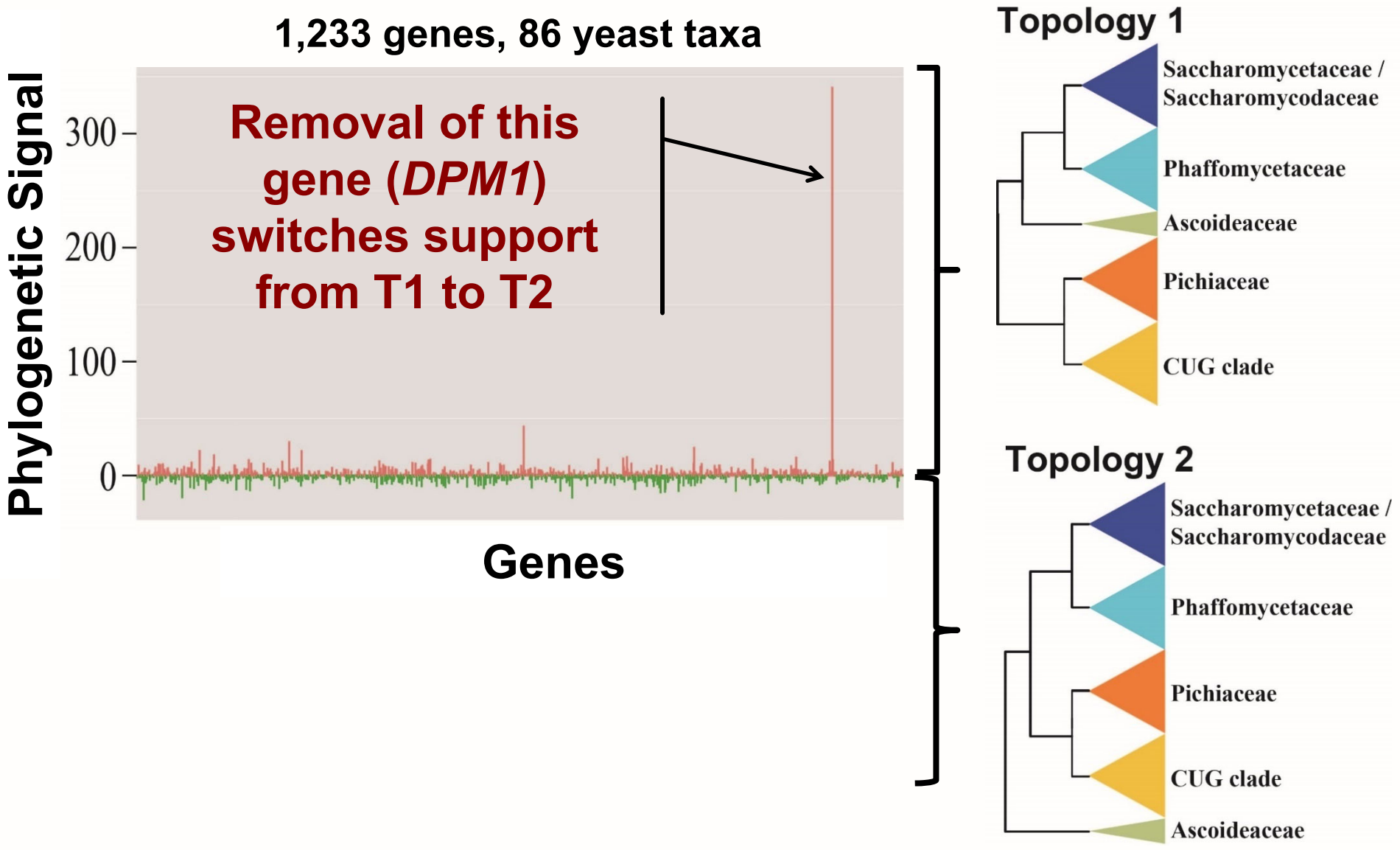
Shen et al. (2016) G3

1,233-gene, 86-taxon data matrix

~13% (11 / 85) of internal branches conflict between analyses

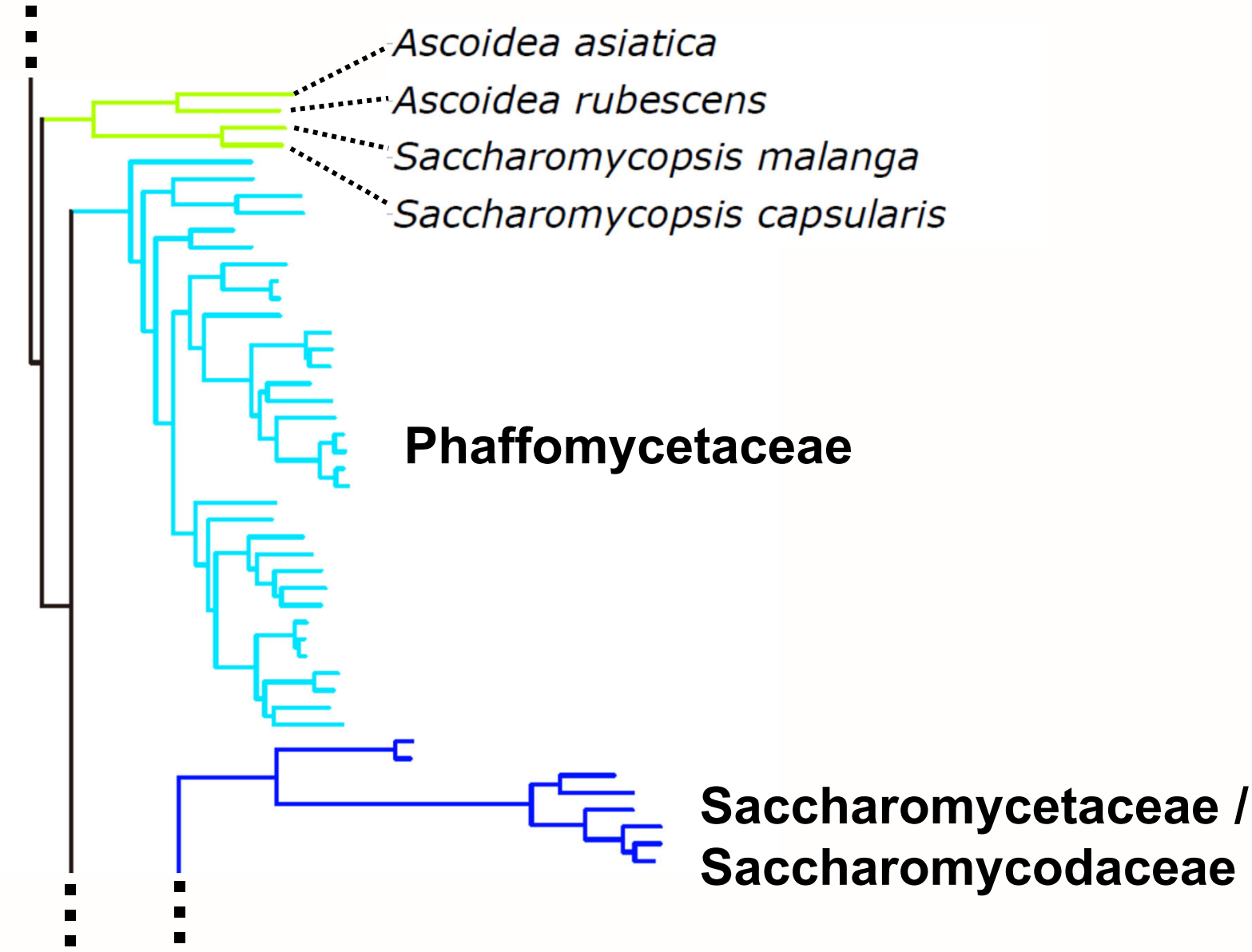
Despite increasing # internal branches ~4X, (85 -> 331), conflict decreased

A Single Gene Governs the Placement of Ascoideaceae



Shen et al. (2017) Nature Ecol. Evol.

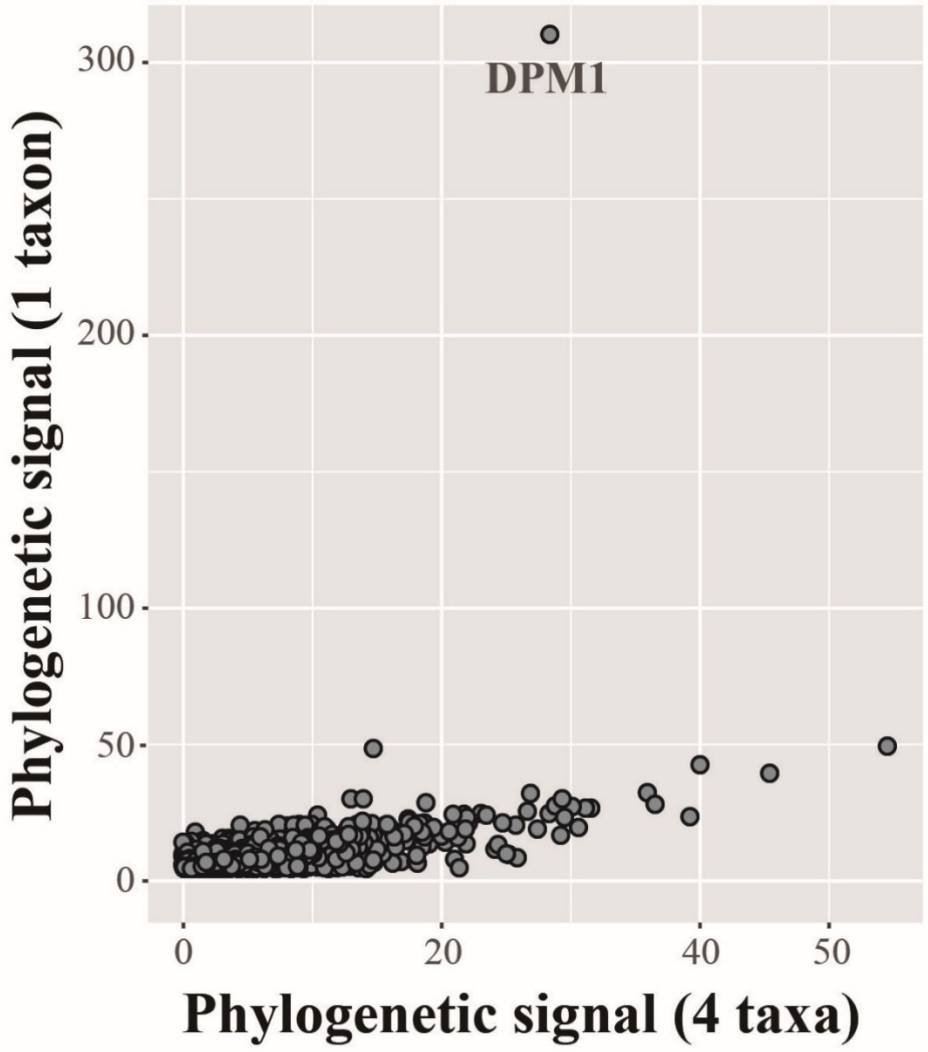
Sampling of 3 Additional Taxa "Breaks" the Long Branch



Shen, Opulente, Kominek, Zhou et al. (2018) Cell

Sampling of 3 Additional Taxa Decreases Gene's Signal

2,408 genes, 329 – 332 yeast taxa



Shen, Opulente, Kominek, Zhou et al. (2018) Cell

Genomfart?

- **Parts of the tree of life are more likely to resemble a bush rather than a tree – do we expect that we can confidently infer every branch and twig?**

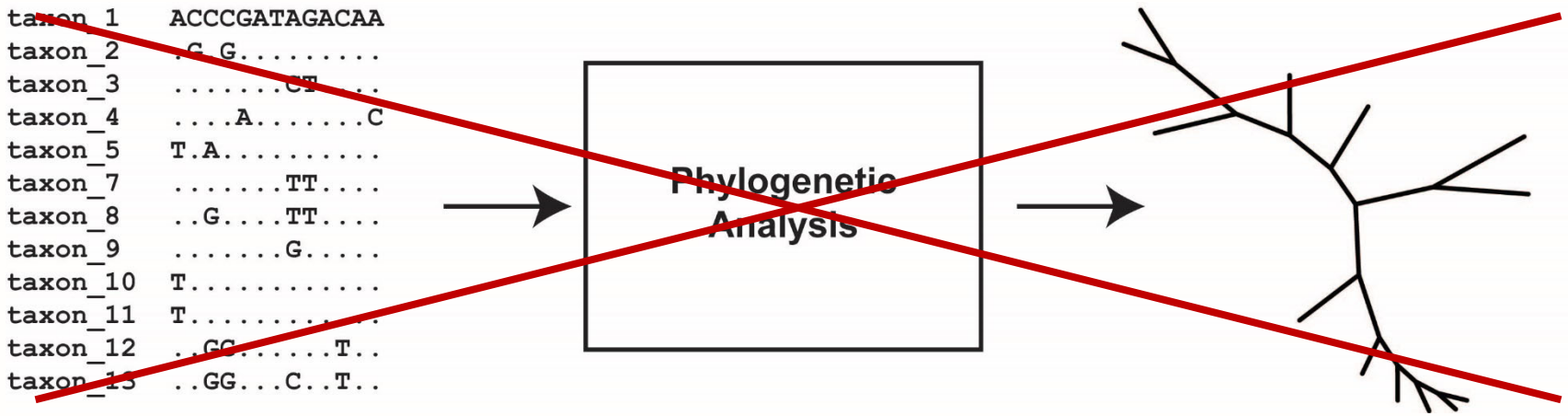
- **Bootstrap-based measures not useful in large data sets**

- **Methods evaluating conflict among data subsets (e.g., internode certainty among genes or sites or concordance factors) are preferable**

- **Explicitly identify internodes that, despite the use of genome-scale data sets, robust study designs and powerful algorithms, are poorly supported**

- **Taxon choice matters & more data will help!**

The Way Forward



Multiple sequence alignment / data matrix reconstruction

Apply different phylogenetic analyses (diff. optimality criteria / diff. approaches)

Assess conflict (e.g., use internode certainty / concordance factors)

Investigate alternative hypotheses for branches showing conflict / assess sensitivity of results

Only report resolution of branches that you have support for

Take Home Messages

“One can use the most sophisticated audio equipment to listen, for an eternity, to a recording of white noise and still not glean a useful scrap of information”

Rodrigo et al. (1994)

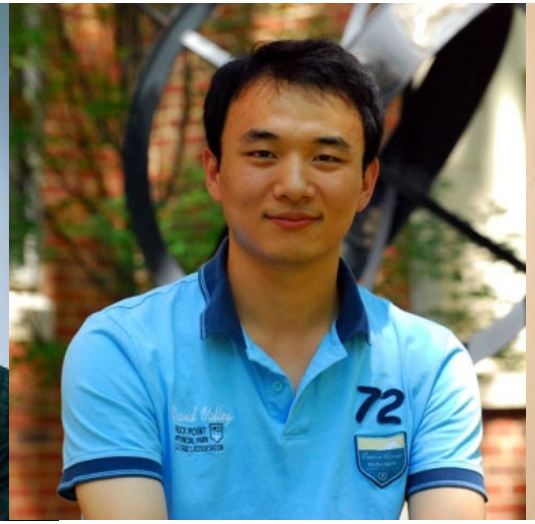
Chapter in: Sponge in Time and Space; Biology, Chemistry, Paleontology

Rokas Lab

polytomies happen...

Acknowledgements






Leonidas Salichos | **Xing-Xing Shen** | **Xiaofan Zhou** | **Alexis Stamatakis**

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