Agricultural Genomics: The Rise of the Genomes



Dr. Doreen Ware, USDA ARS

Agricultural Biotechnology: Emerging Technologies and Insights

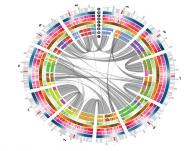
January, 27, 2021

Advancing Agriculture Through Collaborative Research on Crop & Model Species

Outline

U.S. Drought Monitor

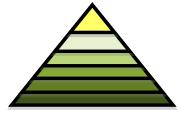
- Agricultural Drivers
- Maize genome 16 years
 - What's in a genome
 - Improvements in Sequencing technology we can Continue to evaluate approaches to develop reference assembly and annotations



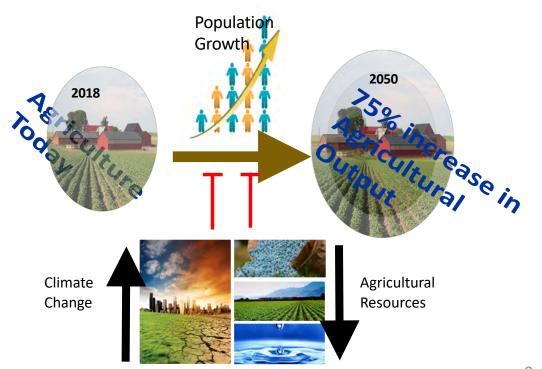
- Genome/Biology enabled agriculture
 - Sorghum EMS population
 - Forward and reverse genetics
 - Breaking down complex trait: Yeild & Quality
 - Plant architecture: flower
 - Response to environment: water, heat, nitrogen, disease
- Biology & "Big Data"
 - collaborative infrastructure
 - Future



LRR



Drivers for Agriculture: Sustainability and Defense



BREEDING FOR 2050 AND BEYOND. Prepare for plant and animal pests and disease while they are still offshore. Design plants for new environments.

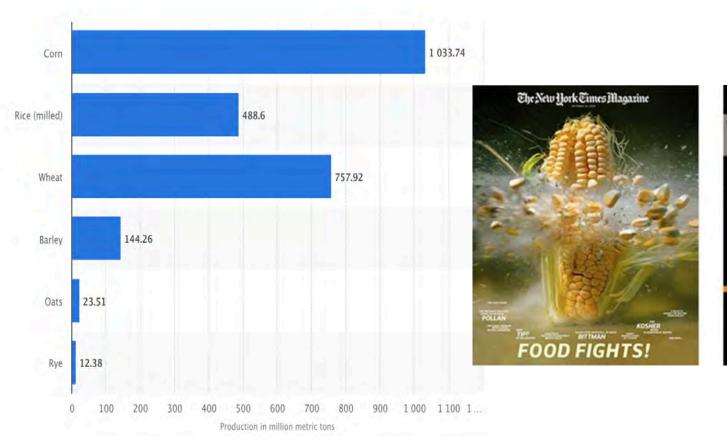
CLIMATE CHANGE. Collect and preserve natural diversity.



Nature Editorial, How to feed a hungry world. 2010

Maize is the highest world-wide production crop

Reference genomes are foundation tools for ensuring food security & environmental sustainability





Acknowledgements

Maize Diveristy Project (NSF & USDA ARS) ~ 2003- present

USD AARS **Ed Buckler**, Sherry Flint-Garcia, Mike McMullen, Jim Holland, Peter Bradbury, Doreen Ware Cornell QiSun, UCDavis Jeff Ross-Ibarra

Maize Genome Project (NSF, DOE, USDA) ~ 2005-2010

University of Washington, Rick Wilson, CSHL: Rob Martienssen, Dick McCombie, Doreen Ware, University of Arizona, Rod Wing, University of Iowa Pat Schnable

B73 Genome & Annotation Improvements V4 (NSF & USDA ARS) ~ 2015 -2017

CSHL staff: Yinping Jiao, Bo Wang, Mike Campbell, Josh Stein, Sharon Wei, Doreen Ware, Dick McCombie, Eric Antinou PacBio: David Rank, Paul Peluso, Jason Chin, Tyson Clark, Ting Hong, Elizabeth Tseng

BioNano: Alex Hestie, Tiffany Liang, Jinghua Shi. USDA ARS Mike McMullen, Kate Guill, University of Georgia: Kelly Dawe, Jonathan Gent, University of Hawaii: Gernot Presting, Kevin Schneider, Thomas Wolfgruber

Institutes: Cold Spring Harbor Laboratory, USDA ARS, Pacific BioSciences, BioNano, University of Georgia, University of Hawaii

Transcriptome Variation (USDA ARS) 2016-2019

CSHL staff: Yinping Jiao, Bo Wang, Mike Campbell, Josh Stein, Sharon Wei, Doreen Ware, Dick McCombie, Sara Goodwin PacBio: Elizabeth Tseng, Tyson Clark, Ting Hong, Kevin Eng, Primo Baybayan Institutes: Cold Spring Harbor Laboratory, USDA ARS, Pacific BioSciences

Maize Nested Association Mapping Panel Reference Assemblies (NSF & USDA ARS) 2018- present

University of Georgia: Kelly Dawe, University of Iowa: Matt Hufford University Minnesota Candy Hirsch 6SHL staff: Josh Stein, Kapeel Chougule Sharon Wei, Doreen Ware, Dick McCombie, Sara Goodwin PacBio: Emily Hatas, Paul Peluso, Jason Chin, Cortiva: Victor Llaca, Kevin Fengler, Greg May, DNA Nexus: Brent Hannigan, Chai Fungtammasan, Brittanny O'Sullivan, NIH: Adam Phillippy, Serge Koren

Institutes: Cold Spring Harbor Laboratory, USDA ARS, University of Georgia, University of Iowa, Pacific BioSciences, DNANexus, NIH

Gramene www.gramene.org (NSF & USDA ARS) 2002- present NSF IOS-1127112

PI: Doreen Ware, PI (USDA ARS, CSHL) & Pankaj Jaiswal, Co-PI (OSU), Paul Kersey (Ensembl Genomes EBI), Helen Parkinson (ATLAS EBI), Lincoln Stein (Reactome OCIR), Crispin Taylor (ASPB)

Gramene @ CSHL Andrew Olson, Joshua Stein, Sharon Wei, : Marcela Karey Monaco,

Institutes: Cold Spring Harbor Laboratory, Oregon State University, EMBL – European Bioinformatics Institute, Ontario Institute for Cancer Research, American Society for Plant Biologists

USDA ARS Sorghum Functional Genomics and Germplasm improvement: 2015- present

USDA Doreen Ware, **Zhanguo Xin**, Chad Hayes, Yinghua Huang, Gloria Burow, Ratan Chopra, John Burke SorghumBase: Nick Gladman, Yinping Jiao, Bo Wang, Kapeel Chouqule Andrew Olson Sharon Wei, : Marcela Karey Monaco Corteva,

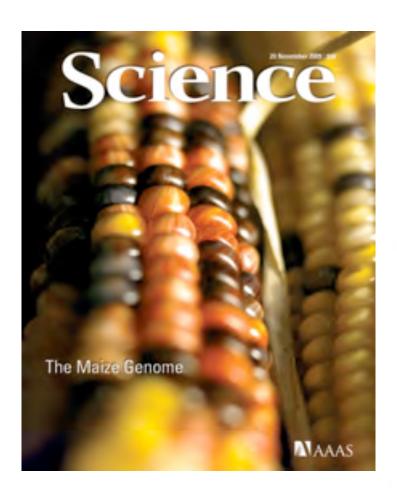


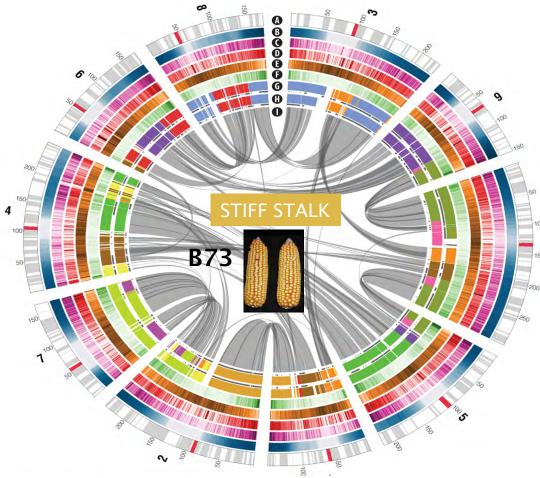






Maize Genome is over 12 years old





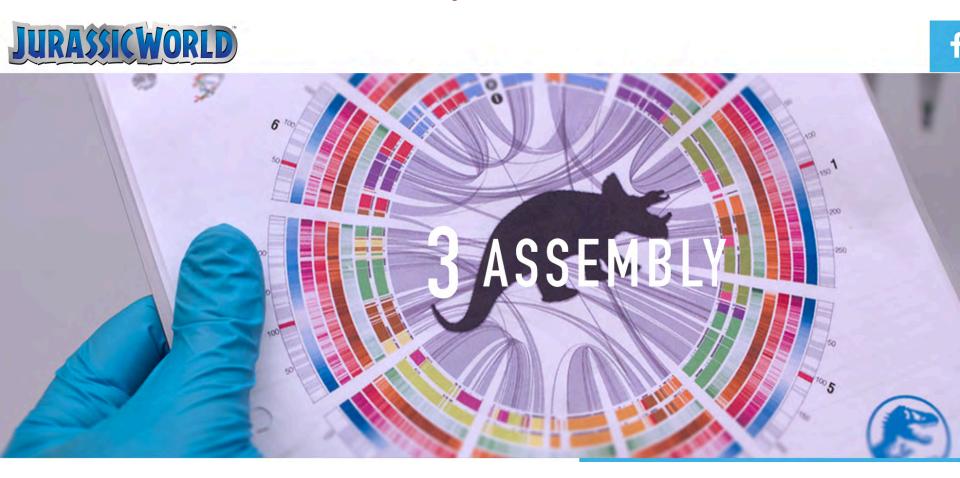






Sequence genomes provides us the parts list and allows us to see what is the same or different between organisms Recombination rate (cM/Mb) Mu insertions (sites/Mb) MF Enrichment (% reads) ■ Repeat coverage (%) Gene density (genes/Mb) Genes in corn, rice, and sorghum are in similar places in the genome 150 Schnable, Ware et al. Science (2009)

Triceratops Genome



Genomes sequences allow us to see all the variations (mutations) that exist in nature

Letters or Single nucleotides polymorphisms (SNPs)

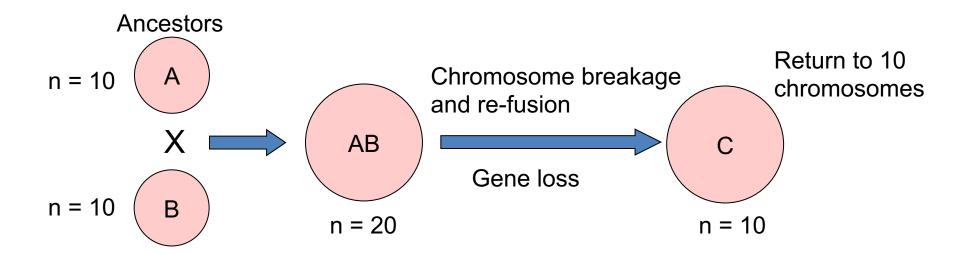
Gene content or parts list, known as copy number variations (CNVs)

Jumping genes, Transposable Elements (TEs) associated with the regulatory sequence between the Genes

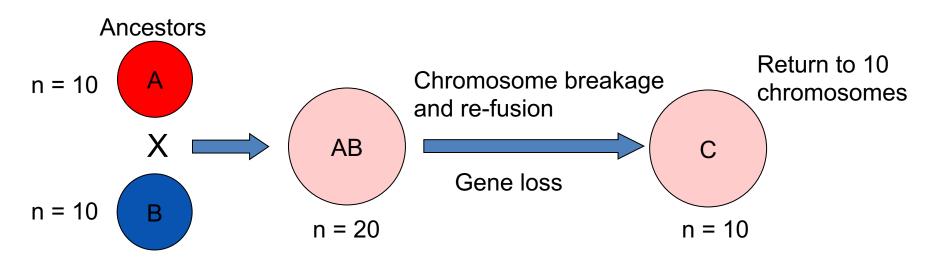


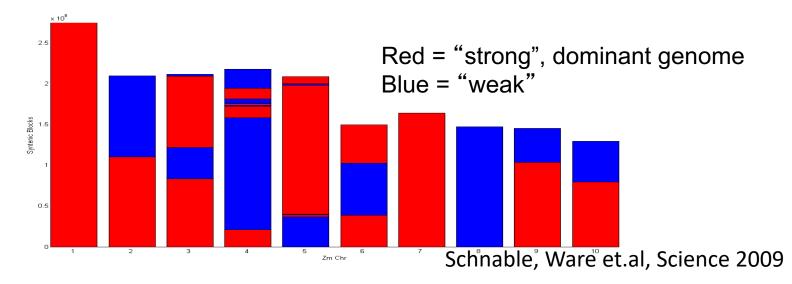
Barbara McClintock
1983 Nobel Prize in Physiology & Medicine

Maize is a "Tale of Two Genomes"



Evolutionary History of the Maize Genome





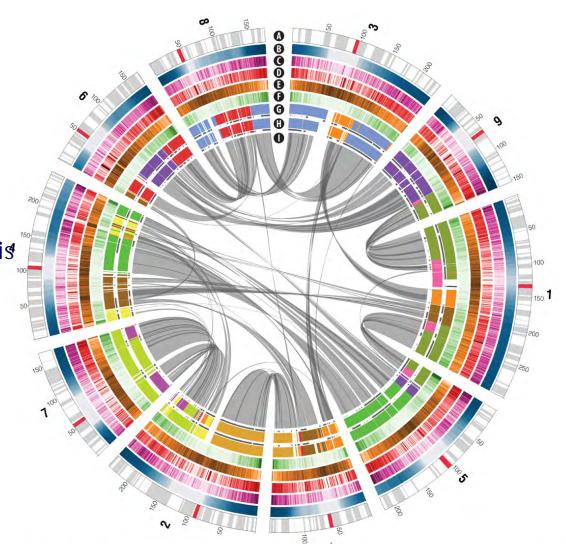
Maize is a "Tale of Two Genomes"

Maize, also known as corn experienced a whole genome duplications and then lost many of the genes

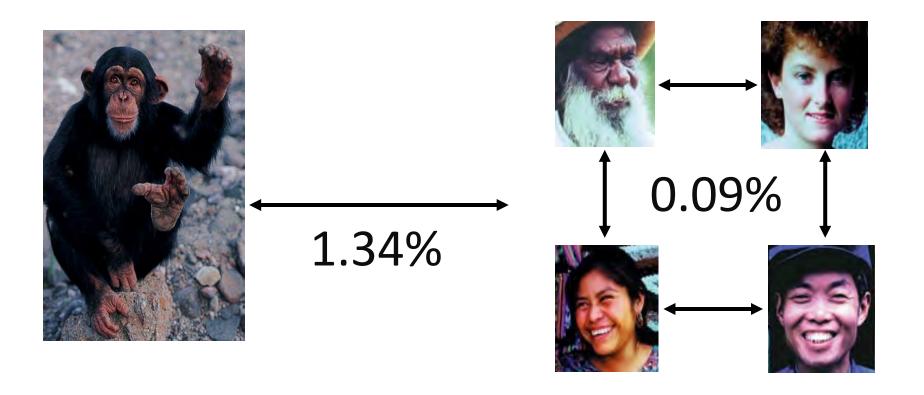
The genes that were kept by corn can tell us about how corn is adapting

Transcription factors, kinases, chromatin modifiers

Not all genomes have the same potential



Humans Have Limited Molecular Diversity



Maize diversity is greater than the difference between human and chimps



Individual Maize lines are very different from each other

The SNPs and gene differences affect how corn plants grow

Access to these sequences can accelerate the time it takes to make new lines of corn

Differences come from locally duplicated genes



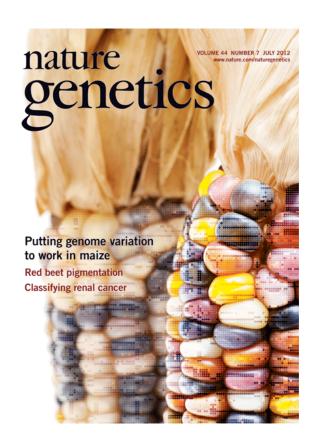




An additional copy of gene confers tolerance to acidic soil



Maize genomes are highly variable



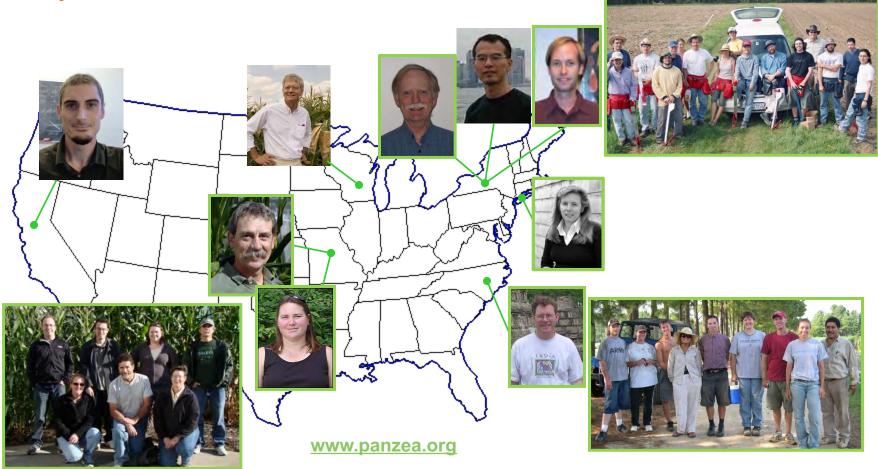
- High rate of SNP and structure variation in the population
- Structure variations are highly associated with phenotypic variation
- Structural variation in non coding region was enriched for phenotypic variation
- One genome is not enough to represent the diversity of the population

Chia JM, Song C, Bradbury PJ, Costich D, de Leon N, Doebley J, Elshire RJ, Gaut B, Geller L, Glaubitz JC *et al*: **Maize HapMap2 identifies extant variation from a genome in flux**. *Nat Genet* 2012, **44**(7):803-807.

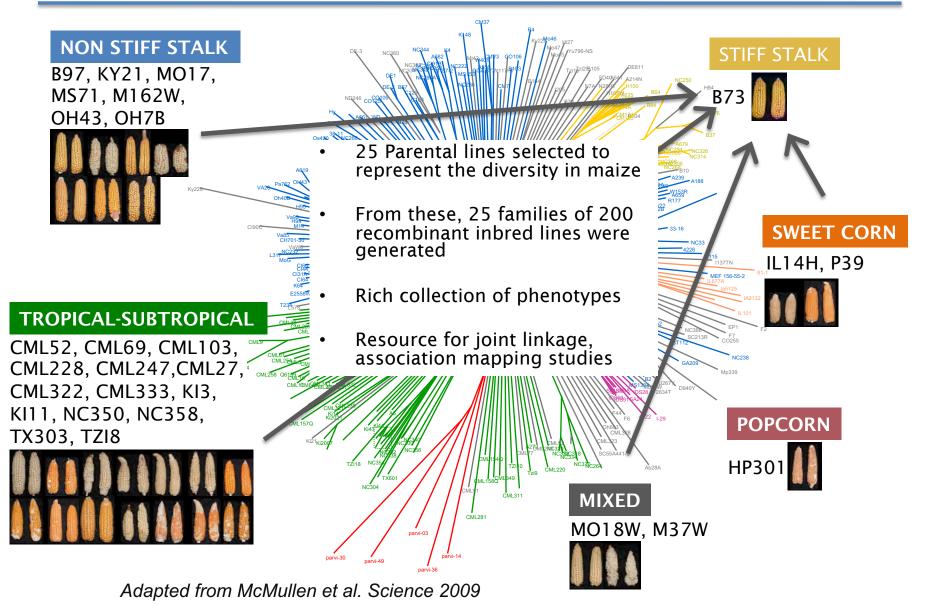
MAIZE DIVERSITY PROJECT



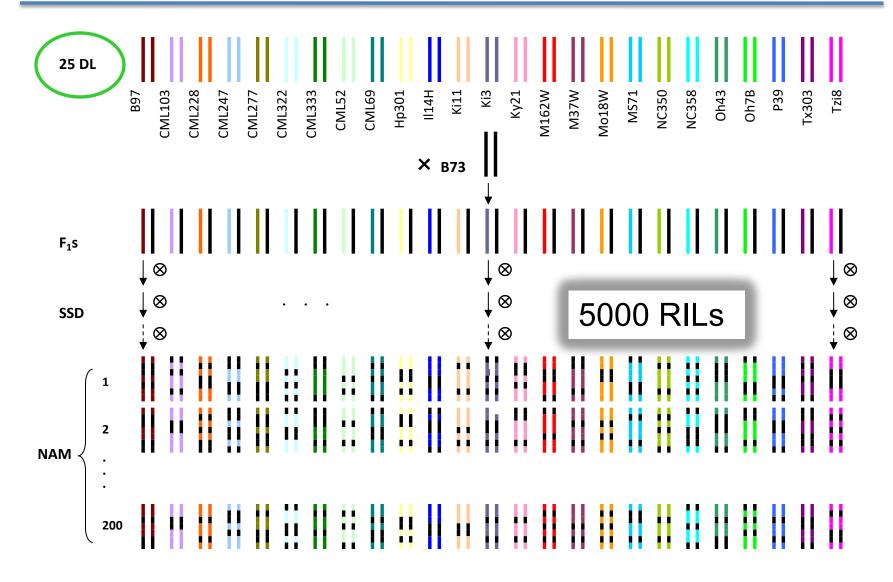
Meet the Family (2002 - present)



26 Reference Assemblies for the Maize Nested Association Mapping (NAM) Population



Maize Nested Association Mapping (NAM) Population



26 Maize NAM founders reference assemblies (2019- 2021)

Arun Seetharam -ISU Margaret Woodhouse - MaizeGDB Kapeel Chougule -CSHL Shujun Ou -ISU Jianing Liu -UGA Xuehong Wei -CSHL Zhenyuan Lu -CSHL Andrew Olson -CSHL Bo Wang -CSHL Sharon Wei -CSHL TingTing Guo -ISU Rafael Della Coletta -UM Xianran Li -ISU John Portwood –MaizeGDB Kevin Fengler -Corteva Victor Llaca -Corteva Amanda Gilbert -UM Nancy Manchanda -ISU Samantha Snodgrass -ISU David Hufnagel -ISU Sarah Pedersen -ISU Michael Syring -ISU Ethy Cannon - MaizeGDB Carson Andorf - MaizeGDB Jonathan Gent –UGA Todd Michael - JCVI Jianming Yu -ISU Candice Hirsch -UM Doreen Ware -CSHL Matthew B. Hufford -ISU

R. Kelly Dawe -UGA



U. Georgia

Kelly Dawe Doreen Ware



Iowa State U.





Candy Hirsch U. Minnesota







USDA-ARS

CSHL























































Bo Ward







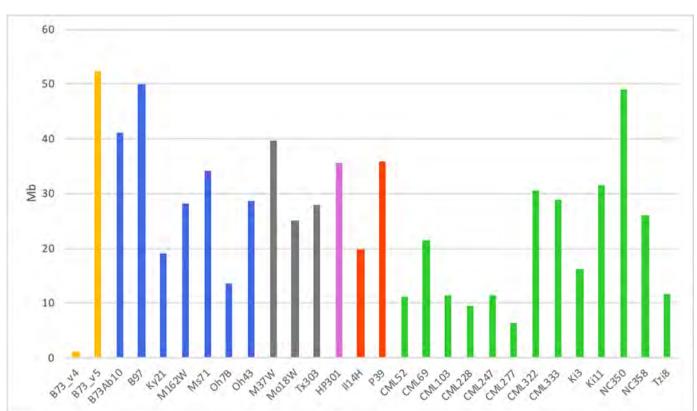


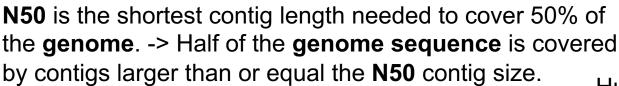






New assemblies have a vast improvement in the contiguity of the sequence









Kevin Fengler

Victor Llaca



Matrices	Assembled Contigs
Total Bases in Assembly	2,180,413,054
Contig Contiguity (NG50)	52,409,415
Number of Contigs	811
Longest Contig	161,290,055

StiffStalk

NonStiffStalk

Mixed

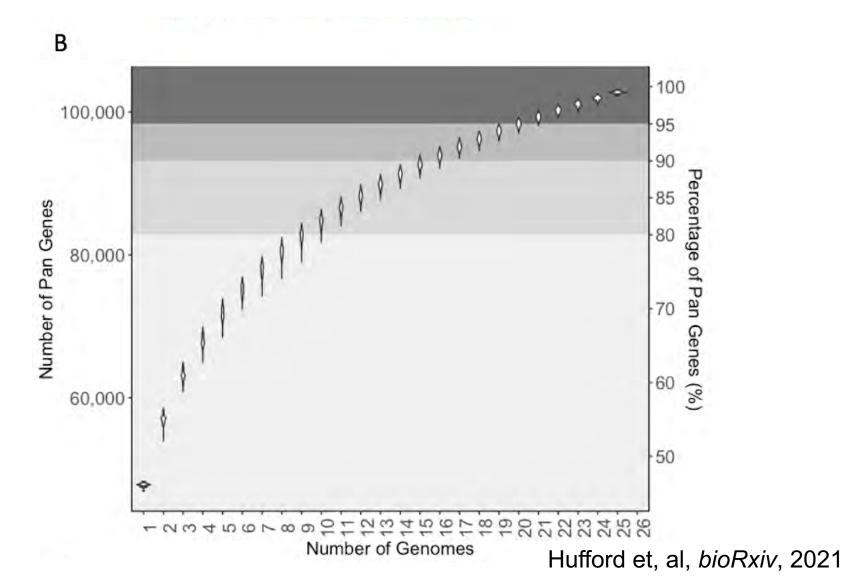
PopCorn

SweetCorn

Tropical

Hufford et, al, bioRxiv, 2021

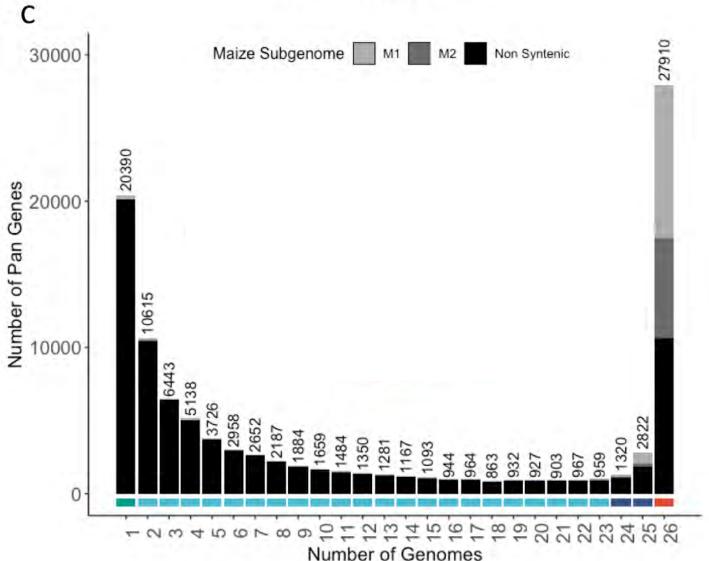
More than 100, 000 genes found in the 26 maize accession



Maize Pan Gene Set



Candy Hirsch U. Minnesota



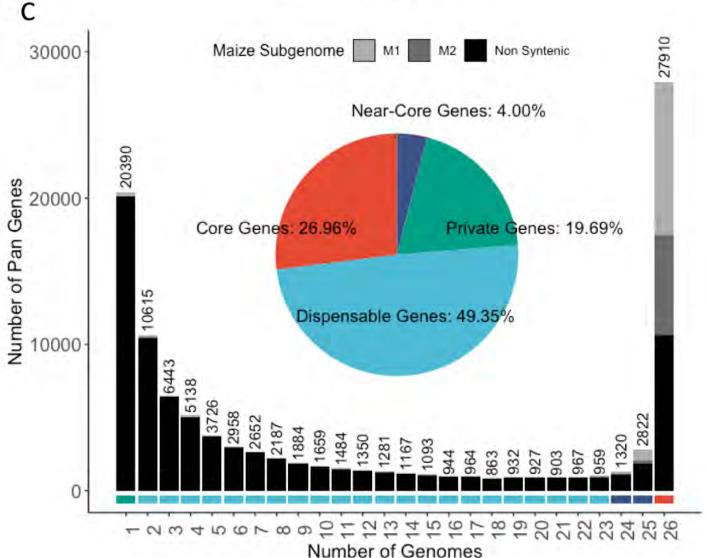
Hufford et, al, bioRxiv, 2021

Maize Pan Gene Set

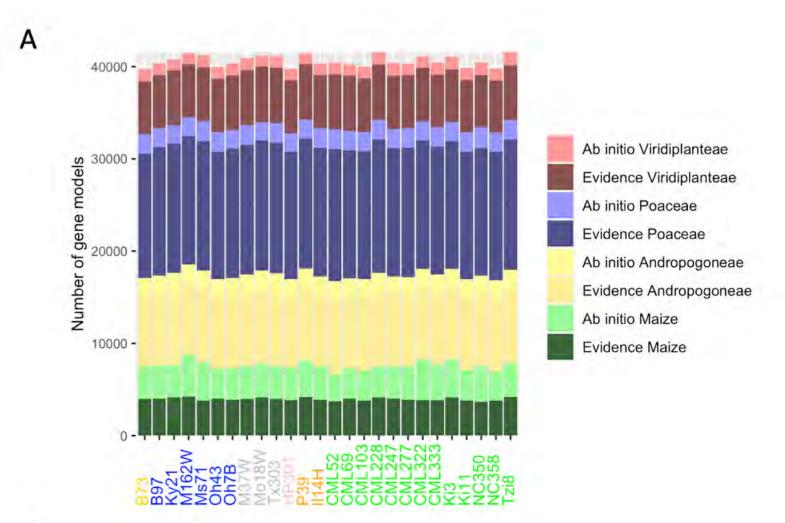
Core genes are more likely to be Syntenic



Candy Hirsch U. Minnesota



The bulk of the genes in maize are found in other species



Genome contains life history of the species

Teosinte Zea mays L. ssp. parviglumis

Landraces



Biology Enabled Agriculture

G X (environment +management) = P

Complex Traits: Yield & Quality

Phenomes



Genome Editing
Genomic Selection
Marker Assisted Breeding

Gene Networks

Metabolomics Expression Epigenetic

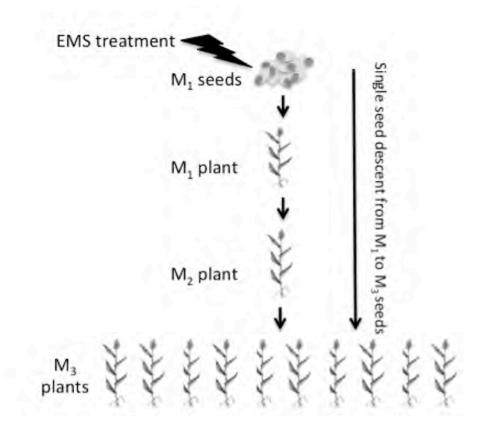
Regulatory Networks Metabolic Pathways

Genomes





Profiling new sorghum genetic & phenotypic variation





Zhanguo Xin
Cropping Systems Research Lab,
USDA-ARS, Lubbock TX

- Parental line: BTX 623
- >10,000 individual M2 seed pools
- >6,400 M3 seeds obtained and
- Phenotyping is on-going and need to be expanded
- High quality DNA prepared for all lines

EMS Mutagenesis

- -Random
- Single nucleotide change
- >99% GC→AT



Mutation Detection by whole genome sequencing of 256 mutants for forward genetics



Yinping Jiao

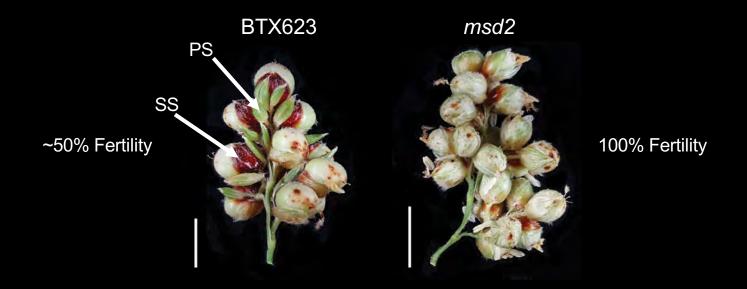
Sequencing summary

- 20 M3 plants pooled together for sequence to averagely 16X
- Average whole genome coverage 86%; gene space coverage -95%
- Quality control of the population: 2 contamination lines + 2 sibling lines
- Mutation detection:
 - 1,862,560 EMS-induced mutations
 - Sanger sequencing validation rate >98%
 - 7,660 mutation/mutant = 1,798 homozygous + 5,862 heterozygous

Mutation Effect:

- 22% of mutations are located in genes, covering 95% of Sorghum genes
- 57% (18,684) of the genes harbor >35,000 disruptive mutations, ~2 disruptive mutations per gene.

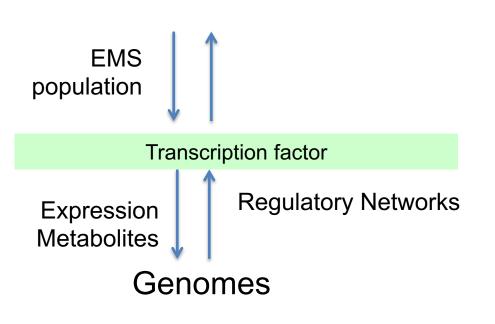
Multiseeded (msd1, msd2, msd3)



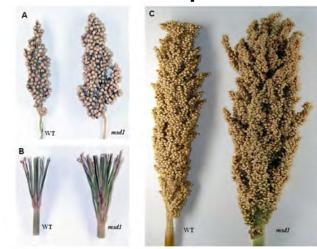
Biology Enabled Agriculture

G X (environment +management) = P

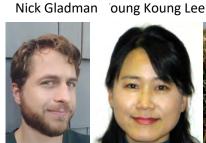
Yield > Flower development > grain number > fertility/branching















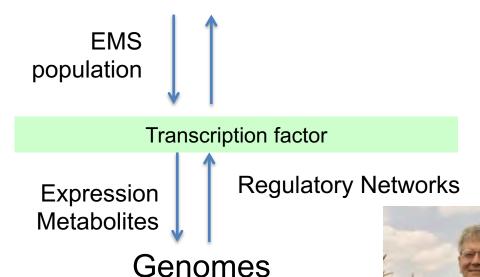
Jiao et al., Nature Comm. (2018)

Biology Enabled Agriculture

G X (environment +management) = P



Yield > Flower development > grain number > fertility/branching



TCP transcription factor



Teosinte Branched

Doebley et al., Geneitics (1995)

News



News

News From the Field

For the News Media

Special Reports

Research Overviews

NSF-Wide Investments

Speeches & Lectures

Multimedia Gallery

NSF Current Newsletter

News Archive

Secrets of Plant Genomes Revealed!

Secrets of Plant Genomes: Revealed! takes viewers on a lively, upbeat journey that explores how these plants got to be the way they are and investigates how we can make better use of them in the future. Plant scientists are hard at work--in the lab, in the field and at the computer--to increase our understanding of nature.

To download each of the parts, click the corresponding download button on the first player.

Introduction



Corn: The Dynamic Genome



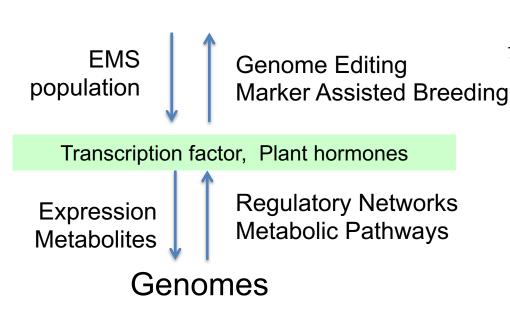
Cotton: Building a Better Plant

Biology Enabled Agriculture

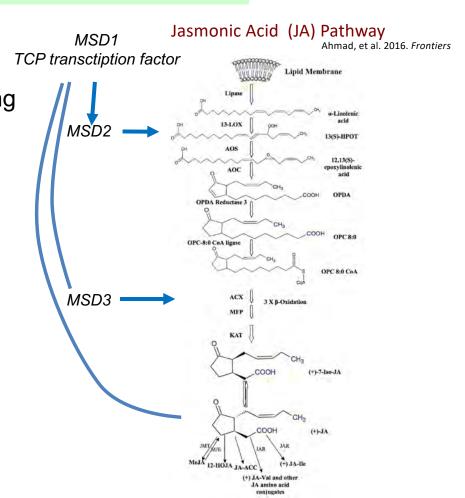
G X (environment +management) = P



Yield > Flower development > grain number > fertility/branching



Jiao et al., Nature Comm. (2018) Gladman et al., Int. J. Mol. Science (2019) Dampanaboina, L., Int. J. Mol. Science (2019)



Nitrogen, soil, and agricultural sustainability

Insufficient Nitrogen fertilizer



Nitrogen deficiencies, LSU (courtesy, Dr. Brenda Tubana)

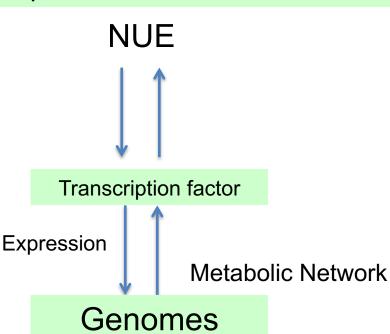
Excess Nitrogen fertilizer



October 2011. Credit, USGS, NASA.

G X (environment +management) = P

Complex Traits: Yield> Fitness> Limiting Nitrogen



Lifang Zhang

Andrew Olson

Christophe Liseron-Monfils



Allie Gaudinier **UC DAVIS**



Siobhan Brady



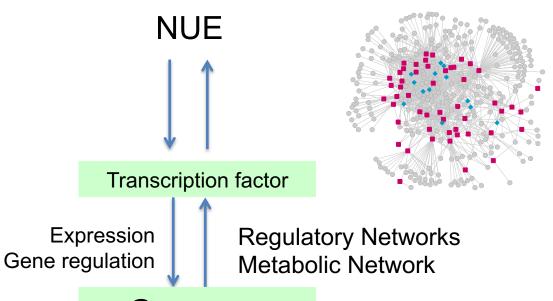




Guardinier et al, Nature 2017

G X (environment +management) = P

Complex Traits: Yield> Fitness> Limiting Nitrogen



Genomes

Lifang Zhang

Andrew Olson



Christophe Liseron-Monfils



Allie Gaudinier **UC DAVIS**



Siobhan Brady



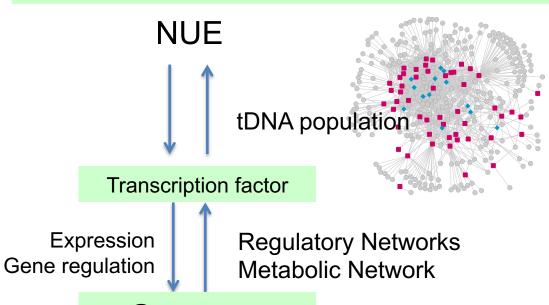




Guardinier et al, Nature 2017

G X (environment +management) = P

Complex Traits: Yield> Fitness> Limiting Nitrogen







Andrew Olson



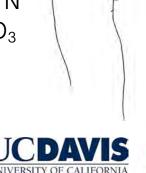
Christophe Liseron-Monfils



Allie Gaudinier **UC DAVIS**



Sufficient N 10mM NO₃

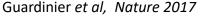


Limiting N 1mM NO₃



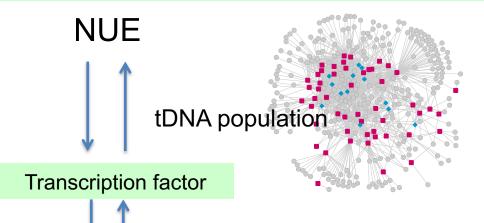






G X (environment +management) = P

Complex Traits: Yield> Fitness> Limiting Nitrogen



Expression Regulatory Networks Gene regulation Metabolic Network

Genomes





Andrew Olson



Christophe Liseron-Monfils

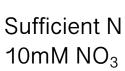


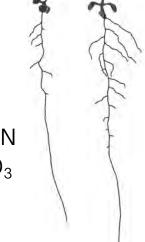
UC DAVIS



Allie Gaudinier





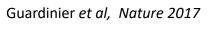


Limiting N 1mM NO₃

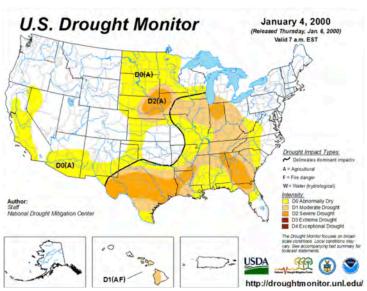


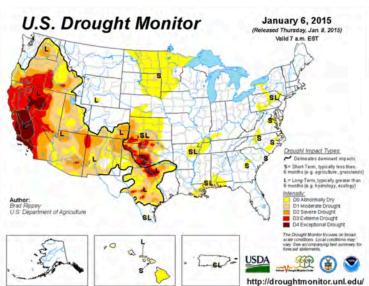


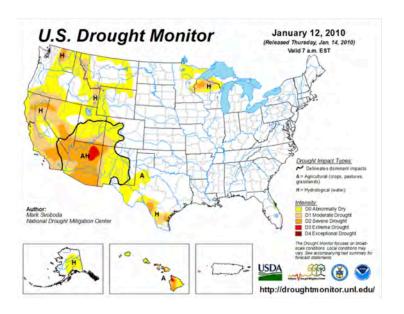


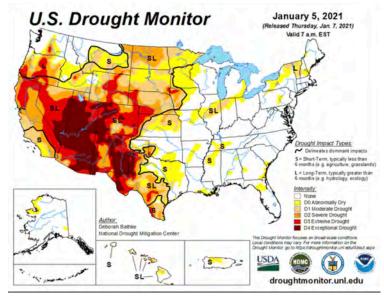


Changing climate increasing temperature and drought





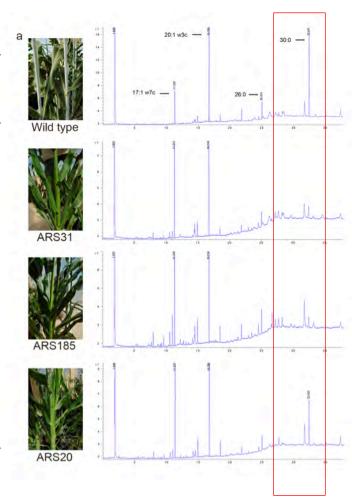




Reverse Genetics: From Gene to Phenotype

Epicuticular wax (bloom) of sorghum plays important roles in tolerance of environmental stress.

Gene in Arabidopsis	Sorghum gene	Amino acid change	Mutant Id
CER6	Sobic.001G453200	E159K	ARS20
KCS12	Sobic.004G341300	R189C	ARS73
CER5	Sobic.009G083300	P581L	ARS73
CER5	Sobic.009G083300	L244F	ARS20
CER6	Sobic.006G020600	A133T	ARS205
KCS7	Sobic.002G268300	P449S	ARS31
KCS4	Sobic.002G268500	A49V	ARS31
KCS20	Sobic.005G168700	R303Q	ARS185
CER1	Sobic.001G222700	L100F	ARS185



Jiao et al. The Plant Cell, 2016

G X (environment +management) = P

Complex Traits: Yeild > Stress > Heat tolerance

Heat tolerance

Fitness impact

EMS population

ATP dependent protease

Regulatory Networks

Genomes





Zhanguo Xin

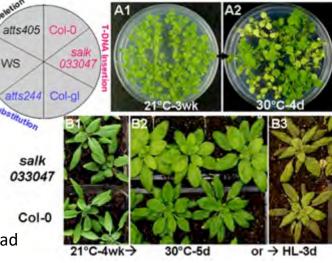


USDA-ARS, Lubbock TX Zhanguo Xin, Gloria Burow, Ratan Chopra, John Burke, Chad Hayes

WS

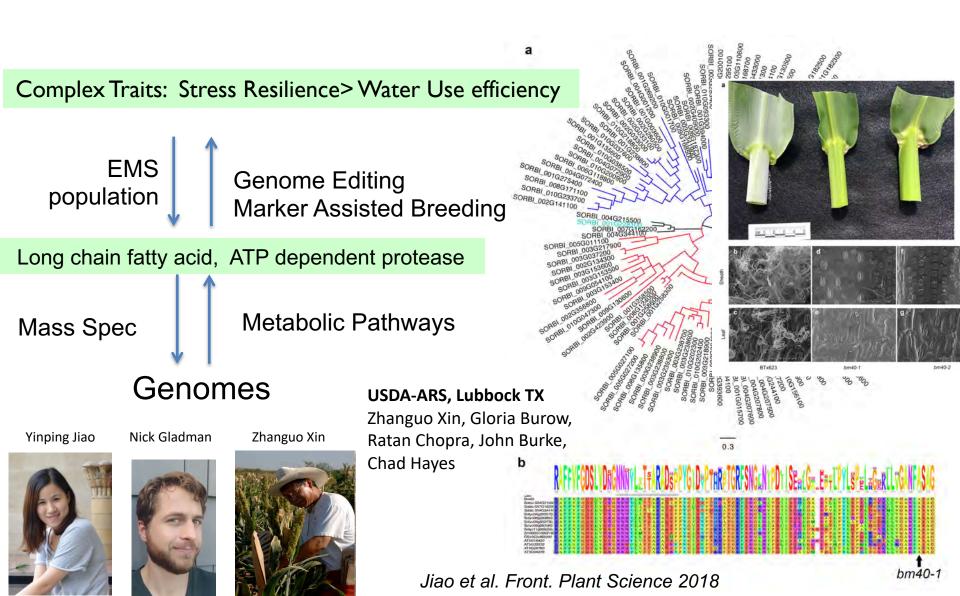
Jiao et al. Plant Cell 2016





Ftsh11 identified in a model plant

G X (environment +management) = P

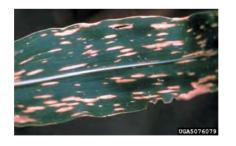


Climate change impacts disease pressures

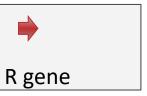




Sorghum Leaf spot http://texassorghum.org/wp-content/uploads/2015/10/Fig.-2.jpg



Resistance to Southern Leaf Blight Kump et al. Nat Genet 2011



- Yield loss: Pesticide spraying increases direct cost and impacts to the environment, and human health
- Disease resistance "R"genes (NLR) are rapidly evolving and often seen in cluster. Good candidate for structural variation
- Pan Genomes: High quality reference assemblies to support characterization of core and dispensible (adaptive) genes

Sugarcane aphids

 Since 2013 in the US sugarcane aphids have been causing enormous damage to sorghum crop

Tx2783 has high resistance to SCA



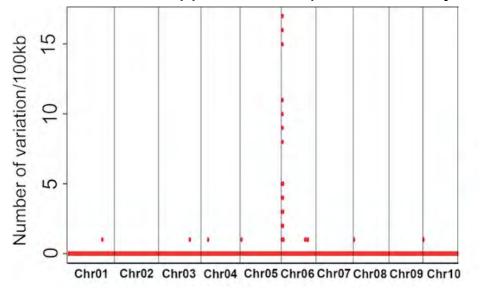




Collaborators:Yinghua Huang (USDA-ARS), Zhanguo Xin (USDA-ARS) Chad Hayes (USDA-ARS)



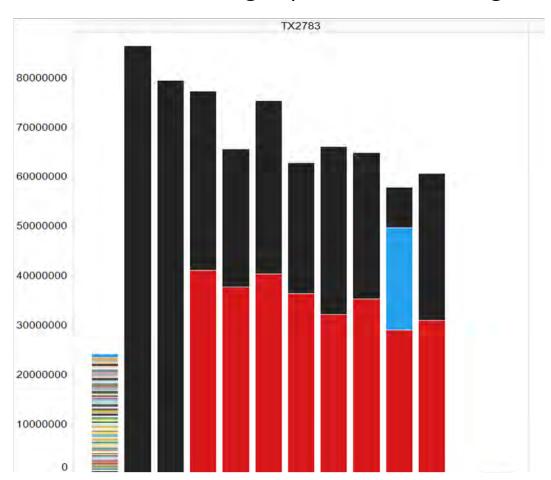
SCA locus mapped to the top of chromosojme 6



Wang et al., bioRxiv, 2021

Sorghum sugarcane aphid tolerant TX2783 reference assembly

447 PacBio contigs (25.6 Mb contig N50)



Sorghum Pan Genomes site: TX2783, BTX623, TX436, TX430, RIO Assembly Gene Neighborhood conservation view

Search Gramene



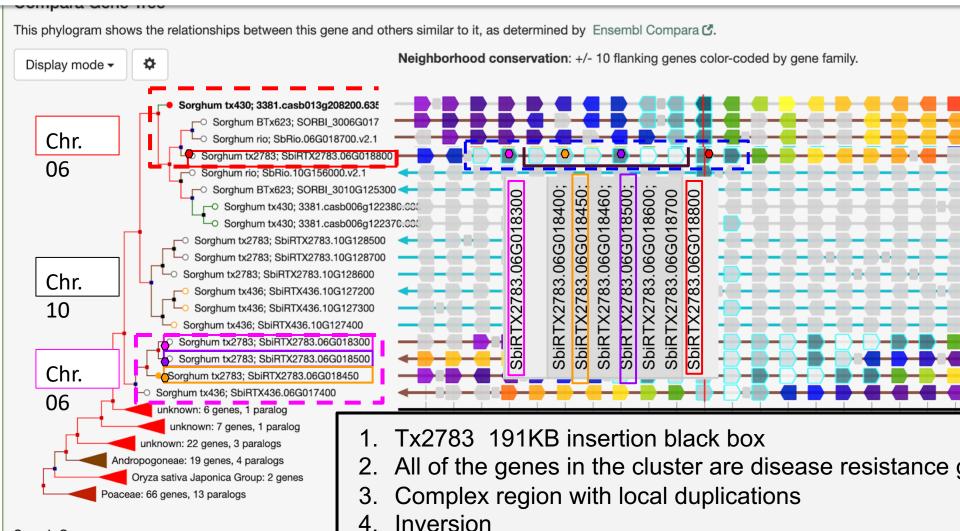




Andrew Olson

Sharon Wei

Kapeel Chou



G X (environment +management) = P

Yield> Biotic Stress > Disease Resistance



Genome Editing
Marker Assisted Breeding

NLR R Genes

Expression

Signaling pathway

Genomes

Yinping Jiao



Zhanguo Xin



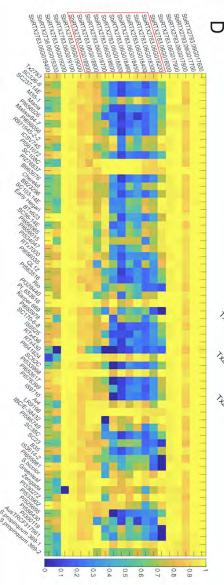
Bo Wang

Liya Wang



TX2783 191 kb structural variant (SV) containing a cluster of R Genes

Resequencing data from 62 accessions identified the SV is segregating at a low frequency in these lines.



Wang et al., bioRxiv, 2021

Exploring Genomes with an eye toward breeding

Biology Enabled Agriculture



QTL **GWAS** Gene Networks Metabolomics Expression Epigenetic Genomes

Genome Editing Genomic Selection Marker Assisted Breeding

Metabolic Pathways **Protein Complexes** Regulatory Networks

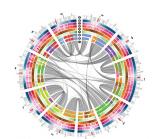
Metabolites **Proteins Transcripts** Chromatin

Association Analyses Genes with major affect

Regulatory and Inference Networks Many genes with minor contributions associated with a complex trait

Comparative analysis Transfer information across species

Improved understanding of biological mechanisms & systems will improve breeding models and support for genetic engineering



Decreasing cost of sequencing leads to increasing computes and data management



\$50 million (2009) Sequencing Centers

BAC library, Sanger sequencing library, finishing libraries, computes

\$250-180 thousand (2016) Sequencing Centers

PacBio long single molecule, Optical map, illumina short read High quality DNA, Library prep, access sequencer & ***compute

\$90- 50 thousand (2017) Sequencing Centers

PacBio long single molecule, Optical map, 10X illumina short read High quality DNA, Library prep, access sequencer & ***compute

\$45- 25 thousand (2018) Local/ Sequencing Centers

\$2-6 thousand (2021) Long single molecule



Decrease Improve Increase Sequence Assembly Compute Cost Quality Cost

Schnable, Ware et al, Science 2009
Jiao et al., Nature, 2017
Ou et. al, Genome Biology 2020
Liu et al, Nature Comm 2020
Wang et. al, submitted 2021
Hufford et. al, submitted 2021







Biology has transitioned to an Information Science

Big Data" Biology Pyramid Quantitative Biology Technologies

Results
Domain
Knowledge

Machine Learning classification, modeling, visualization & data Integration

Scalable Algorithms
Streaming, Sampling, Indexing, Parallel

Compute Systems
CPU, GPU, Distributed, Clouds, Workflows

IO Systems
Hardrives, Networking, Databases, Compression, LIMS

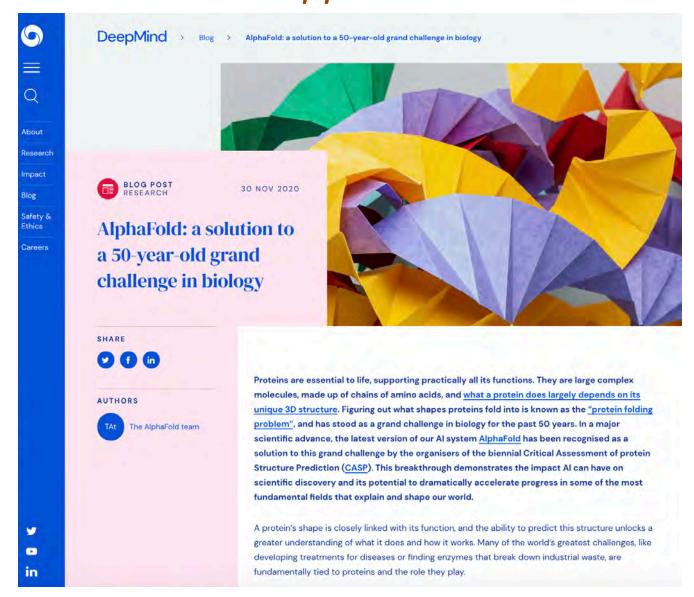
Sensors & Metadata
Sequencers, Microscopy, Imaging, Mass spec, Metadata & Ontologies

1 7

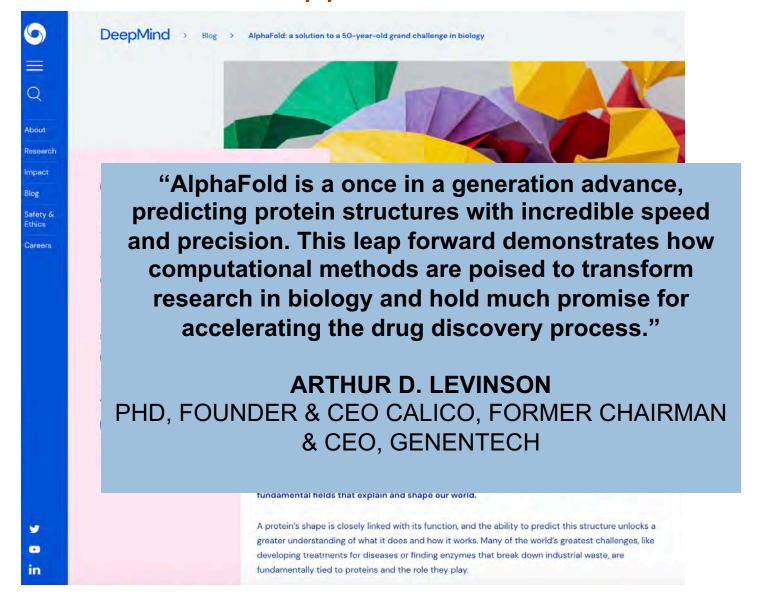
Keystone Big Data in Biology 2014 Stein, Schatz, Ware



Artificial Inteligence (AI) is allowing us to reimagine how we approach science



Artificial Inteligence (AI) is allowing us to reimagine how we approach science



Wild Grapes have natural resistance to disease



Wild Grapes come with baggage they have shed with domestication



Unpleasant taste

This, (2006); Feechan, et.al (2015)



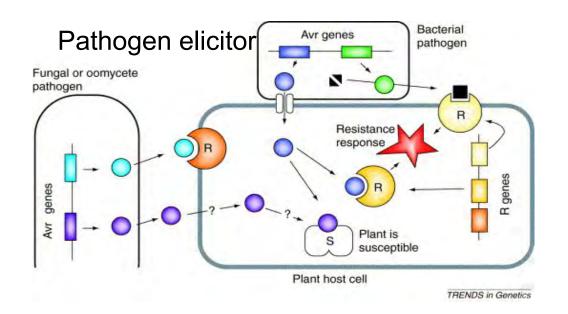
Wild: Male and Female



Domesticated: Hermaphrodite

Plant NLR R-genes

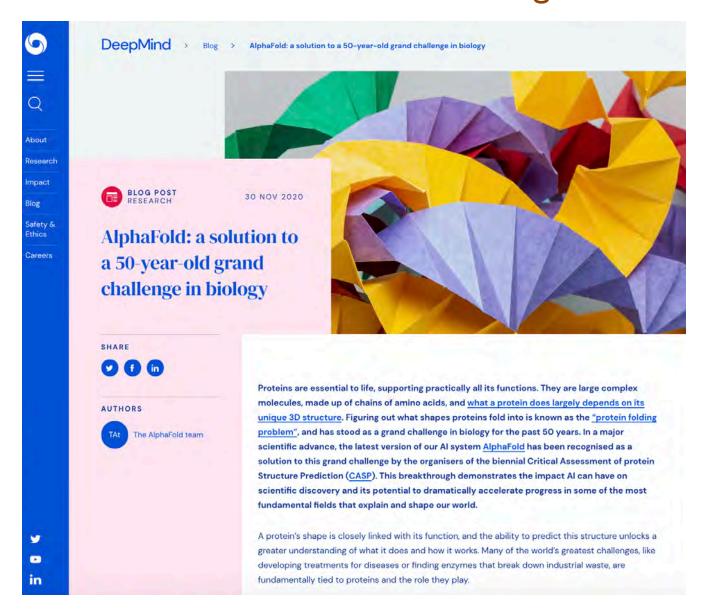




Gene-for-gene hypothesis

Tyler B. (2001). Trends in Genetics. 17(11):611-614

Imagine a world where you can begin to model protein ligand models for disease resitance genes!



The Next Green Revolution will be "Data" and "Design" driven

Agriculture has transition to a Data Science. Massive data generation—genotypes, phenotypes, soil & environment.

Data now exceeds human capacity to formulate and test hypotheses about gene function, regulatory networks, and predictive agriculture.

Develop new approaches and systems that can supply new hypotheses for researchers for crop breeding, fermentation systems, solar energy capture, pest and disease management.

Adapt what has evolved in nature and design the space nature missed



Norman Borlaug Nobel Peace Prize 1970



Ware Lab

Kapeel Chougule Nick Gladman **YInping Jiao Vivek Kumar** Sunita Kumari Forrest Li **Augusto Lima Dinz** Zhenyuan Lu **Andrew Olson** Michael Regulski **Bo Wang Liya Wang** Xiaofei Wang Marcela Tello Ruiz **Sharon Wei** Peter Van Buren **Lifang Zhang**

CSHL

Carol Hu

Dick McCombie Rob Martienssen Dave Jackson Tom Gingeras Dave Micklos

Uplands Farm Tim Mulligan Kyle Schlecht

HPCC resources Todd Heywood

Unraveling Complex Traits using Genomic, Genetic, Systems & Computational Approaches









CSH Spring
Harbor
Laboratory

Advancing Agriculture Through Collaborative Research on Crop & Model Species