The Generation and Analysis of Genomics Information Forms the Foundation for Efficient Virginia-Type Peanut Cultivar Development

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Introduction

- Peanut (Arachis hypogaea L., 2n=4x=40) is a staple crop that is planted and consumed globally as a source of protein and oil
- In the United States, 2,782,290 MT of peanuts were produced in 2020¹
- Runner-Types, Virginia-Types, Spanish-Types, and Valencia-Types, are grown on 85%, 10%, 2%, and 1% of United States peanut acreage respectively²



Figure 1. Peanut Market-Types: Runner, Valencia, Spanish, Virginia (Listed from left to right³)

- NCSU has been involved in Virginia-Type peanut breeding and improvement since the 1930s⁴
- Breeding actives within the NCSU Peanut Breeding and Genetics (PB&G) program were informed by visual ratings and observations.

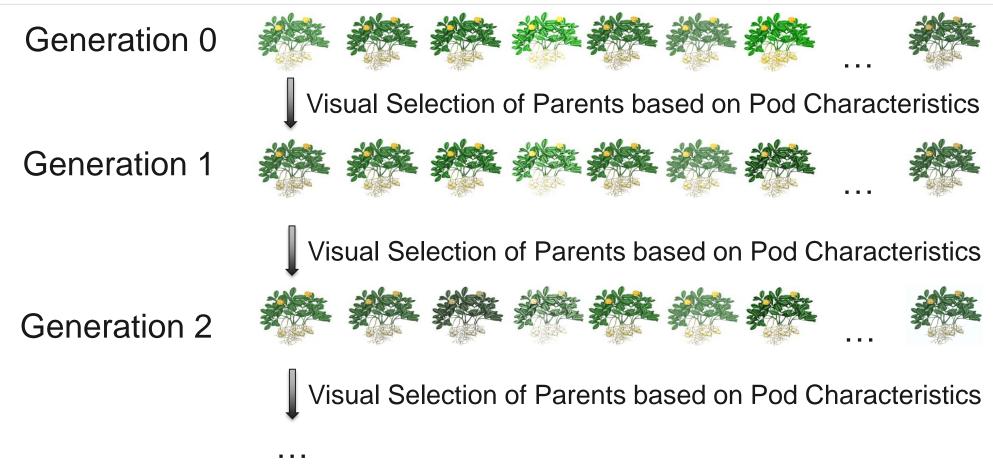


Figure 2. Example of recurrent visual selection for breeding peanut⁵ cultivars used in the NCSU PB&G program

 Novel mating schemes, which added genes from wild species into the peanut breeding population is an vital part of the PB&G program history. The genes from wild species allowed the cultivars produced from NCSU to be resistant to economically damaging pathogens.

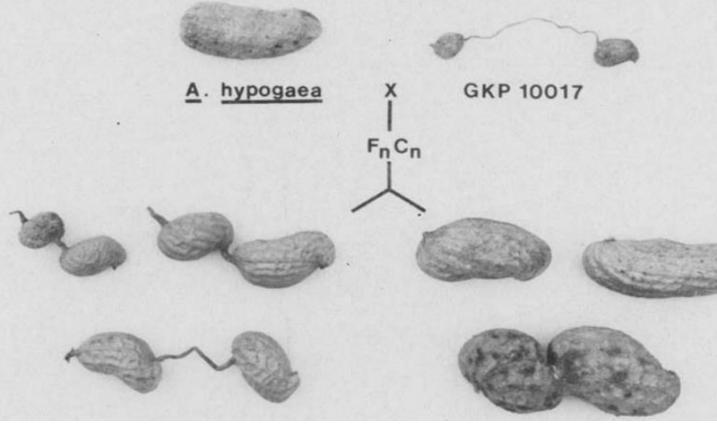
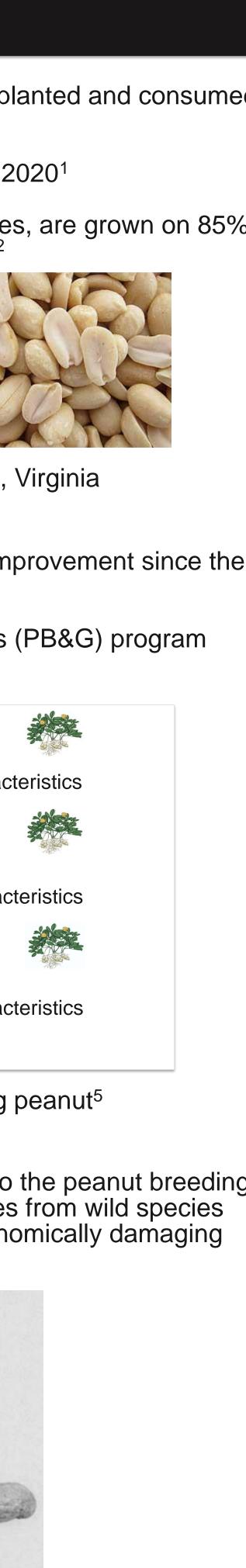


Figure 3. Photograph⁶ showing the result of a wild species (*A. cardenasii*) mated with cultivated peanut (*A. hypogaea*) at NCSU in the 60s

- Contemporary genomic data are cheap, high volume and can be generated quickly
- Using genomic data in breeding is known as genomics-assisted breeding. Integrating genomic technology leads to more efficient and accurate cultivar development.

Objectives

- A) Create Genomic Resources for the NCSU PB&G Program
- B) Use Genomic Data to Characterize Peanut Lines within the Breeding Population
- C) Characterize Historical Wild Species Introgression Events
- D) Generate the Information Needed for Genomics-Assisted Breeding in the NCSU PB&G Program

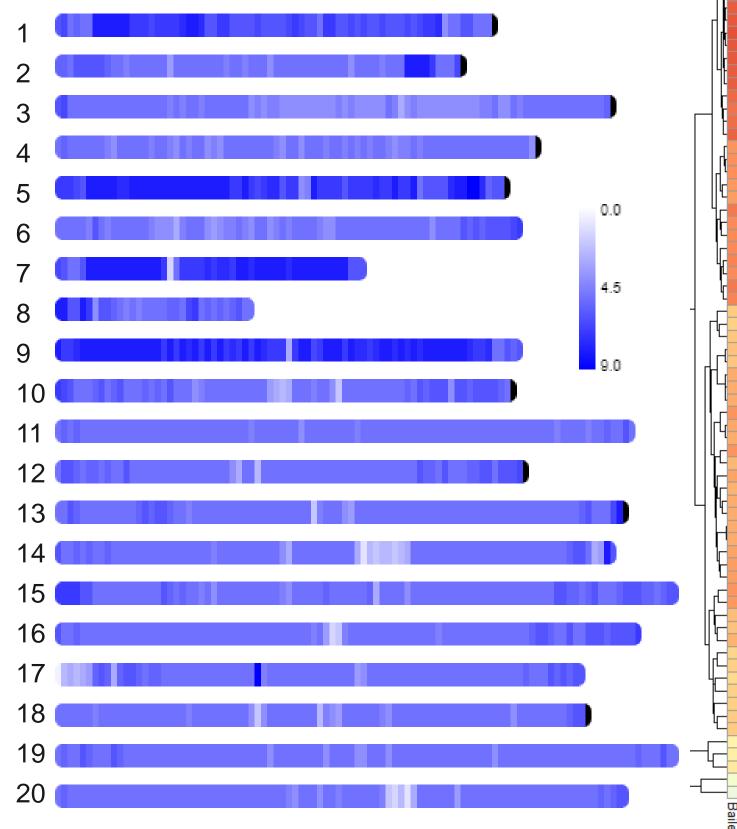


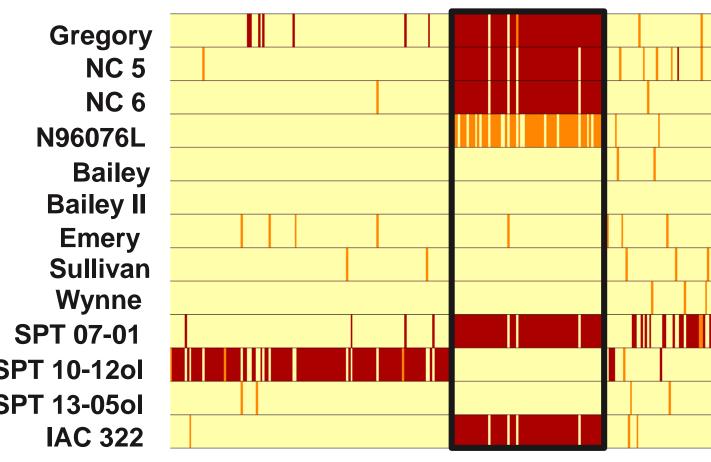


Materials and Methods

- Cultivar Bailey II was chosen to represent Virginia-Type peanut as the reference genomic sequence. This cultivar has excellent product shelf life, outstanding agronomic performance, and good disease resistance to fungal pathogens.

 agronomic performance, and good disease resistance to fungal pathogens. Genome assembly was done with a series of bioinformatics software packages which took raw genomic sequence data and assembled the data into large pieces. Those pieces were further corrected and connected into chromosome-scale linear sequences. The genome was annotated using a modified BIND⁷ pipeline. Variant discovery, i.e. discovering genetic differences, was done for 66 peanut lines, of historical and agronomic significance to the breeding program. Variant discovery was largely done following GATK⁸ best practices. 			Figure 4. The popular bailey II peanut cultivar, chosen for whole genome sequencing and assembly	 represents a large enhancement in terms of contiguity Greater than one million high quality variant sites were identified throughout the genome for the breeding population B) Use Genomic Data to Characterize Peanut Lines within the Breeding Population Markers discovered serve as 'fingerprints' for different individuals The genomic relationship between individuals was quantified C) Characterize Historical Wild Species Introgression Events
Bailey II Genome Sequer	<u>nce:</u>			 Genomic blocks were identified and delimited in better resolution when what was previously possible
Table 1. Comparison of the contextualize the quality of		bly with other cultivated peanut a	ssemblies as a way to	D) Generate the Information Needed to Launch Genomics-Assisted Breeding in the NCSU PB&G program
Genome Assembly	Number of Contigs	Sequence Length (bp)	Complete BUSCOs	Genomics Data
Bailey II Tifrunner (v2) ⁹ Shitoqi ¹⁰ Fuhuasheng ¹¹	591 3496 6289 31747	2,532,481,151 2,534,699,547 2,506,086,556 2,483,343,588	5181 5180 5174 5078	Accurate Sample TrackingGenomic Prediction of Agronomic PerformanceInformed Parental SelectionContamination Screening
		A 83% decrease in the number compared with Tifrunner v2	er of contigs when	Enhanced Development of Peanut Cultivars!
Genomic Variant Discove	ery:			Figure 7. Flowchart demonstrating how the genomic information in this
3 4 5 6 6 7 8 9 9 10 11 12 13 14 15 16 16 17 18 19 20 Figure 5. Twenty hypogae colored by marker density of 500,000 base pairs. Ma been scaled by natural log Evidence of Wild Species	within windows Figur orker density has import organ	e. 6. Genomic state identity matrixant to the NCSU PB&G program ized by Euclidean distance	x of individuals	 PB&G program References Crop Production 2020 Summary 01/12/2021. Crop Prod. 125 (2020). Anthony. Peanut Types and Production. American Peanut Council https://www.peanutsusa.com/about-peanuts/the-peanut-industry3/9-peanut-types-and-production.html. Market Type Photograph. https://www.aboutpeanuts.com/peanut-facts/95-types-of-peanuts. 2019 Virginia-Carolinas Peanut Promotions North Carolina State University, College of Agriculture and Life Sciences, Peanut Breeding Program Records, 1938-1992. Keith Ward. Image. https://www.motherearthnews.com/organic-gardening/growing-peanuts- zw021312zsto/. Stalker, H. T., Wynne, J. C. & Company, M. VARIATION IN PROGENIES OF AN ARACHIS HYPOGAEA x DIPLOID WILD SPECIES HYPRID'. Euphytica 28, 675–684 (1979). Jing Li, Urminder Singh, Priyanka Bhandary, Jacqueline Campbell, Zebulun Arendsee, Arun S Seetharam, Eve Syrkin Wurtele, Foster thy young: enhanced prediction of orphan genes in assembled genomes, Nucleic Acids Research, 2021; gkab1238, https://doi.org/10.1093/nar/gkab1238 Poplin R, Ruano-Rubio V, DePristo MA, Fennell TJ, Carneiro MO, Van der Auwera GA, Kling DE, Gauthier LD, Levy-Moonshine A, Roazen D, Shakir K, Thibault J, Chandran S, Whelan C, Lek M, Gabriel S, Daly MJ, Neale B, MacArthur DG, Banks E. (2017). Scaling accurate genetic variant discovery to tens of thousands of samples bioRxiv, 201178. DCI: 10.1101/201178 Bertoli, D.J., Jenkins, J., Clevenger, J. et al. The genome sequence of segmental allotetraploid peanut Arachis hypogaea. Nat Genet 51, 877–884 (2019). https://doi.org/10.1038/s41588-019-0405- z Zhuang, W., Chen, H., Yang, M. et al. The genome of cultivated peanut provides insight into legume karootypes, polyploid evolution and crop domestication. Nat Genet 51, 865–876 (2019). https://doi.org/10.1038/s41588-019-0402-2 Xiaoping, Chen Qing Lu, et al. Sequencing of Cultivated Peanut, Arachis hypogaea, Yields Insights into Genome Evolution and Crop
Gregory NC 5 NC 6 N96076L Bailey Bailey II Emery Sullivan Wynne SPT 07-01 SPT 10-120I SPT 13-050I IAC 322				934, ISSN 1674-2052, https://doi.org/10.1016/j.molp.2019.03.005. Funding Severice Service Severice Severice Severice Service Severice Severice Severice Severice S
dark red and orange within	the black box represents for low represents the baseline	s (represented as rows) across c preign genetic material from the e peanut genome. These blocks	wild species mating event,	VORTH LA GAOVALINAT NOCESED FIELD INSPECTED VOINN PEDIGARE









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Discussion and Conclusion

A) Create Genomic Resources for the NCSU PB&G Program

- Genome reference sequence was generated and represents a large enhancement in terms of contiguity

