Improving the Production of Oil in an Underutilized Crop: *Crambe hispanica*

A work in progress

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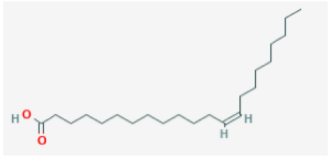
Intro to Crambe Hispanica

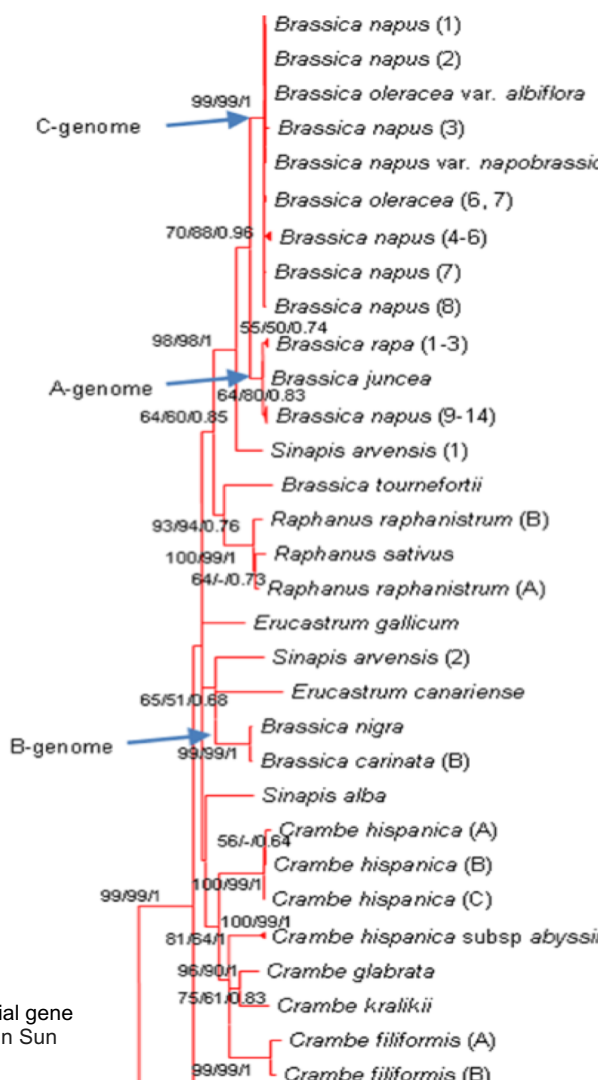
- **History**
  - Native to mediterranean area
  - Official domestication in 1933
- **Characteristics**
  - Cover crop potential
    - Thrives in no-till soils
  - High yield potential
  - Natural Resistance (halophytic)
  - High concentration of Erucic acid in seeds (55-60%)
- **Underutilized**

Erucic acid

- A monounsaturated omega-9 fatty acid
- Known crop genus containing Erucic acid
  - Brassica
  - Crambe
- Used in
  - **Biofuel**
  - Detergent production
  - Ink production



https://pubchem.ncbi.nlm.nih.gov/compound/Erucic-acid



Phylogeny of FAE1 trait (a crucial gene in erucic acid synthesis) (Xiaoqin Sun et al., 2013)

Crambe Hispanica's potential as a biofuel source

- **Issues with biofuel and how Crambe may combat these challenges**
  - *Land impacts/deforestation*
    - Crambe as a cover crop would reduce land area needed
    - Thrives in high salt soils meaning it can grow on a larger variety of land (Primarily Coastal regions)
  - *Fuel crystallizing and clogging vehicle filters (Cold regions)*
    - Erucic acid has a much lower "Cloud point" than standard oil seeds, meaning the fuel will not crystallize as easily in colder regions. (Grewal et al., 1993)
  - *"Food vs Fuel" (Land for fuel replacing land for food)*
    - Crambe may double as a food source, as its leaves/stem are rich with glucosinolates.

Improving Erucic Acid Synthesis in *Crambe hispanica*

Background

- Previous research in related species (Canola) revealing Erucic acid synthesis pathways has significantly contributed to producing high oil quality and content (Lühs et al., 1999; Bao, Pollard, and Ohlrogge, 1998). Similar methods may also work on Crambe hispanica.

https://www.healthline.com/nutrition/is-canola-oil-healthy

Aim

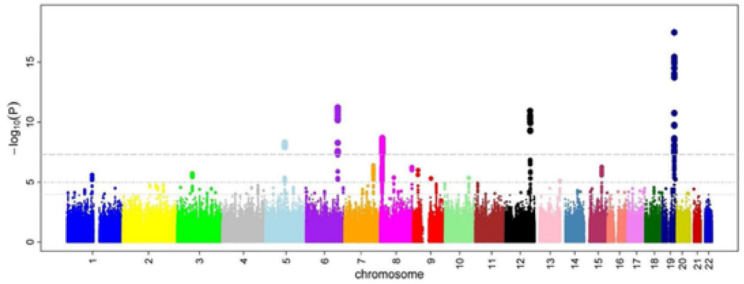
- Contribute data on genes responsible for Erucic acid synthesis to further facilitate Crambe hispanica as a productive biofuel crop.

For Whom?

- Plant breeders use this data when cross breeding and using Crispr-Cas9.

Genome Wide Association Study (GWAS)

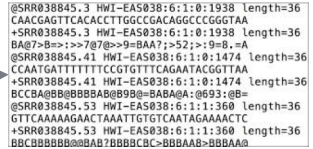
- **Background**
  - Statistical method use in population genetics to identify correlations between single nucleotide bases (SNPs) and phenotype.
  - Outliers signify statistical significance
- **Application in Crambe Research**
  - Using a diverse population of known accesions, we may compare erucic acid levels in seeds to SNPs originating from tissue samples (leaves).



Methods/Timeline

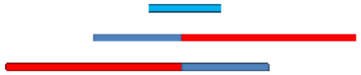
October
Sample leaf tissue and perform RNA isolation

November



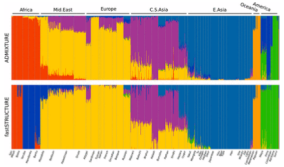
Generate transcriptomes for alignment (RNA-seq)

November



Trim fastq reads using Trimmomatic and map the clean reads to reference genome with HISAT2

December



Call SNPs with GATK and run Faststructure to observe genetic structure

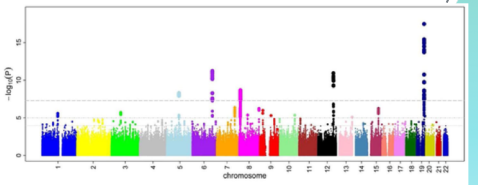
February

Identify the major genes that regulate *C. hispanica* erucic acid biosynthesis.

August
Select 96 diverse accessions

December

Sample seeds for fatty acid composition analysis (GC-MS)



Conduct GWAS analysis using SNPs and phenotypic data (seed fatty acid traits)

January

References

Cover crop - http://www.scielo.org.co/scielo.php?script=sci_arttext&pid=S0304-28472018000208517

Erucic oil content (>55%) - https://www.frontiersin.org/articles/10.3389/fpls.2019.01442/full

Erucic oil oxidation stability - https://www.sciencedirect.com/topics/agricultural-and-biological-sciences/erucic-acid

Crambe Phylogeny - https://journals.plos.org/plosone/article?id=10.1371/journal.pone.0083535

References cont.

Lühs, W. W., A. Voss, J. Han, A. Gräfin zu Münster, D. Weier, F. P. Wolter, M. Frentzen, and W. Friedt. 1999. "Genetic Modification of Erucic Acid Biosynthesis in Brassica Napus." In *Genetics and Breeding for Crop Quality and Resistance: Proceedings of the XV EUCARPIA Congress, Viterbo, Italy, September 20–25, 1998*, edited by G. T. Scarascia Mugnozza, E. Porceddu, and M. A. Pagnotta, 323–30. Dordrecht: Springer Netherlands.

Bao, X., M. Pollard, and J. Ohlrogge. 1998. "The Biosynthesis of Erucic Acid in Developing Embryos of Brassica Rapa." *Plant Physiology* 118 (1): 183–90.

Grewal, V.S., Ramamurthi, S. & McCurdy, A.R. Synthesis and properties of erucic acid triacylglycerols. *J Am Oil Chem Soc* 70, 955–959 (1993). https://doi.org/10.1007/BF02543020