

# Project Overview

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## Introduction

This document is a high-level overview of the experimental design and objectives of the “Jay” yield tests.

## Objectives

The overall objective of this experiment is to identify recombinant inbred lines (RILs) that have yield and agronomic qualities comparable to existing check cultivars, and have a protein + oil content that is greater than the protein + oil content of the check cultivars.

## Experimental design

The experiment consists of two tests, names test 1 and test 2, that are comprised of 15 RILs, and 5 check cultivars. These two tests were grown in a RCBD in 2020 and 2021, in two locations for each year. In 2020, each test was grown at the Clayton (CLA) and Caswell (CAS) research stations, while in 2021 each test was grown at the Plymouth (PLY) research station and CAS. Each test was grown in four replications within each location for both years.

## Population development

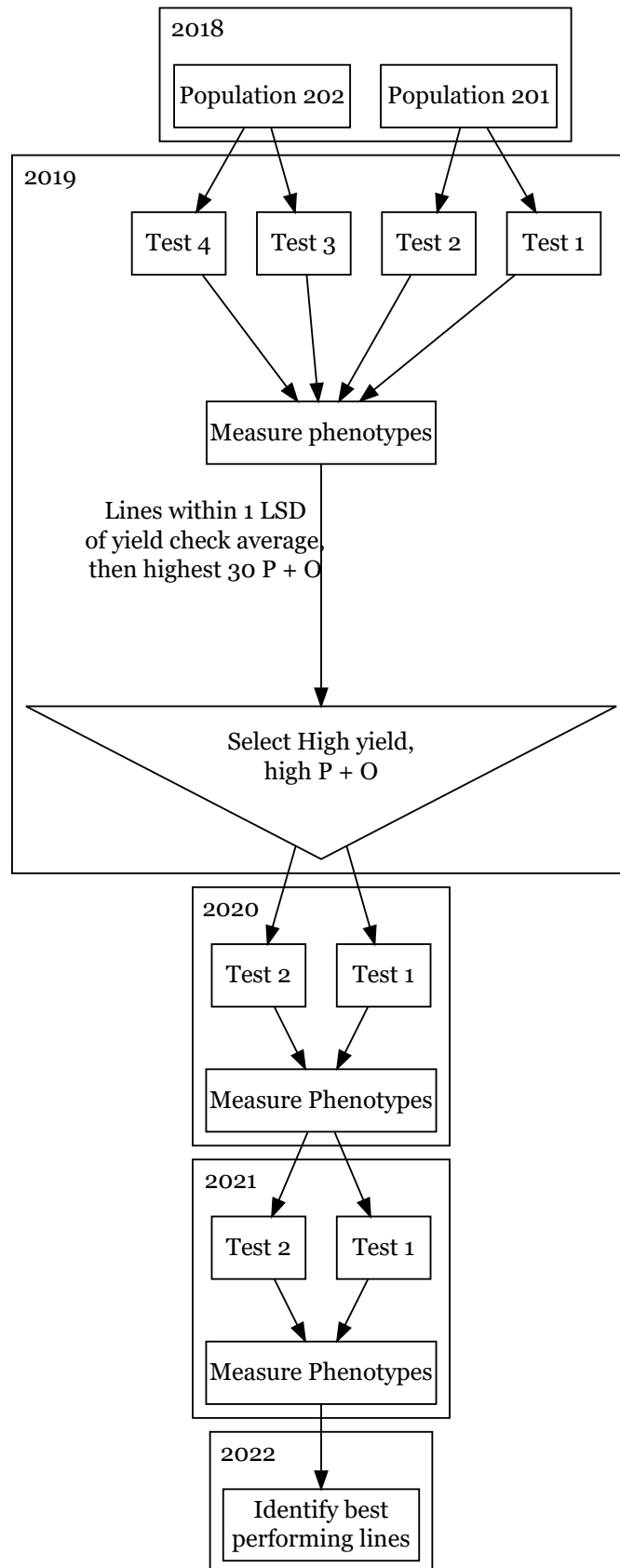
The RILs used in these two tests come from two mapping populations that were initially grown as plant rows in 2018 at CLA. We noticed some of the lines in these two populations had some good combinations of agronomic qualities, and seed composition traits and this is how we first got the idea to more rigorously evaluate the yield performance potential of a selection of the lines from these two populations. These two populations were too large to test every line so we first made some selections to identify 80 lines from each population that would be tested for yield performance. Briefly, this selection scheme first removed any lines from consideration that had poor lodging, seed load, or bulk weight, and any lines with extreme maturity dates. Lines were then selected so that the entire distribution of protein and oil content of the starting mapping populations would be represented in the selection. The 80 lines for each population were then split into two tests of 40 RILs based on maturity date. Three check cultivars and the mapping population parents were then included in the tests as well for a final set of four tests with 45 lines each.

These four tests were grown in PLY and CAS in 2019. Agronomic notes including lodging, height, maturity date, and an agronomic score based on overall appearance were taken in the field and yield, seed weight, protein, and oil content were measured after harvesting.

After this data was collected, another round of selection was done that focused on the observed yield and agronomic performance of the lines. Lines with poor agronomic qualities like significant lodging or poor pod load were first excluded from advancement. Then, only lines that were within or above a least significant difference of the average of the checks for each test were kept for consideration. Then, the thirty lines with

the highest protein + oil content across all four tests were identified from among those that passed the yield threshold. These thirty lines were then split into two halves based on maturity date. Five checks were then added to each test for two final tests of 20 genotypes. These two tests were then grown over two years in the design from the section above. The same phenotypes were measured in field and after harvest that were measured in the preliminary trials in 2019.

I've added a small diagram of this overall development in the flowchart below.



## Collected data

The main traits we're interested in analyzing/summarizing are yield and the seed composition traits seed protein and oil. We do however have data on several agronomic traits including height, maturity date, seed weight, seed quality, and lodging. Yield was measured for all reps for both years while seed protein and oil were measured for reps 1-2 for both years.

## Analysis and questions

We used a linear model to analyze the data with the formula:

$$y = \mu + GEN + ENV + GEN : ENV + REP(ENV) + \epsilon$$

Where  $y$  is the value of some phenotype,  $\mu$  is the overall mean,  $GEN$  is the genotype effect,  $ENV$  is the environment effect,  $GEN:ENV$  is the genotype x environment interaction effect,  $REP(ENV)$  is the effect of replication nested within environment, and  $\epsilon$  is the measurement error. This model was used to perform an analysis of variance and to obtain genotype least square means.

Going back to the overall goal of the experiment, we want to identify genotypes which have a yield comparable to or superior to that of some existing high-yielding cultivars, and seed composition traits that are superior to those high-yielding cultivars. The criteria by which we can declare that a RIL has these qualities is where I have the most questions. I've seen a few papers that used a least-significant difference value to compare means of new cultivars to other established cultivars and was wondering if this would be an appropriate test to use. I have also read a fair amount of criticism for using the least significant difference to compare means since it doesn't correct for multiple comparisons. Because of that, I was considering using Tukeys HSD instead, or another correction to the standard LSD test to account for multiple comparisons. We also used multiple check cultivars within each test, but as far as I can tell the multiple comparison statistics are only strictly appropriate for comparing the means of one cultivar with the means of one other, and I didn't know how appropriate it would be to use a LSD/HSD value to compare the mean of one RIL with the average of the checks for each test.