Selection of *A. stenosperma*-derived advanced lines with strong resistance to LLS using association analyses

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This project builds on a previously developed resource: a BC_3F_2 population derived from (*Arachis batizocoi* x *A. stenosperma*)^{4x} crossed and backcrossed with peanut elite locally adapted lines. This induced allotetraploid has resistances to various peanut pests and pathogens (Leal-Bertioli et al., 2021). Individuals were selected with markers linked to RKN resistance (Ballen et al., 2019), each individual is more than 90% elite peanut with a small proportion of genetic material from *A. stenosperma*. We observed the plants also segregated for late leaf spot (LLS) resistance in greenhouse. We proposed to perform genetic analysis, made possible by the peanut genome sequencing project, on selected advanced lines of this population to identify the genetic regions that confer the resistance, create specific LLS resistance associated DNA markers, and use these in breeding to advance and improve lines.

The first analysis calculated the correlation between LLS incidence severity in the greenhouse with genetic markers. This indicated that the presence of wild segments on chromosome A06 and the top of chromosome B02 may be associated with resistance to LLS (Figure 1). With this analysis, we selected families to perform the genetic study including resistant plants with low wild DNA introgression.

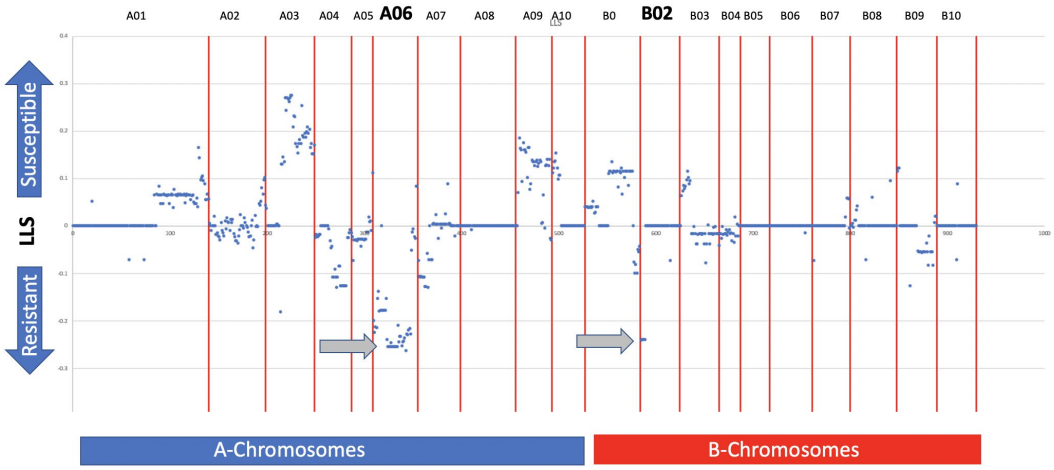

Figure 1: Graph of LLS greenhouse screening x SNP markers

In the spring 2020, these selected BC_3F_2 families were planted in the field at Midville. Plants were placed on 2x2m grids, well-spaced, so that we could observe disease resistance and growth habit of each plant. Leaf material was collected from all plants at the beginning of the season for DNA extraction. This year TSWV was very high and LLS pressure was low. Nevertheless, highly disease resistant and productive plants were selected to advance a generation and test in the field next year. To complement field assays, from these same families, we did *in vitro* assays for LLS, Early Leaf Spot and Rust. These experiments were able to identify lineages which are resistant to both Early Leaf Spot and Rust, but the quality of inoculum only allowed low confidence results to be produced for LLS.

In summary, we were able to show that this same genetic population has resistance to Early Leaf Spot, Rust and, very probably TSWV. We remain confident that this material has very promising LLS resistance. However, because of the vagaries of field conditions and *in vitro* assay results, we were unable to refine our genetic analysis this year. Especially because of the very promising results with Early Leaf Spot, Rust and TSWV, we remain confident that this work will lead to cultivars with much improved disease resistance, allowing for a reduction of spraying regimes.