

Final Report for 2019 National Peanut Board funding to the Texas Peanut Producers Board.

I. Subject area: Molecular Genetics & Breeding

Project Title: Breeding to Increase Peanut Yields and Production Efficiency by Developing Breeding Lines with Improved Drought and Heat Tolerance combined with Multiple Disease Resistance

Funding Year: 2019

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Sub-Project I-1 Multiple Disease Resistant Runner-type trials

The TAMU Peanut project had replicated yield trials located in South Texas, (Pearsall, Dilley, and Derby), in Central Texas (Proctor) and in North West Texas, (Seminole and Seagraves). We conducted 6 Advanced Line Tests (ALT's) across Texas in 2019 as well as one replicated screening nursery for Sclerotinia resistance. We continued to move lines developed for drought tolerance, into the more widespread ALT test in order to get more accurate information on each lines performance across the state. A total of 13 breeding lines were included from the drought tolerance program. There are also 3 lines developed for Root knot nematode resistance and Sclerotinia resistance, as well as 4 commercial checks Tamrun OL11, AG18, Georgia 09B, and Georgia 14N. Table 1 is a combined analysis of the ALT's across Texas in 2019. Six of the 7 lines in the top statistical grouping all came from the drought crossing program. It should be noted that the lines developed for drought tolerance are some of the first generation of crossing completed for the drought project and while they are not the most drought tolerant lines in the project, they appear to have excellent yield potential under normal irrigation to the point that they continue to perform at the top of the tests. The best performing lines are currently being used as parents for development of future breeding lines.

Table 1.
Combined Analysis of Advanced Line Yield Trials Across Texas in 2019

Entry	Lbs/a	%TSMK	Value/Ac	g/100 sd
TxL100212-03-03	5786.3 a	72.2 e-g	1013.39 ab	70.1 cd
TxL100212-05-09	5770.6 ab	72.2 e-g	1030.28 a	73.7 ab
Tx144370	5759.5 ab	74.0 b-e	1040.61 a	65.8 e-g
TxL100212-02-05	5588.7 a-c	71.4 fg	984.96 a-c	71.0 bc
TxL100225-03-08	5565.9 a-d	74.3 b-d	1021.05 a	60.5 i
TxL100225-03-13	5438.6 a-e	71.4 fg	960.76 a-d	59.5 i-k
TxL100212-07-08	5427.4 a-e	72.4 d-g	969.02 a-d	67.5 d-f
TxL100212-05-03	5324.1 b-f	72.9 c-f	954.35 a-d	70.9 bc
Tx121082	5291.9 c-f	75.5 ab	982.51 a-c	63.3 gh
TxL100212-03-06	5269.1 c-f	74.6 bc	967.14 a-d	67.8 de
Tx144342	5242.5 c-f	73.0 c-f	942.43 a-d	71.9 a-c
Tx144485	5196.2 c-f	73.7 b-e	944.66 a-d	73.8 a
TxL100225-03-04	5124.1 d-g	72.9 c-f	920.31 b-d	56.9 kl
TxL100212-02-06	5119.0 d-g	71.4 fg	904.67 cd	61.6 hi
TxL100212-03-08	5073.1 e-g	70.6 g	892.13 cd	64.8 fg
Georgia 09B	5031.6 e-g	72.2 e-g	881.15 d	66.7 ef
Georgia 14N	5029.2 e-g	77.2 a	954.66 a-d	60.2 ij
Tamrun OL11	4963.0 fg	75.1 b	914.11 b-d	65.7 e-g
TxL100212-07-12	4726.6 g	66.1 h	778.31 e	54.7 l
TxL100212-07-07	4726.4 g	67.6 h	777.53 e	57.7 jk
Mean	5285.8	72.65	945.62	65.5
LSD	457.1	2.0	99.7	2.8
p≤0.05	≤0.0001	≤0.0001	≤0.0001	≤0.0001

Sub-Project I-2 Initial Drought Tolerant Runner-type yield trials

We continued testing three runner-type drought populations again in 2019 which represented the third year of replicated trials for this material. In addition to these original tests the best performing lines from previous years have been included in the statewide advanced line trials. The dedicated drought trials were tested in South Texas near Pearsall under full irrigation and in West Texas under reduced irrigation. The location for the 2019 trials for yield had two major events that led to inconsistency in the plot data. A heavy rain the night after plants lead to washing through the plots and a mid-season outbreak of Cotton Root Rot (*Phymatotrichum omnivorum*) in the field led to an almost total loss of the location. Plots were harvested but no significant differences were observed in yield data. Based on this an abbreviated table is presented for each of the 3 drought trials. In 2020 all of the lines will be tested under drought conditions again. In addition, the top performing lines based on 2017-2018 combined data will be merged into a single test for testing under normal irrigation. One positive result from this project this year was the collection of canopy cover data using Unmanned Aircraft Systems (UAS) on the diseased plots right before harvest. This represented Texas A&M Peanut Breeding programs new UAS program and will be used to develop disease rating algorithms.



Figure 1. Orthomosaic images generated from UAS imagery (left) and canopy cover (%) within each 0.9×0.9m grid calculated from the orthomosaic image

Drought #1 Yield Test

The Drought #1 Trial consisted of 23 breeding lines plus the donor parent for the drought tolerance trait (ICGV-76) and the elite parent which was the commercial line Tamrun OL11. The 2019 Drought #1 test heavily infested with Cotton root rot and the data collected was highly variable shown by the coefficient of variation (CV) of 47. Overall no statistical differences were observed in this test so only the top performing line and the commercial check Tamrun OL 11 are presented in an abbreviated table (Table 2).

Table 2. Drought #1 Yield Trial south of Pearsall, Texas in 2019

Entry	g/plot	%TSMK	g/100 seed
TXL100212-03-11	7171 NS	69.2	63.2
Tamrun OL11	4187 NS	69.1	64.9
Mean	5093		
p≤0.05	≤0.5858		

TxL100212-03-11 had the highest yield in the test at 7171 g/plot. However, twenty other breeding lines performed in the top statistical grouping for yield as well. Many of the lines in the test performed superior to the parent Tamrun OL11 which only yielded 4187 g/plot which was also in the top grouping. TxL100212-02-05, TxL100212-02-06, TxL100212-03-06 and TxL100212-03-08 and were also previously mentioned in the ALT (Table 1). These lines performed so well in 2018 that they were added to the multiple location ALT in 2019 to test them over multiple environments. Many of the top performing lines were moved forward for the 2020 testing.

Drought #2

There were 23 breeding lines and two parents (ICGV-76 & Tamrun OL11) in the Drought #2 Yield Trial. To conserve space only data from the top performing line and the commercial check Tamrun OL11v are shown in an abbreviated table (Table 3). TxL100212-05-09 had the highest yield at 7239 g/plot. There were ten breeding lines that performed in the top grouping for yield ranging from 5146 g/plot to 7239 g/plot. The commercial check Tamrun OL11 performed towards the middle of the test with a yield of 4984 g/plot. As mentioned earlier in the text, the 2019 Drought #2 test heavily infested with Cotton root rot and while the test did show statistical significance the data collected was highly variable which is evidenced by the coefficient of variation (CV) of 37%. Tamrun OL11 typically grades upper 70's at this site and as seen in Table 3 it graded 61.7 %TSMK. As above only the top performing line and the commercial check Tamrun OL 11 are presented in an abbreviated table (Table 3).

Table 3. Drought #2 Yield Trial south of Pearsall, Texas in 2019

Entry	g/plot	%TSMK	g/100 seed
TXL100212-05-09	7239 NS	67.6	64.4
Tamrun OL11	4984 NS	69.4	61.7
Mean	4597		
p ≤0.05	≤0.0014		

Drought #3 Yield Test

The Drought #3 yield trial had eighteen breeding lines and the two parents as check varieties. Six of the eighteen breeding lines performed in the top grouping for yield with TxL100225-05-07 having the highest yield at 7076 g/plot (Table 4). Grades for this test were even lower than the Drought #1& 2 with Tamrun OL 11 having an average grade of 65.5%. These lines have not performed as well as the lines from the other two tests in south Texas under full irrigation over the past three years and most will be eliminated for 2020.

Table 4. Drought #3 Yield Trial south of Pearsall, Texas in 2019

Entry	g/plot	%TSMK	g/100 seed
TxL100225-05-07	7076 NS	68.5	62.2
Tamrun OL11	5135 NS	65.5	60.1
Mean	4678		
p ≤0.05	≤0.0127		

Other Drought Tolerance Trials.

Runner trials.

We began testing a different set of materials in previous years, designed to combine tolerance to water deficit, nematode resistance, and the high oleic trait. We received NIFA funding for this project beginning in 2017, and so are continuing the project under that funding source, and data are not presented here except to summarize.

However, we made additional selections from breeding lines using markers for all three traits, and are presenting data for the selected breeding lines below (Table 5). This was the first year of the test. The trial was conducted in West Texas, at a target of 25% ET replacement (essentially dryland) from June through August, chosen to represent the times of greatest water deficit on farms with limited irrigation. The low yields, grades and seed weights demonstrate the degree of water deficit stress that the test was under. But what is important is that yields of several of the breeding lines were statistically higher than Tamrun OL11 and numerically higher than Flavorunner 458, suggesting that some of these could be more tolerant to water deficit than standard varieties. Of particular interest are lines TxL144301-128 and -027, which were in the top 3 for yield and also had the highest 100 seed weight and numerically the highest grades in the test. Grades under water deficit were numerically as good as or better than Tamrun OL11 and Flavorunner 458. The large gap between Total Kernels and TSMK is due to the presence of many small kernels that graded as Other Kernels, as there was either not enough water for many of the kernels to fill out, or development of pegs was delayed by lack of water so that the seeds could not mature by harvest. These conditions were more extreme than we expect growers to experience, but were chosen to stress the different breeding lines to find which can survive water deficit the best.

Table 5. Yield under water deficit stress, 2019.

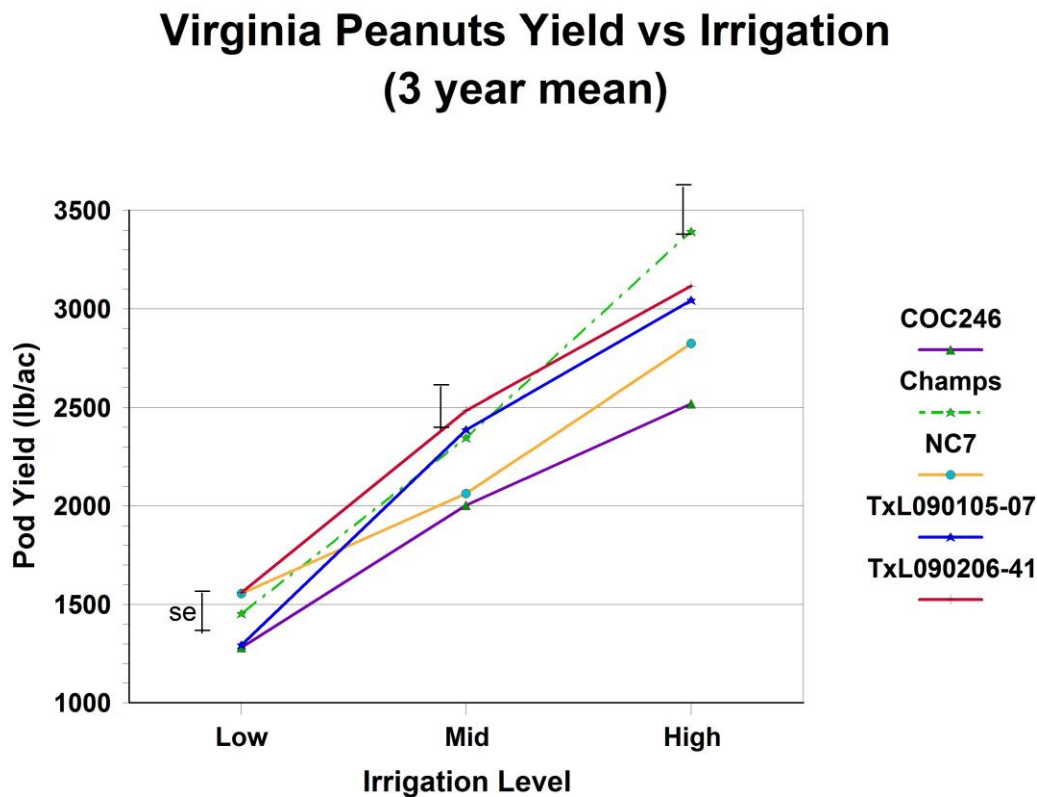
Accession	Pod Yield (lb/ac)	Shellout (%TSMK)	Shellout (%Total Kernels)	Seed Weight (g/100 SMK)
TxL144301-150	2666 a	28.3 ns	55.9 ns	39.7 f-i
TxL144301-128	2650 a	46.2	57.9	46.9 a-c
TxL144301-027	2644 a	36.0	54.7	46.1 a-d
TxL144301-171	2484 ab	29.2	57.9	37.5 g-i
TxL144301-016	2447 a-c	31.0	57.5	36.8 hi
TxL144301-100	2416 a-d	32.8	53.9	44.6 a-f
TxL144301-119	2401 a-d	29.0	54.8	41.9 b-h
TxL144301-166	2361 a-e	29.7	53.4	41.3 c-h
TxL144301-025	2347 a-e	24.8	53.7	42.4 a-h
TxL144301-003	2329 a-e	35.7	59.2	37.8 g-i
...				
FlvRun458	2068 a-g	33.3	57.8	39.2 f-i
TxL144301-117	2062 a-g	35.2	55.7	38.6 g-i
...				
TxL144301-158	1482 gh	32.4	53.3	41.3 c-h
TamrunOL11	1441 gh	31.8	56.0	37.2 hi
TxL144301-184	1169 h	35.9	56.4	40.0 e-i
Mean	2041	32.3	56.1	40.5
LSD	766	---	---	5.8
CV	23.1%	21.8%	5.9%	6.9%
p	0.013	0.274	0.423	0.008

These materials have also been photographed using weekly UAS overflights by Dr. Payton at the USDA-ARS in Lubbock. We expect that, when aerial images are analyzed and correlated with ground-based measurements and pod yield at harvest, this will give us new ways to measure and select for favorable responses of peanut to water deficit stress.

Virginia trials.

A multiyear test was made of several high oleic Virginia breeding lines, check varieties, and a minicore accession over 3 years. The test was grown under three irrigation levels - “H” high (75% evapotranspiration potential throughout the entire growing season), “M” mid (50% ET replacement from approx. 40 to 105 days after planting), and “L” low (a target of 25% ET replacement – essentially dryland 40 to 105 days after planting). Significant differences were found across irrigation treatments, with high oleic breeding line TxL090206-41 yielding the highest, and COC246 the worst (Figure 2). The high oleic release candidate TxL090105-07 as well as TxL090206-41 and Champs were in the top yield categories under high and mid irrigation levels. Differences under low irrigation were not significant statistically different, but TxL090246-41 was numerically at the top. We have planted an expanded test with more experimental entries and check varieties in 2020 at two locations under low irrigation to determine whether additional data will give evidence that one or more of the Virginia breeding lines can be released as drought-tolerant varieties that will be useful to growers who have less irrigation water available.

Figure 2. Graph of pod yields of Virginia accessions over 3 years under 3 irrigation treatments. Irrigation levels were “H” high, “M” mid, and “L” low.



Current Runner releases

We have received final approval from the Texas A&M plant release committee for a proposal for release of a high yielding, high grading runner varieties, to be named AG18. It is a high-yielding, high grading, high-oleic runner-type peanut cultivar released from Texas A&M AgriLife Research with good resistance to Sclerotinia blight (*Sclerotinia minor* (Jagger)). It exhibits yield and grade attributes equal to or better than Georgia 06G and Georgia 09B and increased consistency when compared to Tamrun OL11. AG18 seed size is smaller than that of Tamrun OL11 and similar to Florunner. No differences in flavors were noted in flavor analysis between AG18 and Tamrun OL11. In both 2017 and 2018 we conducted a 1 acre breeders seed increase of the variety. This seed was delivered to Texas A&M AgriLife Foundation Seed and a 55 acre increase was conducted in 2019. In 2020, we submitted for release NemaTAM II to the Texas A&M plant release committee. NemaTAM II is a high yielding, high oleic nematode resistant Runner variety.

We have also submitted for release Tamrun OL18L which is a high oleic early maturing runner with a large seed form our early maturity program, and Tamrun OL19 which has a somewhat smaller seed. Early maturing runner. Tamrun OL18L was submitted for release in 2019 with approval being given in 2020. We will need to increase breeders seed in 2020 to have sufficient seed to send for production of Foundation Seed. Tamrun OL19 was submitted for release in 2020, with approval pending.

We have also submitted a high oleic nematode resistant runner, NemaTAM II, from our multiple disease resistance program for release in 2019. Some of these lines are being used in crossing programs prior to release to cross with drought and heat tolerant accessions.

Future Runner Releases

The Tx144300's were developed for resistance to Root knot nematodes and Sclerotinia. While they performed lower in yield to the drought lines mentioned above, Tx144370 has performed well in South Texas which is where they were developed to give growers a nematode resistant line with better characteristics than the previously released Webb and NemaTAM II (under review) variety. These two lines have yielded from 400-600 lbs/a better than Webb and graded 1-3 percentage points higher. Additionally, they have a much shorter growth habit than NemaTAM II and a slightly smaller seed size in most of the trials over the past four years. The decision was made to move forward with the breeding lines for release. Tx144370 is in initial purification and increase in the 2020 season.

Spanish-type Yield Trials

We continued testing advanced Spanish-type yield trials in West Texas during the 2019 growing season. The tests consisted of 17 advanced Spanish breeding lines and three commercial checks: OLin, Schubert, and Tamnut OL06. In order to conserve space only the top statistical grouping for yield is presented. This represents the 3 year of testing for this population.

In 2019 fourteen breeding lines and two checks, OLin, and Schubert performed in the top grouping for yield with TxL076221-34 having the highest numerical yield at 5,047 lbs/a (Table

6). The top two yielding lines TxL076221-34 and TxL076239-16 graded equal to the commercial checks Schubert and OLin with grades of 70.92, 71.08, 71.1 and 71.7 respectively. TxL076224-08 had the highest numerical grade although it was not statistically higher. TxL076239-16 was significantly larger than either Schubert or OLin with g/100 seed of 56.0, 48.6 and 52.4 respectively.

Table 6. Analysis of Advanced Spanish-type Yield Trials in West Texas 2019

Entry	Lbs/a	%TSMK	Value/Ac	g/100 sd
TxL076221-34	5047.4 a	70.92 a-c	885.94 a	51.733 cd
TxL076239-16	4971.9 a	71.08 a-c	865.14 ab	56 ab
TxL076225-48	4947.6 a	69.213 b-d	849.08 a-c	50.033 d-f
Schubert	4908.6 a-c	71.133 a-c	858.73 ab	48.6 d-f
TxL076224-08	4892.4 a	72.787 a-c	873.97 a	51.533 cd
Tx144932	4489 ab	64.267 f	726.41 a-e	47.133 e-g
TxL076236-04	4445.2 a-c	70.573 a-d	772.07 a-d	52.267 b-d
OLin	4429.4 a-c	71.733 ab	778.88 a-d	52.467 b-d
TxL076225-28	4359.4 a-c	72.013 ab	773.23 a-d	48.8 d-f
TxL076229-53	4340.9 a-c	70.2 a-d	751.7 a-e	49.567 d-f
TxL076239-12	4198.3 a-c	70.16 a-d	726.4 a-e	52.433 b-d
TxL076221-06	4170.2 a-c	70.667 a-d	725.81 a-e	48.333 d-f
TxL076225-24	4158.5 a-c	71.253 a-c	730.23 a-e	50.333 c-f
TxL076224-24	4155.8 a-c	70.76 a-c	720.73 a-e	54.367 a-c
TxL076224-15	4088.9 a-c	68.067 c-e	685.07 b-e	48.767 d-f
Mean	4251	69.88	734.06	50.2
LSD	965.13	3.19	182.39	4.19
p≤0.05	≤0.0014	≤0.0001	≤0.0017	≤0.0001

Virginia Type Yield Trials

Testing of Virginia type lines continued in 2019. A replicated trial with 7 breeding lines and 3 checks was planted in West Texas and evaluated for yield and grade. Heavy pod rot pressure affected both yield and grade resulting in the test as a whole being highly variable. Analysis of Variance for yields for the test were not statistically significant, although very close ($p \leq .0526$). TxL09106-05 was the top numerical yielder in the 2019 test with a yield of 4059 lbs/a as compared to 3601 lbs/a for the commercial check Bailey.

In a separate location in West Texas without pod rot pressure, the top-yielding line TxL090105-38 had a yield of 2756 lb/ac, which was numerically but not statistically higher than Bailey at 2256 lb/ac. Other accessions yielded statistically similar but numerically lower than Bailey. These included TxL090105-07 and TxL090106-15.

In addition, selected lines were tested in South Texas as part of other projects. In South Texas testing TxL09106-15 and TxL090105-07 both performed numerically better than the commercial check Bailey with a yield of 6425 lbs/a, 6077 lbs/a and 6075 lbs/a respectively. At the present time TxL09106-15 and TxL090105-07 being considered for release based on several years of combined data.

Developing Wild Species Pathway for Introgression of Drought Tolerance

This project continues as part of the long-term drought project. It was initially funded through internal funding but has been absorbed into our overall drought program. The initial phase of this project was to identify 14 candidate genes associated with drought tolerance by conducting an imposed drought transcriptomics study. Specifically, we identified transcription factors that occur early in genetic pathways and represent excellent candidates for marker development. We are actively seeking additional funding to validate the presence of the candidate genes in the original drought tolerant species and to expand the study to include other related species. This project remains a vital part of our overall drought program. Transcriptomics is a powerful tool that can not only tell if a gene is present but can also determine how strongly a gene is expressed. Once the genes are validated for expression level then marker development can be conducted which will be used to aid in introgression of the genes into our elite material.

Crossing and chromosome doubling continues as part of the wild species introgression pathway is under development. When completed will allow the genes identified to be moved into the cultivated peanut. At this point we have made the initial cross and confirmed hybridization. The next step in pathway development is to double the chromosome number. This is proving to be very challenging. However, progress is being made and we remain hopeful. In addition, we are exploring alternate pathways using alternative species and or accessions to move the genes if the original route is not successful. One related complex hybrid has already been doubled and has cultivated materials that are being backcrossed into more advanced materials. Tamrun OL11 is the recurrent parent in this crossing program. Several more backcrosses are needed.

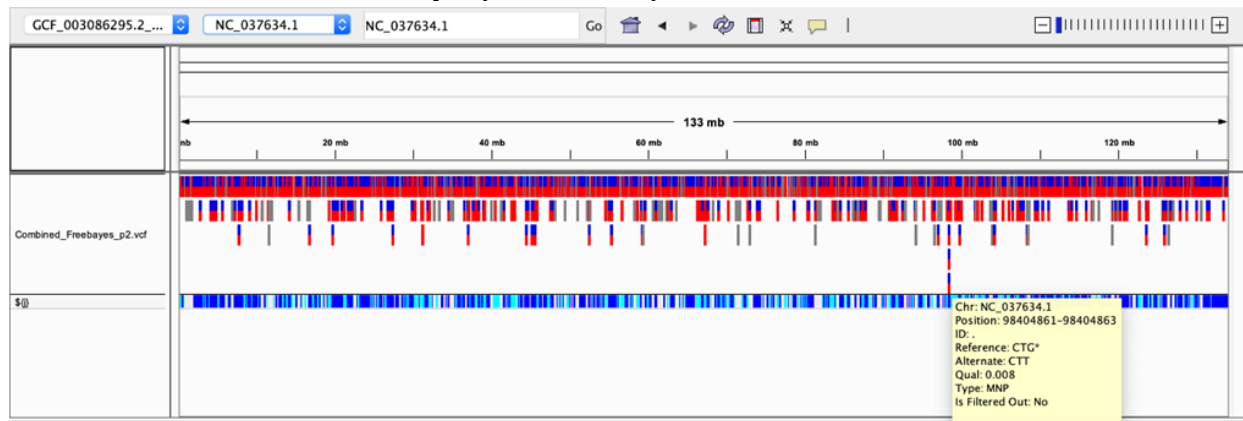
Identify Markers for Drought Tolerance in Mini-core Collection

We sent DNA of the minicore collection and additional accessions to the Texas A&M AgriLife Bioinformatics and Genomics Center for RAD-Seq sequencing. We received approx. 45 GB (gigabytes) (compressed) of sequence data back. Data are being analyzed, and preliminary results have been obtained using two software programs – Freebayes and GATK (Table 7). The significance of this type of analysis is that it is expected to give us a much more complete way to identify differences among peanut accessions than was possible using earlier types of marker analysis. But the data analysis is more complicated initially because of the far larger amounts of data (Figure 3).

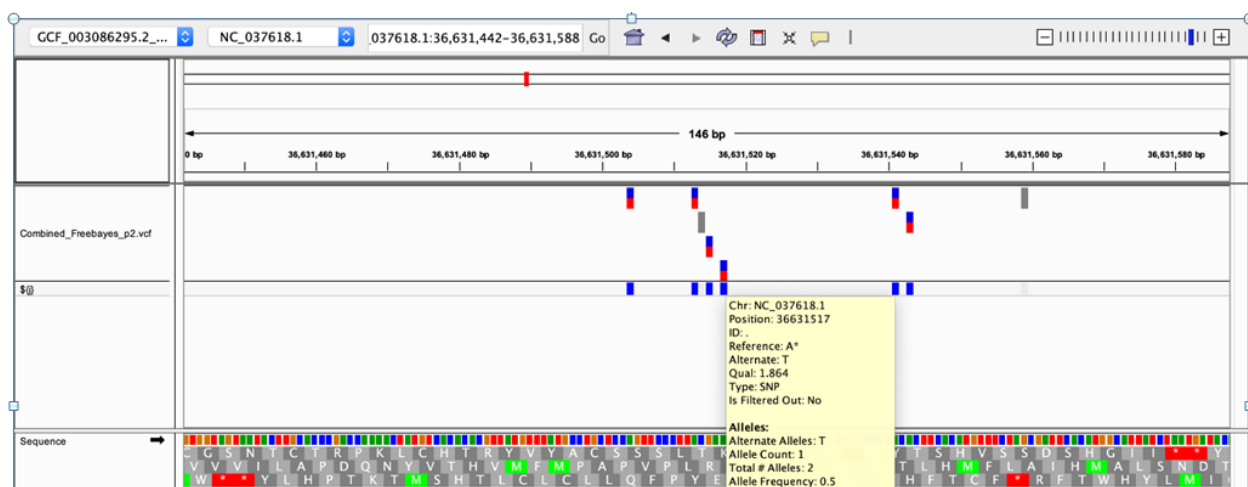
Table 7. Peanut mini-core RAD seq results: number of contigs and SNPs detected with and without filters by using Freebayes.

Criterion	Number of Chromosomal Sequence Contigs Mapped To	Number of SNPs
Raw SNPs	84	109,389
Filter for Quality Score > 10	58	43,113
Filter for Read Depth > 10	57	45,158
Filter for Allele Frequency > 0.5	68	37,974
Filter for all above	34	4,676

Figure 3. Examples of SNPs using the Integrated Genome Viewer. A. This shows an entire chromosome (136 million nucleotides), with the SNPs shown in the second line. Because of the scale, each bar may represent multiple SNPs, as can be seen in B.



B. This zooms in on a very small part of the chromosome, 146 nucleotides in all. It can be seen that there are 6 SNPs in this small region.



Analysis of an earlier set of data with lower read depth using GATK has given somewhat more successful results. After filtering, we obtained 3,265 SNPs. Although this is fewer than obtained by Freebayes, the data span 394 contigs, and the distance represented spans an estimated 18.3% of the genome. We expect that using the full data set and analysis with GATK will produce a larger number of SNPs and genome coverage at least as good as the preliminary analysis.

Accessions were also sent for analysis on the peanut community SNP chip (Figure 4). Of 47,837 SNPs, there were 9,000 polymorphic markers found in the minicore collection. Although this number is higher than the results obtained so far by RAD-Seq, because the SNP chip interrogates a single nucleotide only and lacks data on flanking sequences, it is expected that there may be more false positives due to the presence of unexpected closely-related (“paralogous”) that are present in other places in the peanut genome.

Figure 4. Examples of SNP calls obtained from the peanut community SNP chip. Eight minicore accessions (across) are shown here, and 14 SNPs (dnwn) are displayed. Calls are given as AA or BB, or occasionally as the heterozygote AB.

Affy_SNP_ID	Chr_id	Start	ConversionType	a550846-43969	a550846-43969	a550846-43969	a550846-43969	a550846-43969	a550846-43969	a550846-43969	a550846-43969
				45_P1429420	46_P1355271	47_P1407667	48_P1240560	49_P1339960	50_P1502040	51_P1493693	52_P1331314
Affx-15207 Aradu.A01	230496	PolyHighResolution	BB	AA	BB	BB	BB	BB	BB	BB	BB
Affx-15206 Aradu.A01	231463	PolyHighResolution	AA	BB	AA	AA	AA	AA	AA	AA	AA
Affx-15207 Aradu.A01	413869	PolyHighResolution	BB	AA	BB	BB	BB	BB	BB	BB	BB
Affx-15203 Aradu.A01	1193638	PolyHighResolution	BB	BB	BB	BB	BB	BB	BB	BB	BB
Affx-15208 Aradu.A01	1398141	PolyHighResolution	AA	BB	AA	AA	AA	AA	AA	AA	AA
Affx-15205 Aradu.A01	1455124	PolyHighResolution	AA	BB	AA	AA	AA	AA	AA	AA	AA
Affx-15205 Aradu.A01	3808034	PolyHighResolution	BB	BB	BB	BB	BB	BB	BB	BB	BB
Affx-15206 Aradu.A01	3910739	PolyHighResolution	BB	AA	BB	BB	BB	AA	BB	BB	BB
Affx-15207 Aradu.A01	4679934	PolyHighResolution	BB	AA	BB	BB	BB	BB	BB	BB	BB
Affx-15204 Aradu.A01	6315233	PolyHighResolution	AA	AA	AA	AA	AA	AA	AA	AA	AA
Affx-15207 Aradu.A01	7180725	PolyHighResolution	BB	AA	BB	BB	BB	BB	BB	BB	BB
Affx-15206 Aradu.A01	9205209	PolyHighResolution	BB	AB	BB	BB	AB	BB	BB	BB	BB
Affx-15202 Aradu.A01	11382044	PolyHighResolution	BB	AB	BB	AB	AB	AA	AB	AB	AB
Affx-15206 Aradu.A01	18338848	PolyHighResolution	BB	BB	BB	BB	BB	BB	AA	BB	BB

Leafspot Resistant Spanish-types

We increased a population BC3-43-09-03-02 X Schubert to introduce leafspot resistance into Spanish peanuts. These materials were segregating for market type, with most of the materials being runners (because runner type is dominant to Spanish type). We harvested the different plant types separately and have planted Spanish selections where possible for increase and DNA extraction for scoring for markers for leafspot resistance in 2020. Data on the markers that we identified (PM3, PMc348) were presented in the 2018 report.

Nematode Resistant Spanish-types

We have made crosses with breeding lines Tx144423 and Tx144432. Both lines are high oleic and nematode resistant lines from a spanish MDR program. The seed size of the lines is below average and further crossing is needed. In our 2018 spring crossing program we made several crosses with released varieties OLin, Tamnut OL06 and Schubert as well as selected other germplasm. These were increased in our greenhouses in summer 2018. In the 2019 season

they were planted as individual plants in Yoakum Tx. Selections were made in the field and material was moved forward for increase and plant row selection on 2020.

Sub-Project IV. SNP Marker Development

Development of a B Genome Mapping Population

This effort is being conducted with other funding but will be important to our efforts in the variety development program when it is completed. We are in the fourth generation of developing a B-genome mapping population involving two diploid (20 chromosome) wild species. The fourth generation (F_3) is actually the third generation of single seed descent which we are conducting. We will carry this project one more generations and then establish the map which will be integrated with the A-genome map and then be used for "prescription breeding."

We have cut ends off the seeds of each generation, and the remainder of each seed has been planted for increase. The cut seed ends will be used for DNA extraction. It is becoming increasingly difficult to maintain the population. We believe this is due to genetic incompatibility between species. We will continue to carry the material forward unless the population size becomes too small to collect accurate information. At that time the project will be reevaluated. An alternate population is available.

Screening for Root Knot Nematode Resistance

A total of 904 seeds representing 94 breeding lines were screened previously by SNP markers to identify lines with resistance to root-knot nematodes, drought, and having high oleic seed. This was done to purify the breeding lines, to verify that they had nematode resistance, and to purify them for markers for drought tolerance, as they were segregating for markers for drought tolerance. To obtain sufficient seed, these were harvested in 2019 and planted again for increase in 2020.

A related population, called Population 1B, was previously screened for markers with resistance to root-knot nematodes, drought, and having high oleic seed. Forty selections were made from the 180 breeding lines. These were planted and evaluated for drought tolerance (see the Additional Drought Tolerance section above).

Screening for the High O/L Trait

As part of our continuing efforts to develop high-oleic varieties and maintain purity of breeding lines and TAMU varieties, 6,327 seeds were tested for the high oleic trait by NIR (Table 8). Additional seeds were scored using DNA markers instead, mainly where we were also testing for other traits (such as root-knot nematode resistance or tolerance to water deficit) with markers at the same time.

Table 8. Numbers of Seeds screened by NIR for the High Oleic Trait.

Experiment	Seeds Screened
Pod Rot Resistant Runner	389
Large Seeded Runner	40
Valencia High Oleic Increase	674
AG18 Runner	653
Tamrun OL18L and Tamrun OL19	230
Other Runner Increase	127
Virginia High Oleic Increase	947
Spanish Drought Tolerant	90
Others including F1 and F2 Single Plants	132
Check Variety Testing	45
Stephenville Samples	2,821
Total	6,327

Closing Comments

The future of the the Texas A&M Peanut Breeding Program is very bright. New personnel and funding have allowed for several new projects to begin or reach preliminary approval. Projects in high throughput phenotyping using UAS and handheld Raman spectroscopy are underway, as well as development of new populations for drought, yield, leafspot and organic production. In addition, we are also starting new projects that specifically deal with nutrition and health of the peanut. The Texas A&M Peanut Breeding Program received significant new support from Texas A&M AgriLife Research in 2019. Dr. John Cason was hired as a full-time peanut breeder at the Texas A&M AgriLife Research and Extension Center at Stephenville. His hiring in this position restored a full-time position that was lost when Dr. Charles Simpson's retired 16 years ago. Texas A&M AgriLife provided a generous startup up package of almost \$450,000. These funds are were used for equipment purchases and greenhouse renovations at the Stephenville center that serve the entire peanut breeding program. In addition, the Texas Peanut Producers Board in conjunction with the Southwestern Peanut Shellers Association provided an additional \$100,000 gift for additional equipment purchases for the breeding program with were used to purchase additional equipment to help us serve the peanut producers of Texas. Finally, Texas A&M AgriLife Research also provided \$1,000,000 for the construction of a dedicated peanut sheller for the Texas A&M University System that will allow the breeding program to provide high quality pure seed to the peanut industry. As all these pieces are put in place as well as the expansion of the Texas A&M peanut program personnel has taken place we have already seen an increase in research proposals dealing with peanut and that will eventually bring in even more research dollars that will directly impact the producers of Texas and even the world.