Marker Assisted Selection for Nematode Resistance - Progress

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Initial Crossing in 2007

- **Dr. Bell develops BC1F1 (College Stn)**
  - **Lonren lines x MAFES Lines**
- **Thaxton-Wallace Lonren Crosses**
  - **DREC-MSU field & greenhouse**
- **Initially efforts directed at Reniform alone**

2007 – Misc. Breeding Lines x Lonren

- 8824-1-2-25-192-10-57 RKN
- STV 732N(h/ne)/8806-2-2-21
- STV 7A/Tamcot Sphinx
- STV 213/SPNXCDUG8H-1-95
- DPL 90/LBCHGP12KS-1-96
- DPL 90/DG2387
- DG 2387/STV 474
- ACSI1017/DP 5415 NE
- FM 832/DES 24NE
- 96WD-18/F5(89E-51/93WA126ne)
- SG747/DES 24NE
- DES227/H01-09-03
- 0110-1ne
- 0141-15ne
- 0023-11ne

- Lonren 63-11
- Lon O-15
- Lon A-2
- Lon 84-4
- Lon 83-1
- Lon 83-31
- Lon 64-1
- Lon M-3
- Lon 83-8
- Lon 84-4

2008 – 1st Year of MAS

- Planted 27 F2 and 95 BC1F2 populations with 40 parents
- Tagged 3566 plants
- BNL3279 Results†
  - 1865 null
  - 980 heterozygous
  - 850 homozygous

†USDA-ARS Genomics Lab – Stoneville, MS
Brian Scheffler
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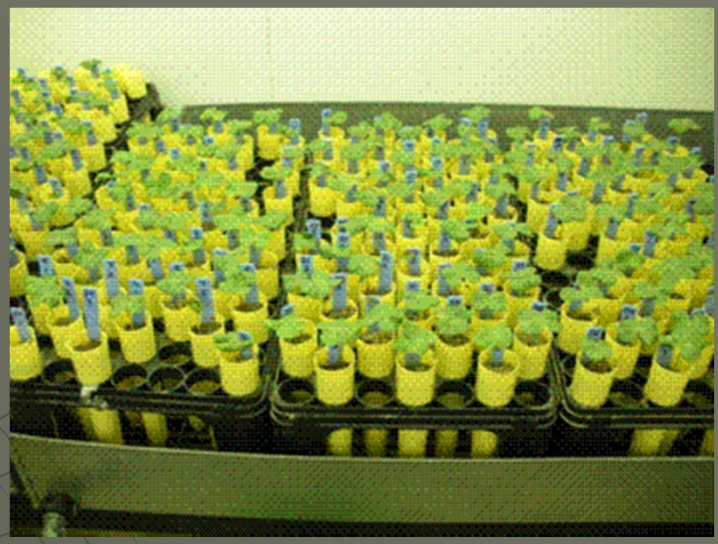
2008 – MAS Plant Selections

- ~300 F2 IP's
- Criteria
  - BNL3279
  - lack of stunting
  - # bolls
  - general appearance
- Lint%
- Fiber Quality

2009

- F2:F3–Confirm phenotype
  - random sample
  - winter greenhouse – (Stetina)
- MAS F2:F3 planted to progeny row
- Testing
  - Homogeneity
    - Segregants discarded
  - Lint% - Fiber Quality
- 68 rows (F3) selected for further evaluation



F3 Pro-Rows

2010 F4 Yield

Table. Least square means for lint yield and fiber for Lonren dervied F4 yield trial- Stoneville, MS,2010.

2010 ENTRY	2009 PLANT TAG #	Pedigree	Lint Yield † lbs/a	Lint Percent %	MIC mic	UHM %	STRN g/tex	QS1 ‡	QS2 ‡
57	289-229	161)FM 832/DES 24 NE//(804)L	1060	35.56	5.40	1.11	42.60	58.00	79.50
72	FM 958	Commercial Check	1057	37.47	5.20	1.19	34.70	79.00	76.00
71	SG 105	Commercial Check	1018	35.92	5.20	1.15	31.60	70.00	72.50
70	DP 393	Commercial Check	998	38.16	5.00	1.17	33.30	79.00	76.50
64	392-239	161)FM 832/DES 24 NE//(804)L	986	35.98	5.10	1.11	40.05	62.50	82.50
63	376-238	161)FM 832/DES 24 NE//(804)L	952	35.72	4.90	1.14	35.40	72.00	76.00
62	369-237	161)FM 832/DES 24 NE//(804)L	951	35.54	5.55	1.15	38.85	65.50	77.50
14	239, 124	-2-25-192-10-56 RKN/84-38 Lo	900	39.79	5.20	1.11	38.50	59.00	76.00
56	281-228	161)FM 832/DES 24 NE//(804)L	875	36.05	4.90	1.12	36.75	66.50	71.00
60	345-235	161)FM 832/DES 24 NE//(804)L	872	35.61	5.15	1.14	36.70	67.50	74.50
1	532-551	-2-25-192-10-10 RKN/83-30 Lo	855	34.20	4.40	1.18	33.05	88.50	78.00
31	712-171	0029-5/LONREN-1	851	38.08	4.80	1.09	35.00	59.00	68.00
43	997,199	23)ACSI 1017/DP 5415 NE//(804	827	37.80	4.60	1.12	37.40	70.00	76.00
17	312-133	-2-25-192-10-56 RKN/84-38 Lo	820	39.93	4.95	1.09	33.10	53.50	59.50
69	LONREN-	Reniform Parent	699	36.55	4.70	1.13	35.60	71.50	71.50
41	969-197	23)ACSI 1017/DP 5415 NE//(804	671	35.65	4.70	1.09	39.25	61.00	74.00
16	272-129	-2-25-192-10-56 RKN/84-38 Lo	661	37.02	4.55	1.09	33.05	60.50	64.50
53	193-220	23)ACSI 1017/DP 5415 NE//(804	659	37.53	5.20	1.05	35.00	37.50	55.50
44	025-203	23)ACSI 1017/DP 5415 NE//(804	450	33.92	4.05	1.13	35.20	76.50	78.00
15	252-127	-2-25-192-10-56 RKN/84-38 Lo	449	34.86	4.95	1.10	34.65	56.00	62.50
6	872-891	1-2-25-192-10-3 RKN/84-37 Lor	447	34.32	4.60	1.07	35.70	59.50	75.00
13	212-123	-2-25-192-10-56 RKN/84-38 Lo	443	38.08	5.35	1.03	35.80	32.00	61.00
23	512-151	0036-10/LONREN-1	429	33.12	5.50	1.05	36.85	33.00	57.50
65	416-242	D-18/F5(89E-51/93WA-126ne)/	406	34.33	4.40	1.14	37.00	78.50	79.00
Mean			764	36.30	4.93	1.11	36.05	63.17	71.75
LSD (.05)			287	2.31	0.41	0.06	2.18	19.70	
CV(%)			28.95	3.23	4.26	2.67	3.05	16.08	
R-Square			0.50	0.81	0.83	0.77	0.90	0.79	
Reps			4	2	2	2	2	2	2

2011 F5 Yield

Table 1. LSmeans for performance of 2011 Lonren F5 Yield A (Stoneville, MS).

Entry	ID	Pedigree	Lint Yield lbs/a	r	Lint Percent %	MIC mic	UHM %	STRN g/tex	QS1†	QS2†
3	0762-9	8824-1-2-2	1314	1	37.69	4.97	1.21	30.85	60.25	67.25
20	0774-6	0110-1ne/L	1175	2	32.88	4.74	1.23	37.08	69.25	81.75
26	(06-22 ne)	(06-22 ne)9	1082	3	35.61	5.17	1.17	33.48	46.50	55.75
11	0763-4	8824-1-2-2	1081	4	36.92	4.96	1.16	29.78	43.75	56.50
4	DP 5415NI	DP 5415NE	719	19	31.83	4.81	1.24	32.95	70.50	77.25
16	Lonren-1/(	Lonren-1/0	719	20	34.89	4.20	1.30	32.40	92.00	91.50
19	0774-7	0110-1ne/L	708	21	31.89	4.21	1.29	31.13	89.50	90.50
35	**FM958**	**CK**	**687**	22	**37.29**	**4.82**	**1.24**	**34.05**	**74.25**	**78.25**
6	0750-5	0050-14 / L	681	23	35.75	4.70	1.18	30.30	50.00	63.00
31	DES 227/H	DES 227/H(	674	24	32.94	4.57	1.21	32.30	66.50	73.25
33	**DP393**	**CK**	**667**	25	**35.65**	**4.82**	**1.21**	**32.20**	**65.00**	**71.00**
18	Lonren-1/(	Lonren-1/0	664	26	32.12	4.15	1.30	33.68	97.75	93.75
7	0778-2	0141-15ne/	563	31	33.90	5.09	1.12	31.03	32.00	47.75
36	**DP 491**	**CK**	**538**	32	**37.13**	**4.46**	**1.30**	**32.35**	**91.50**	**90.50**
29	0723-5	0038-4 / LC	470	33	33.69	5.31	1.17	33.05	42.25	54.25
34	**SG105**	**CK**	**431**	34	**32.92**	**4.64**	**1.19**	**30.33**	**60.50**	**66.75**
25	(04PST-12	(04PST-123	418	35	33.09	4.81	1.15	36.45	49.50	63.25
24	0708-3	0029-5/LOI	391	36	33.38	4.22	1.22	34.55	64.50	72.25
Mean			721		34.42	4.70	1.21	32.66	64.75	71.92
LSD (.05)			230		1.18	0.28	0.04	1.84	12.55	9.12
CV(%)			18.77		2.40	4.22	2.13	4.00	14.47	9.39
R-Square			0.77		0.86	0.76	0.83	0.80	0.83	0.86
Reps			4		4	4	4	4	4	4

2012

- 2010-2011 Yield testing –marginal
- 2012
  - Multi-location testing of top picks
    - pending root-tip homogeneity
  - Infested & Non-Infested
  - Checks
    - Lonren / Barbren / Commercial

SUMMARY

- Three additional cycles of crossing and MAS since initial crosses in 2007
- New sources of resistance added each cycle
  - RKN, GB713 derived
- Learning experience each year
  - Mechanics –
    - tagging
    - #samples
    - testing homogeneity
    - Infested field yield trial

SUMMARY-continued

- Approach – very applied, screen max # plants possible for rare recombinant
- Define, refine protocol, improve efficiency
- Tools / Aides – new for 2012

Root Tip Testing

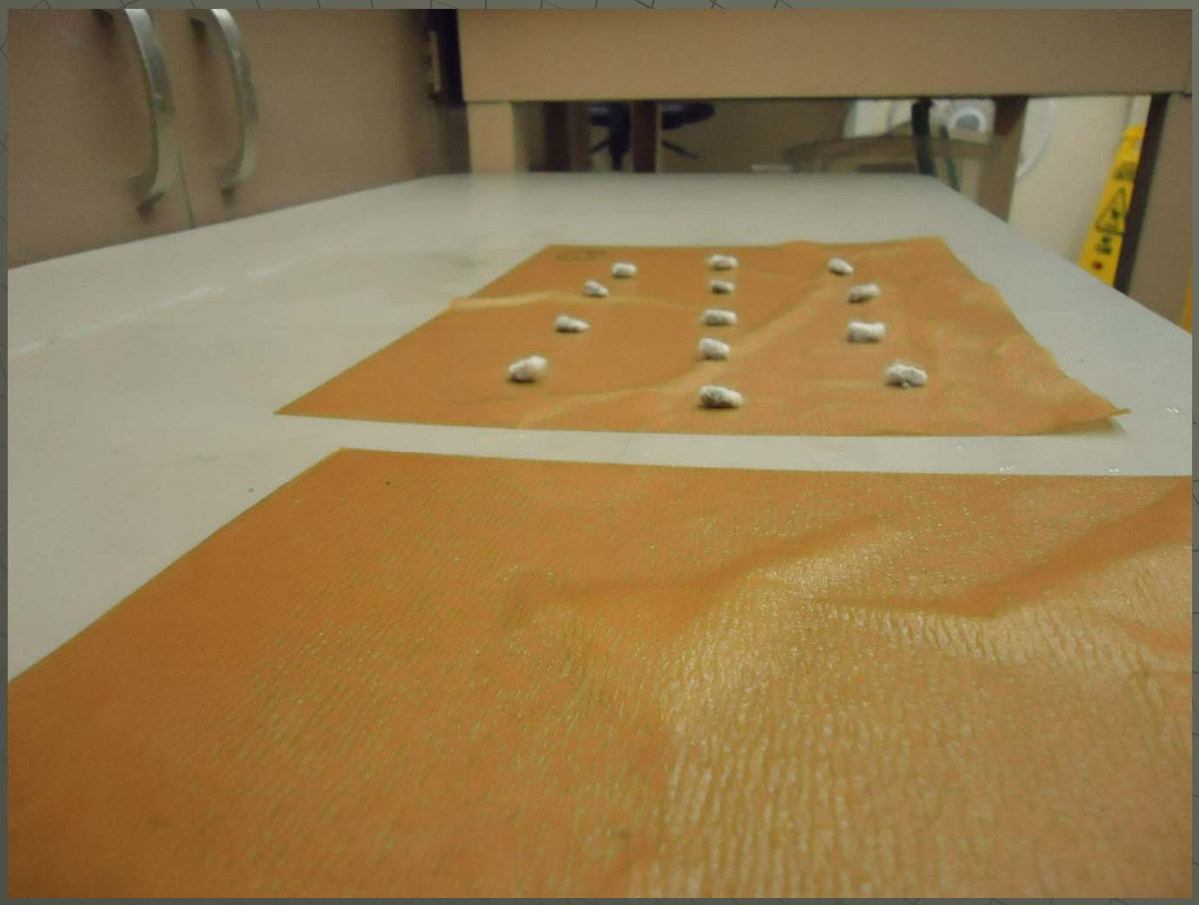
- **F3RKN IP**
  - **7 Homozygous RKN**
- **F2IP**
  - **84 Homo Ren**
  - **35 RKN**
  - **29 Het Ren, Homo RK**
- **F2F3IP**
  - **45 Homo Ren**
  - **52 Het RN, homo RKN**
  - **3 Het RN, het RKN**
- **F2F3 Progeny Rows**
  - **29 Homozygous**

Root-Tips
Confirm MAS in Field

Zygosity of marker assisted selected IP's and rows in 2011

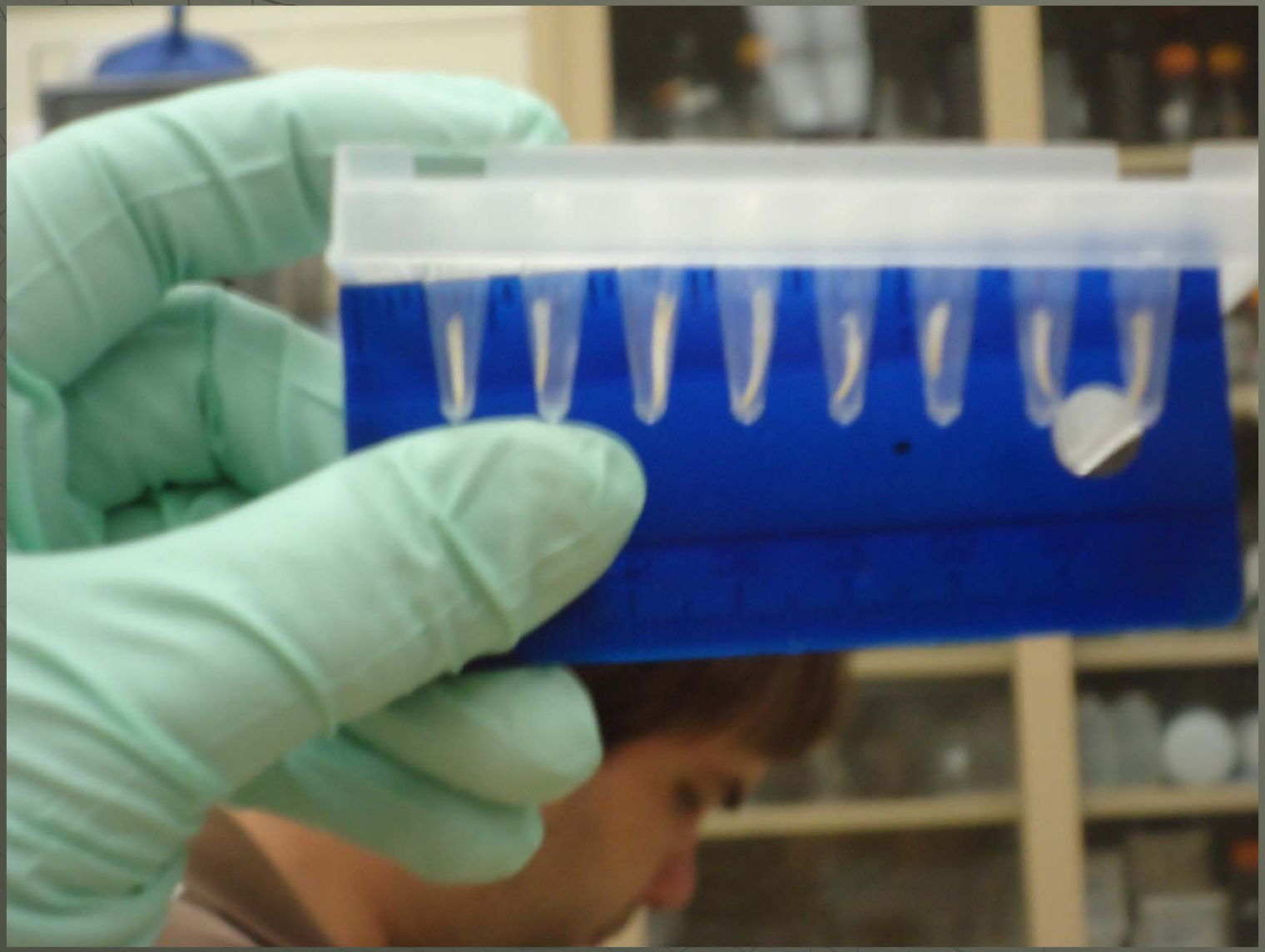
2011 Generation	Ren Hom	RKN Hom	Ren / RKN Hom / Hom	Ren / RKN Het / Hom	Ren / RKN Het / Het	TOTALS
F2 IP's	84	35		29		148
F2F3 IP's	41		4	51	4	100
F2F3 Rows	20	9				29
Total Selected	145	44	4	80	4	277

MAKE RAG DOLLS



Incubate at 85F for 48 hrs

4 seed/entry
.5 to 1 cm length



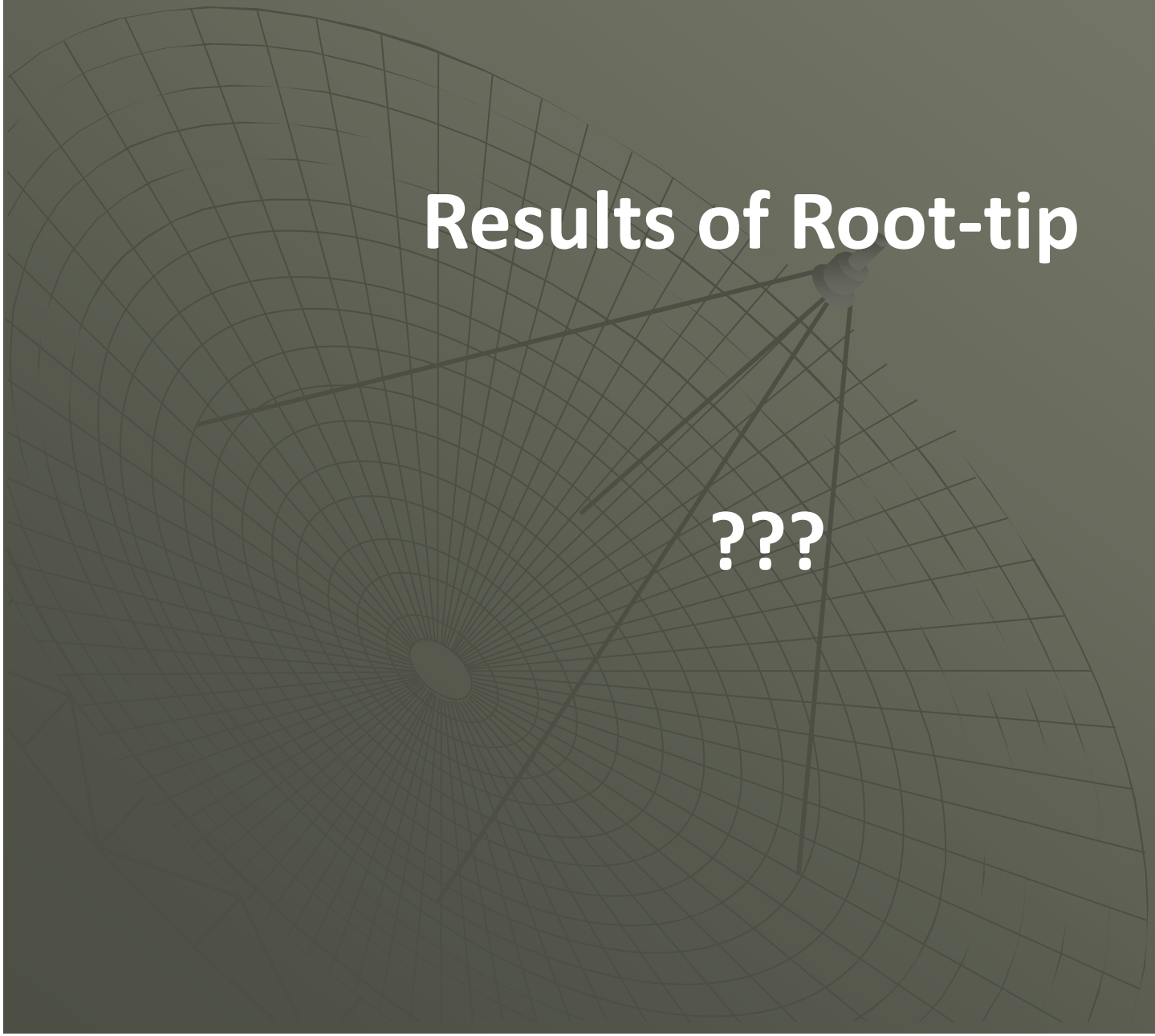
Add buffer A
Seal plate with film

Incubate plates in thermocycler for 10 min with heated lid

Add buffer B
Put film back on
Vortec briefly

Spin plates in
Centrifuge
4000 for 1 min

Results of Root-tip



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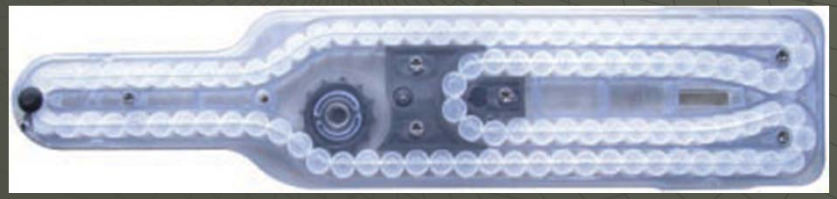
PLANT TRAK
Genetic Sampling and Barcoding



Planttrak Bench Top Unloader