

Optimization of Doubled Haploid Procedures in Burley Tobacco

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University of Kentucky

Background

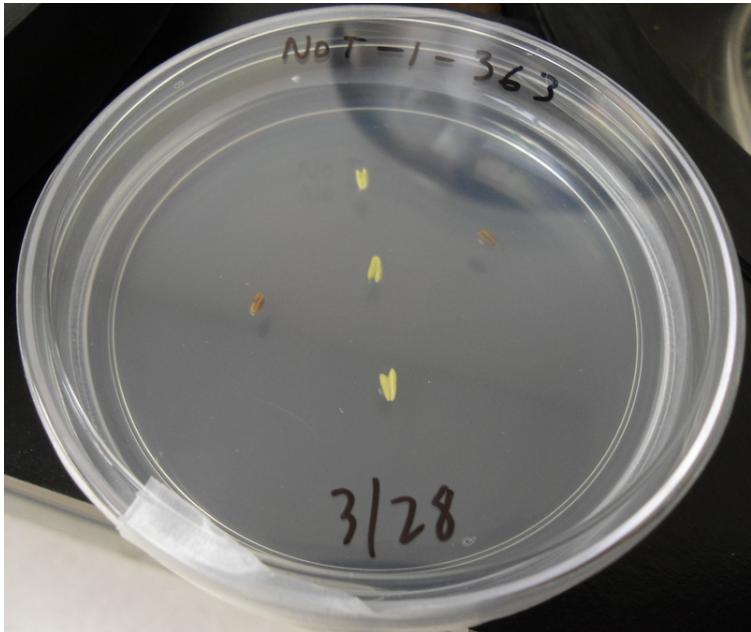
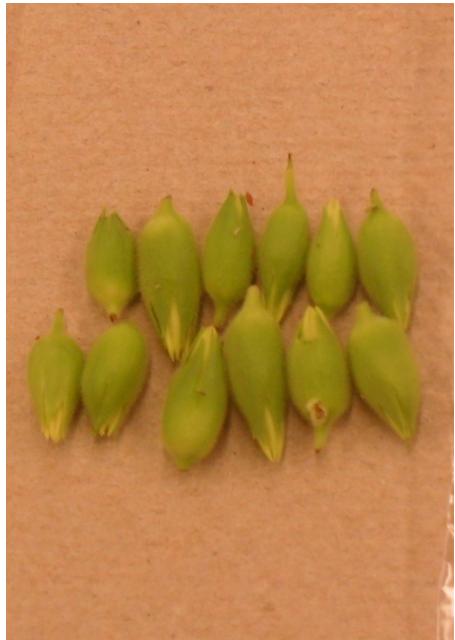
- Production of doubled haploid (DH) lines is often used to significantly reduce the amount of time required to achieve homozygosity in tobacco breeding populations.

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- Production of doubled haploid (DH) lines is often used to significantly reduce the amount of time required to achieve homozygosity in tobacco breeding populations.
- There are two methods of obtaining DH lines in tobacco.

WAYS TO GET HAPLOID PLANTS

→ **Androgenic Derived Haploid (ADH):** Culturing of the male gametes (**anther culture**)



Immature buds

Plate anthers on specialized media

WAYS TO GET HAPLOID PLANTS

→ **Androgenic Derived Haploid (ADH):** Culturing of the male gametes (**anther culture**)



Haploid plantlets generated



Plantlets placed on rooting media

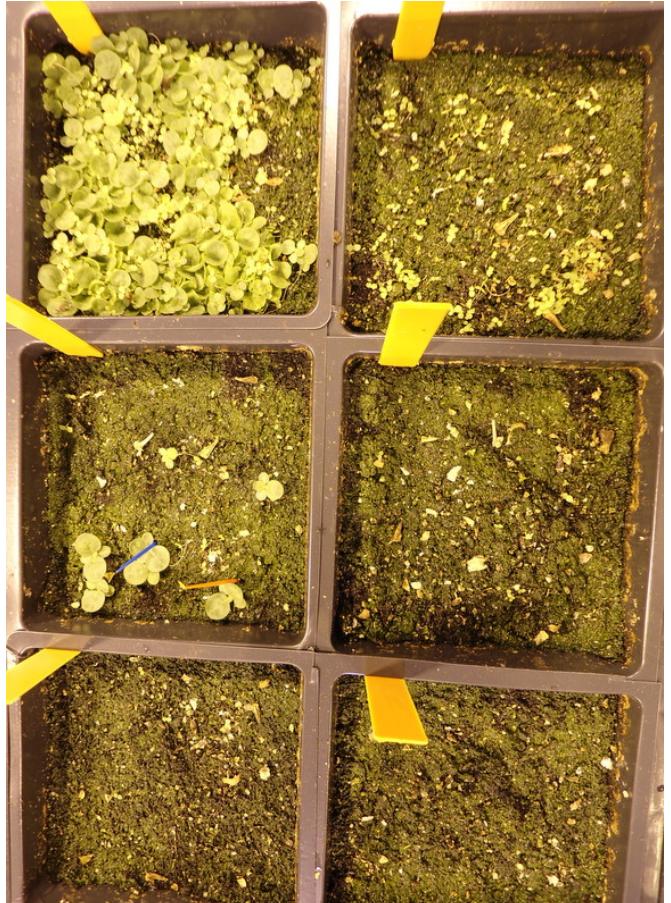
WAYS TO GET HAPLOID PLANTS

Maternally or Gynogenic derived haploids (MDH or GDH):

→ Interspecific crosses *N. tabacum* X *N. africana* – Chrom. elimination



N. tabacum X *N. africana* cross



Identification of haploid plantlets

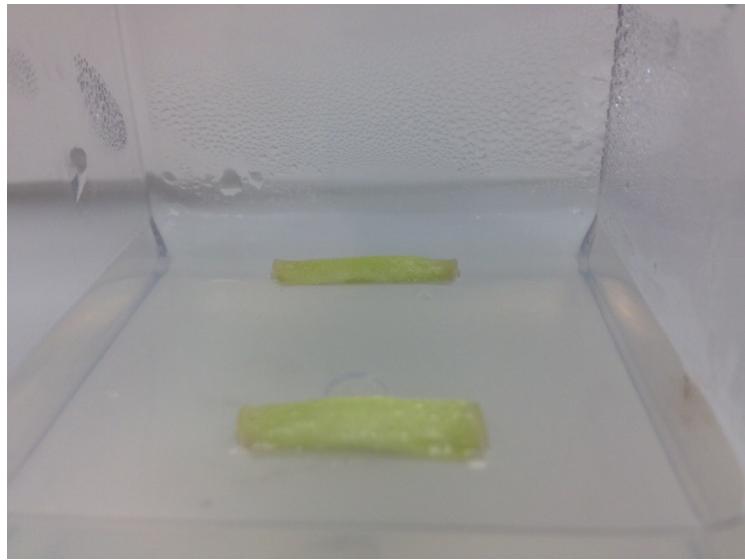
Chromosome Doubling of Haploid Plants

ADH:

or

→ Doubled Haploids

GDH:



Mid-Vein Culture



Shoot Initiation

Chromosome Doubling of Haploid Plants

ADH:

or

→ Doubled Haploids

GDH:



Plantlets on rooting media

Root initiation

Background

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Background

- Historically, the most prominent reported disadvantage with regard to using DH techniques in tobacco is their unsatisfactory agronomic performance.
- ADH lines have been shown to be inferior to the cultivars from which they were derived when compared for overall agronomic performance, particularly in flue-cured tobacco.
- Although GDH lines performed better than ADH lines, neither displayed equal yielding ability compared to the selfed progenies of the original parental lines.

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- Due to the inferiority of ADH lines in comparison to GDH lines, the GDH *N. africana* method was adopted by most tobacco DH breeding programs.
- Virtually all tobacco varieties are now released as male sterile hybrid varieties.
- Although GDH lines have performed very well as male parents in the production of hybrid varieties, little or no published research has investigated the performance of ADH parental lines for the development of hybrid varieties.

Objectives of Current Research

- Objective One – Determine the relative efficiency of androgenic versus gynogenic methods of obtaining doubled haploid lines in two burley tobacco varieties.

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- Objective Two – Compare the agronomic performance of ADH versus GDH lines developed from two burley varieties.
- Objective Three – Compare the performance of two hybrid varieties having ADH versus GDH lines as the male parent.

OBJECTIVE 1

What is the most effective method of generating DH materials – ADH or GDH?

→ Time: **ADH: 473 days** **GDH: 400 days**

ADH – Germination response - Source of anthers

Seed Nursery	24 %	TN 90LC
	14.3%	GR 149LC
Disease Nursery	16.9%	<i>Fusarium</i> Wilt
	11.6%	Bacterial Wilt
	9.1%	Black Shank

OBJECTIVE 1

What is the most effective method of generating DH materials – ADH or GDH?

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GDH – Differential response of Genotypes

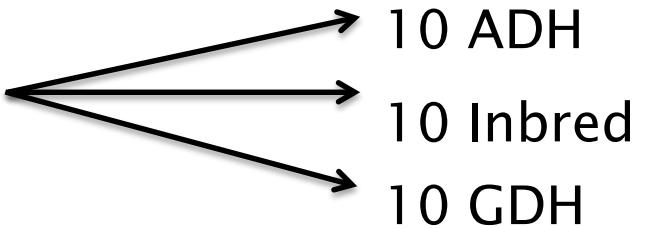
	crosses	haploids	Hap/cross
TN 90LC	70	10	1 / 7
GR 149LC	460	6	1 / 76

OBJECTIVE 2

Agronomic performance of Doubled Haploids

→ Genotype:

TN 90LC



109-RECGRN-05
TN 90 DiHaploid Study
Planting Plan - Example

Rep 1		Rep 2		Rep 3	
Plot	Entry	Plot	Entry	Plot	Entry
	Border		Border		Border
1201	TN 90ADH1	1301	TN 90ADH3	1401	TN 90ADH4
1202	TN 90LC	1302	TN 90LC	1402	TN 90LC
1203	TN 90MDH1	1303	TN 90MDH3	1403	TN 90MDH4
1204	TN 90ADH2	1304	TN 90ADH5	1404	TN 90ADH6
1205	TN 90LC	1305	TN 90LC	1405	TN 90LC
1206	TN 90MDH2	1306	TN 90MDH5	1406	TN 90MDH6
1207	TN 90ADH3	1307	TN 90ADH7	1407	TN 90ADH8
1208	TN 90LC	1308	TN 90LC	1408	TN 90LC
1209	TN 90MDH3	1309	TN 90MDH7	1409	TN 90MDH8
1210	TN 90ADH4	1310	TN 90ADH9	1410	TN 90ADH10
1211	TN 90LC	1311	TN 90LC	1411	TN 90LC
1212	TN 90MDH4	1312	TN 90MDH9	1412	TN 90MDH10

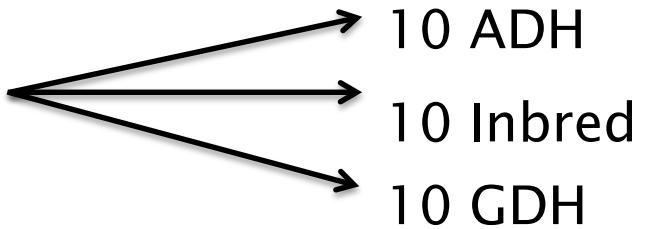


OBJECTIVE 2

Agronomic performance of Doubled Haploids

→ Genotype:

TN 90LC



→ Design: **Split Plot Design - 3 Locations**

3 Blocks (reps) per Location

30 plants per row

→ Data:

- > Plant height - 50 D.A.T.
- > Plant height after topping
- > Leaf length
- > Leaf width
- > Number leaves/ plant
- > Yield

Results

Mean Values for the Inbred Source, ADH and GDH Populations

TN 90LC at Three Locations - 2013

Note - Mean values are for nine triplets

DH Method	Height 50 (cm)	Height topping (cm)	Leaf length (cm)	Leaf width (cm)	Leaves per plant	Yield (Kg/ha)
TN 90LC	117.7	138.9	60.6	27.4	19.7	2889
ADH Lines	111.0	135.7	59.6	26.0	19.8	2778
F value	26.03***	5.49*	3.42	31.35***	0.33	5.28*
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TN 90LC	117.7	138.9	60.6	27.4	19.7	2889
GDH Lines	122.2	141.4	60.4	26.8	20.0	2945
F value	10.96**	4.86*	0.46	5.04*	3.19	1.84

(*) P<0.05, (**) P<0.01 and (***) P<0.0001



TN 90 ADH5

TN 90LC

TN 90 GDH5

H5

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**ANOVA for Differences among Individual Lines *within* Populations
*TN 90LC Populations***

Population	Statistic	Height 50 (cm)	Height topping (cm)	Leaf length (cm)	Leaf width (cm)	Leaves per plant	Yield (Kg/ha)
TN 90 ADH (10 Lines)	MS F value	616.87 2.27*	494.9 3.04**	54.26 6.79***	32.59 11.76***	1.75 1.05	1041164.4 12.24***
TN 90LC (10 Lines)	MS F value	267.91 0.6	147.63 0.84	12.82 1.23	6.03 1.55	0.877 0.77	387927.1 3.83**
TN 90 GDH (10 Lines)	MS F value	4107.3 11.96***	1230.9 3.91**	40.58 3.47**	15.58 3.12**	6.72 3.82**	2064789 16.42***

(*) P<0.05, (**) P<0.01 and (***) P<0.0001

TN 90 ADH Lines in Comparison to Their Paired TN 90LC

TN 90 Triplet	Doubled Haploid Method	Height 50 (cm)	Height topping (cm)	Leaf length (cm)	Leaf width (cm)	Leaves per plant	Yield (Kg/ha)
Line 4	TN 90LC	121.9 a	138.8 a	59.5 a	26.2 a	19.2 a	2629 a
	TN 90 ADH	122.1 a	143.4 a	59.2 a	25.4 a	19.5 a	2842 a +213
Line 1	TN 90LC	118.9 a	137.2 a	60.4 a	27.0 a	19.9 a	2778 a
	TN 90 ADH	113.9 a	136.6 a	59.4 a	25.6 a	20.0 a	2840 a +62
Line 2	TN 90LC	107.4 a	131.2 a	58.9 a	26.6 a	19.6 a	2756 a
	TN 90 ADH	100.9 a	128.0 a	56.8 a	24.3 b	20.4 a	2795 a +39
Line 8	TN 90LC	117.1 a	137.1 a	60.2 a	27.2 a	19.3 a	2929 a
	TN 90 ADH	107.0 b	135.2 a	60.3 a	27.1 a	19.7 a	2892 a -37
Line 7	TN 90LC	125.3 a	147.0 a	61.8 a	28.6 a	20.1 a	3047 a
	TN 90 ADH	119.0 a	140.6 a	62.3 a	27.9 a	19.4 a	2883 a -164

Separation of means within comparisons are based on Fischer's LSD; means followed by the same letter within a column and within a TN 90LC are not significantly different at 5% level of significance.

TN 90 ADH Lines in Comparison to Their Paired TN 90LC (Cont.)

TN 90 Triplet	Doubled Haploid Method	Height 50 (cm)	Height topping (cm)	Leaf length (cm)	Leaf width (cm)	Leaves per plant	Yield (Kg/ha)
Line 10	TN 90LC	117.3 a	141.5 a	61.0 a	27.9 a	19.9 a	2883 a
	TN 90ADH	110.5 a	135.0 a	59.0 b	25.5 b	19.6 a	2644 a
Line 6	TN 90LC	117.3 a	138.1 a	59.9 a	26.8 a	19.7 a	2951 a
	TN 90ADH	102.8 b	129.8 b	60.7 a	24.6 b	20.0 a	2683 a
Line 3	TN 90LC	122.9 a	143.5 a	62.2 a	27.6 a	20.2 a	3013 a
	TN 90ADH	114.6 a	141.1 a	59.7 b	27.5 a	20.3 a	2748 b
Line 9	TN 90LC	111.5 a	136.1 a	61.9 a	28.1 a	19.7 a	3016 a
	TN 90ADH	108.4 a	131.9 a	59.4 a	25.9 b	19.6 a	2673 a
Line 5	TN 90LC	113.1 a	135.0 a	62.5 a	29.0 a	20.2 a	3271 a
	TN 90ADH	95.5 b	113.9 b	52.3 b	20.5 b	18.7 a	1900 b

-239

-268

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TN 90 **GDH** Lines in Comparison to Their Paired TN 90LC

TN 90 Triplet	Doubled Haploid Method	Height 50 (cm)	Height topping (cm)	Leaf length (cm)	Leaf width (cm)	Leaves per plant	Yield (Kg/ha)
Line 4	TN 90LC	121.9 a	138.8 a	59.5 a	26.2 a	19.2 a	2629 b
	TN 90 GDH	129.6 a	144.0 a	59.2 a	26.9 a	19.9 a	3024 a +395
Line 10	TN 90LC	117.3 a	141.5 a	61.0 a	27.9 a	19.9 a	2883 a
	TN 90 GDH	111.5 a	135.7 a	62.0 a	27.3 a	19.0 a	3148 a +265
Line 2	TN 90LC	107.4 a	131.2 a	58.9 a	26.6 a	19.6 a	2756 a
	TN 90 GDH	117.8 a	135.7 a	58.8 a	26.1 a	20.6 a	2841 a +85
Line 8	TN 90LC	117.1 b	137.1 b	60.2 a	27.2 a	19.3 a	2929 a
	TN 90 GDH	126.6 a	147.6 a	58.9 a	27.6 a	19.7 a	2940 a +11
Line 9	TN 90LC	111.5 a	136.1 a	61.9 a	28.1 a	19.7 a	3016 a
	TN 90 GDH	111.6 a	135.9 a	62.1 a	26.8 a	20.2 a	3025 a +9

Separation of means within comparisons are based on Fischer's LSD; means followed by the same letter within a column and within a TN 90LC are not significantly different at 5% level of significance.

TN 90 **GDH** Lines in Comparison to Their Paired TN 90LC (Cont.)

TN 90 Triplet	Doubled Haploid Method	Height 50 (cm)	Height topping (cm)	Leaf length (cm)	Leaf width (cm)	Leaves per plant	Yield (Kg/ha)
Line 3	TN 90LC	122.9 a	143.5 a	62.2 a	27.6 a	20.2 a	3013 a
	TN 90 GDH	127.6 a	144.0 a	62.0 a	26.1 b	20.2 a	3005 a
Line 1	TN 90LC	118.9 a	137.2 a	60.4 a	27.0 a	19.9 a	2778 a
	TN 90 GDH	121.8 a	142.3 a	58.6 a	25.7 a	20.4 a	2765 a
Line 7	TN 90LC	125.3 a	147.0 a	61.8 a	28.6 a	20.1 a	3047 a
	TN 90 GDH	125.8 a	146.3 a	62.0 a	28.5 a	20.2 a	2964 a
Line 6	TN 90LC	117.3 a	138.1 a	59.9 a	26.8 a	19.7 a	2951 a
	TN 90 GDH	127.1 a	141.3 a	59.8 a	26.0 a	19.5 a	2794 a
Line 5	TN 90LC	113.1 a	135.0 a	62.5 a	29.0 a	20.2 a	3271 a
	TN 90 GDH	57.8 b	102.0 b	54.7 b	23.1 b	17.3 b	1687 b

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Separation of means within comparisons are based on Fischer's LSD; means followed by the same letter within a column and within a TN 90LC are not significantly different at 5% level of significance.

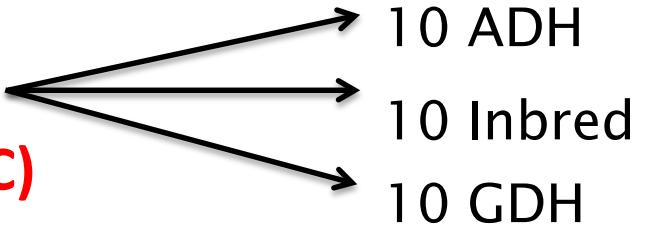
OBJECTIVE 3

Agronomic performance of Hybrid Varieties

→ Genotype:

KT 204LC

KT 204LC = (TKS 2002LC X TN 90LC)



→ Design: **Split Plot Design - 3 Locations**

3 Blocks (reps) per Location

30 plants per row

→ Data:

- > Plant height - 50 D.A.T.
- > Plant height after topping
- > Leaf length
- > Leaf width
- > Number leaves/ plant
- > Yield

Results

**Mean Values for Hybrid Varieties Having the
Inbred Source, ADH, or GDH Lines as Male Parent**
KT 204LC at Three Locations - 2014

Source of Male Parent	Height 50 (cm)	Height topping (cm)	Leaf length (cm)	Leaf width (cm)	Leaves per plant	Yield (Kg/ha)
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TN 90LC	82.8	130.3	63.1	28.5	20.5	3211
ADH Lines	80.8	128.8	64.0	28.4	20.3	3195
<i>F value</i>	4.99*	3.97	4.18*	0.09	3.57	0.61
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TN 90LC	82.8	130.3	63.1	28.5	20.5	3211
GDH Lines	82.0	129.1	63.7	28.8	20.4	3342
<i>F value</i>	0.96	2.64	1.75	1.14	1.32	3.56

(*) P<0.05, (**) P<0.01 and (***) P<0.0001

ANOVA for Differences among Individual Lines **within Populations**
KT 204LC Populations

Source of Male Parent	Statistic	Height 50 (cm)	Height topping (cm)	Leaf length (cm)	Leaf width (cm)	Leaves per plant	Yield (Kg/ha)
TN 90 ADH (10 Lines)	MS	58.55	99.57	21.41	3.58	2.23	147819.8
	F value	0.72	1.27	2.72*	1.00	1.91	1.88
TN 90LC (10 Lines)	MS	59.07	88.55	7.96	1.9	0.81	74214.4
	F value	2.31*	2.78**	1.01	0.58	0.81	0.96
TN 90 GDH (10 Lines)	MS	112.17	144.12	13.27	4.09	0.88	137434.2
	F value	1.47	2.57*	1.72	1.33	1.11	1.14

(*) P<0.05, (**) P<0.01 and (***) P<0.0001

KT 204 ADH Hybrid lines in Comparison to KT 204LC

KT 204LC Triplet	Male Parent	Height 50 (cm)	Height topping (cm)	Leaf length (cm)	Leaf width (cm)	Leaf number	Yield Kg/ha
Line 1	KT 204LC	84.0 a	135.1 a	63.4 a	28.8 a	21.0 a	3159 a
	KT 204 ADH	76.2 b	127.9 b	64.0 a	28.8 a	19.9 b	3317 a
Line 6	KT 204LC	83.2 a	132.2 a	64.4 a	29.1 a	20.7 a	3331 a
	KT 204 ADH	78.6 a	126.3 a	65.1 a	28.1 a	20.1 a	3437 a
Line 2	KT 204LC	83.1 a	134.8 a	63.0 a	28.7 a	20.8 a	3276 a
	KT 204 ADH	81.5 a	130.6 a	64.2 a	28.8 a	20.5 a	3350 a
Line 5	KT 204LC	79.7 a	129.6 a	62.0 a	28.2 a	20.7 a	3186 a
	KT 204 ADH	84.3 a	133.2 a	60.4 a	27.0 a	21.4 a	3256 a
Line 10	KT 204LC	78.1 a	126.6 a	64.2 a	28.9 a	20.2 a	3052 a
	KT 204 ADH	80.9 a	129.0 a	64.1 a	28.8 a	20.4 a	3108 a

Separation of means within comparisons are based on Fischer's LSD; means followed by the same letter within a column and within a TN 90LC are not significantly different at 5% level of significance.

KT 204 ADH Hybrids in Comparison to KT 204LC (Cont.)

KT 204LC Triplet	Male Parent	Height 50 (cm)	Height topping (cm)	Leaf length (cm)	Leaf width (cm)	Leaf number	Yield Kg/ha
Line 8	KT 204LC	84.4 a	126.8 a	62.7 a	28.1 a	20.1 a	3152 a
	KT 204ADH	81.5 a	128.2 a	63.1 a	27.6 a	20.1 a	3135 a
Line 4	KT 204LC	87.0 a	132.4 a	63.5 a	28.3 a	20.4 a	3119 a
	KT 204ADH	80.8 a	132.1 a	64.1 a	28.5 a	20.4 a	3101 a
Line 7	KT 204LC	81.2 a	127.5 a	63.0 b	28.7 a	20.4 a	3237 a
	KT 204ADH	78.5 a	122.5 a	66.4 a	29.1 a	19.7 a	3145 a
Line 9	KT 204LC	83.1 a	128.9 a	63.9 a	28.3 a	20.0 a	3223 a
	KT 204ADH	84.5 a	125.7 a	64.4 a	28.5 a	19.4 a	3022 a
Line 3	KT 204LC	84.5 a	129.0 a	61.3 b	27.8 a	20.4 a	3377 a
	KT 204ADH	81.0 a	131.7 a	64.0 a	28.6 a	20.7 a	3094 a

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Separation of means within comparisons are based on Fischer's LSD; means followed by the same letter within a column and within a TN 90LC are not significantly different at 5% level of significance.

KT 204 **GDH** Hybrid lines in Comparison to KT 204LC

KT 204LC Triplet	Male Parent	Height 50 (cm)	Height topping (cm)	Leaf length (cm)	Leaf width (cm)	Leaf number	Yield Kg/ha
Line 1	KT 204LC	84.0 a	135.1 a	63.4 a	28.8 a	21.0 a	3159 a
	KT 204GDH	86.1 a	130.5 a	63.8 a	29.3 a	20.2 b	3489 a
Line 4	KT 204LC	87.0 a	132.4 a	63.5 a	28.3 a	20.4 a	3119 b
	KT 204GDH	85.6 a	129.6 a	63.8 a	28.5 a	20.4 a	3422 a
Line 10	KT 204LC	78.1 a	126.6 a	64.2 a	28.9 a	20.2 a	3052 a
	KT 204GDH	78.6 a	131.9 a	66.1 a	29.8 a	20.5 a	3344 a
Line 8	KT 204LC	84.4 a	126.8 a	62.7 a	28.1 a	20.1 a	3152 a
	KT 204GDH	82.6 a	129.8 a	62.3 a	28.3 a	20.7 a	3389 a
Line 5	KT 204LC	79.7 a	129.6 a	62.0 b	28.2 a	20.7 a	3186 a
	KT 204GDH	74.0 b	119.8 a	64.7 a	27.9 a	19.8 a	3390 a

Separation of means within comparisons are based on Fischer's LSD; means followed by the same letter within a column and within a TN 90LC are not significantly different at 5% level of significance.

KT 204 **GDH** Hybrid lines in Comparison to KT 204LC (*Cont.*)

KT 204LC Triplet	Method	Height 50 (cm)	Height topping (cm)	Leaf length (cm)	Leaf width (cm)	Leaf number	Yield Kg/ha
Line 9	KT 204LC	83.1 a	128.9 a	63.9 a	28.3 a	20.0 a	3223 a
	KT 204GDH	82 a	127.6 a	64.1 a	29.6 a	20.3 a	3431 a
Line 2	KT 204LC	83.1 a	134.8 a	63.0 a	28.7 a	20.8 a	3276 a
	KT 204GDH	83.4 a	133.4 a	64.0 a	29.3 a	20.9 a	3396 a
Line 3	KT 204LC	84.5 a	129.0 a	61.3 a	27.8 b	20.4 a	3377 a
	KT 204GDH	84.0 a	133.3 a	63.0 a	29.0 a	20.9 a	3318 a
Line 6	KT 204LC	83.2 a	132.2 a	64.4 a	29.1 a	20.7 a	3331 a
	KT 204GDH	82.3 a	127.5 a	62.8 a	27.8 a	20.3 a	3175 a
Line 7	KT 204LC	81.2 a	127.5 a	63.0 a	28.7 a	20.4 a	3237 a
	KT 204GDH	81.5 a	127.4 a	62.3 a	28.3 a	20.0 a	3078 a

+208

+120

-59

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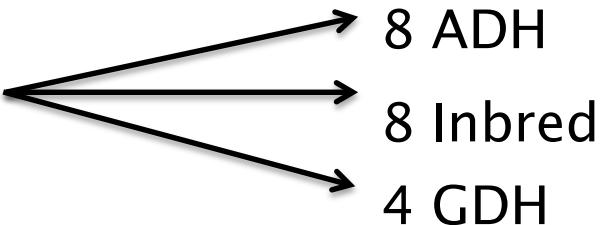
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OBJECTIVE 2

Agronomic performance of Doubled Haploids

→ Genotype:

GR149LC



→ Design: **Split Plot Design - 3 Locations**

3 Blocks (reps) per Location

30 plants per row

→ Data:

- > Plant height - 50 D.A.T.
- > Plant height after topping
- > Leaf length
- > Leaf width
- > Number leaves/ plant
- > Yield

Mean Values for the Inbred Source, ADH and GDH Populations

GR 149LC at Three Locations - 2014

DH Method	Height 50 (cm)	Height topping (cm)	Leaf length (cm)	Leaf width (cm)	Leaves per plant	Yield (Kg/ha)
GR 149LC	76.6	131.2	59.3	27.1	21.2	3351
ADH (8 Lines)	72.6	128.8	59.1	28.4	20.2	3357
F value	14.89**	5.41*	0.32	28.34***	36.7***	0.02
GR 149LC	77.7	131.3	59.2	26.8	21.4	3360
GDH (4 Lines)	73.3	131.6	58.6	28.4	20.2	3343
F value	17.05**	0.05	1.38	22.7***	24.35***	0.1

(*) P<0.05, (**) P<0.01 and (***) P<0.0001

ANOVA for Differences among Individual Lines **within Populations**
GR 149 Populations

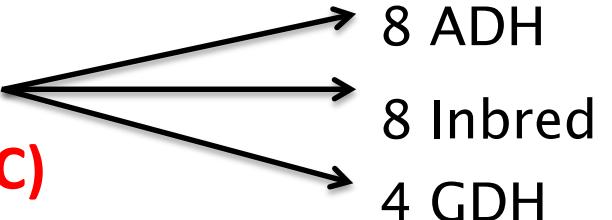
Population	Statistic	Height 50 (cm)	Height topping (cm)	Leaf length (cm)	Leaf width (cm)	Leaves per plant	Yield (Kg/ha)
GR 149 ADH (8 Lines)	MS F value	62.82 1.28	83 1.16	47.47 7.66***	98.43 32.91***	5.67 3.32**	112415.7 1.54
GR 149LC (8 Lines)	MS F value	35.45 0.73	19.13 0.46	6.58 1.09	3.82 1.66	1.73 1.72	66004.9 0.79
GR 149 GDH (4 Lines)	MS F value	163.06 6.14**	73.86 1.4	41.06 5.04**	251.66 76.44***	6.37 4.21**	335614.4 4.98**

(*) P<0.05, (**) P<0.01 and (***) P<0.0001

OBJECTIVE 3

Agronomic performance of Hybrid Varieties

→ Genotype: **TN 97LC**
TN 97LC = (ms TN 90LC X GR 149LC)



The diagram consists of three black arrows pointing to the right from the text "TN 97LC = (ms TN 90LC X GR 149LC)". The first arrow is labeled "8 ADH", the second "8 Inbred", and the third "4 GDH".

→ Design: **Split Plot Design - 3 Locations**
3 Blocks (reps) per Location
30 plants per row

→ Data:

- > Plant height - 50 D.A.T.
- > Plant height after topping
- > Leaf length
- > Leaf width
- > Number leaves/ plant
- > Yield

**Mean Values for Hybrid Varieties Having the
Inbred Source, ADH, or GDH Lines as Male Parent**

TN 97LC at Three Locations - 2014

Source of Male Parent	Height 50 (cm)	Height topping (cm)	Leaf length (cm)	Leaf width (cm)	Leaves per plant	Yield (Kg/ha)
GR 149LC	83.1	124.1	59.9	27.1	20.1	2968
ADH Lines	79.0	120.6	61.1	28.6	19.6	3033
<i>F value</i>	21.91***	11.02**	8.93**	31.86***	5.75*	2.88
GR 149LC	83.2	125.3	59.8	27.2	20.3	3002
GDH Lines	77.6	122.4	60.8	28.3	19.7	3010
<i>F value</i>	15.94**	4.78*	3.39	8.84**	6.73*	0.03

(*) P<0.05, (**) P<0.01 and (***) P<0.0001

ANOVA for Differences among Individual Lines **within Populations**
TN 97LC Populations

Source of Male Parent	Statistic	Height 50 (cm)	Height topping (cm)	Leaf length (cm)	Leaf width (cm)	Leaves per plant	Yield (Kg/ha)
TN 97 ADH (8 Lines)	MS F value	45.39 0.71	64.33 0.57	8.5 0.76	6.04 1.37	1.74 0.74	67314.9 0.77
TN 97LC (8 Lines)	MS F value	27.56 0.49	38.12 0.86	1.73 0.33	0.34 0.22	1.57 1.3	44094.6 0.84
TN 97 GDH (4 Lines)	MS F value	25.25 0.47	22.37 0.35	19.56 2.39	40.48 10.93***	3.52 1.95	72226.6 1.34

(*) P<0.05, (**) P<0.01 and (***) P<0.0001

Summary

- On average, GDH lines had superior agronomic performance in comparison to ADH lines; for the TN 90 population, the difference between ADH and LC lines was statistically significant.

Summary

- On average, GDH lines had superior agronomic performance in comparison to ADH lines; for the TN 90 population, the difference between ADH and LC lines was statistically significant.
- However, it was much easier to obtain large numbers of haploid plants using the ADH method; this was particularly true for GR 149LC.

Summary

- In comparison to the GDH method, the ADH method of obtaining haploid plants required approximately 73 additional days.

Summary

- In comparison to the GDH method, the ADH method of obtaining haploid plants required approximately 73 additional days.
- Individual ADH and GDH lines that were equal to or superior to their source variety were identified.
- When ADH or GDH lines were used as pollinators in hybrid combinations, mean yields of the hybrid lines were not different from their respective LC hybrids.

Acknowledgements

Appreciation is expressed to Altria, Philip Morris International, R.J. Reynolds, BAT, The American Snuff Company, the Council for Burley Tobacco, Burley Growers' Cooperative, and KTRDC for funding the Kentucky-Tennessee Tobacco Improvement Initiative.