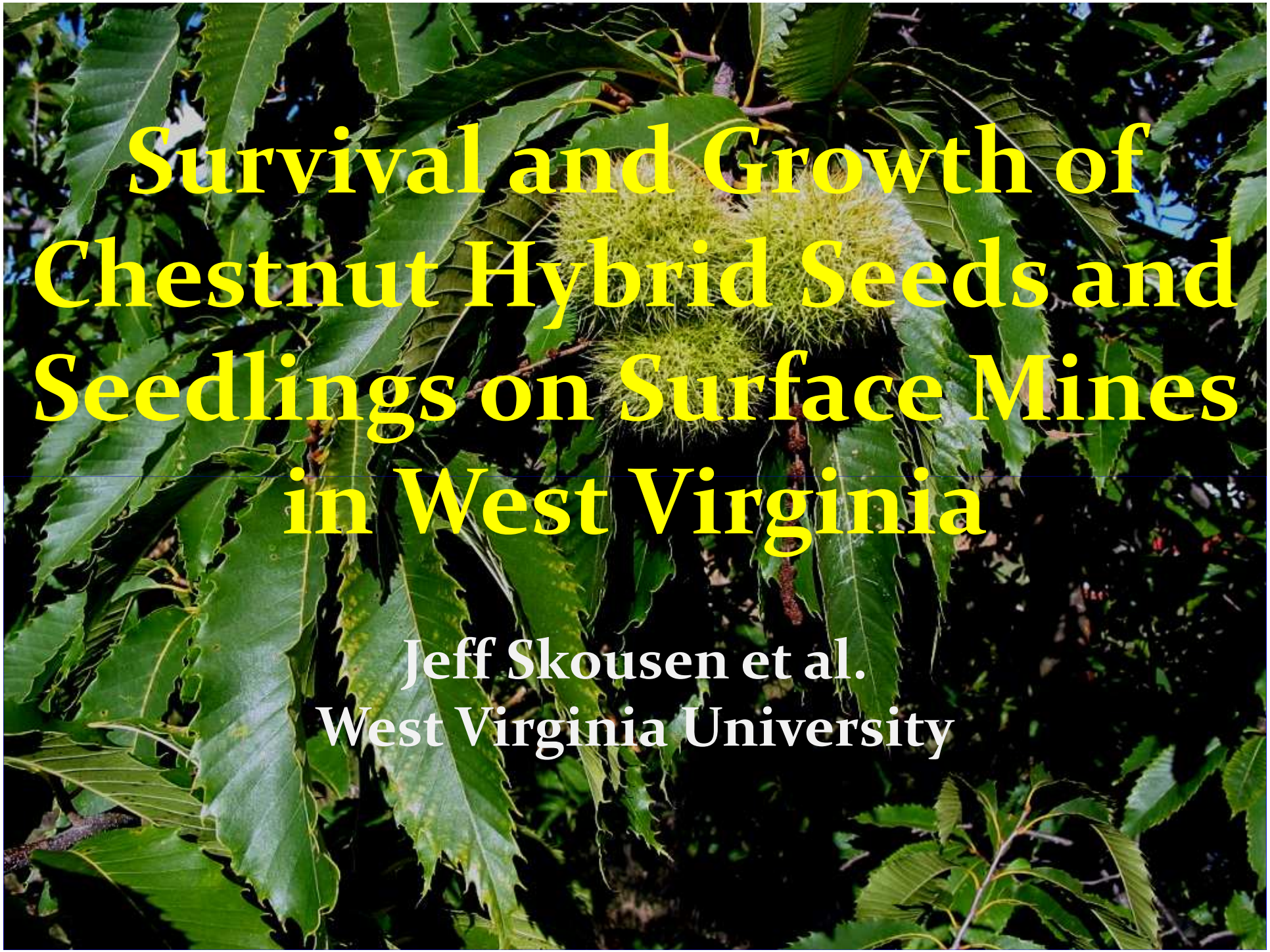


Survival and Growth of Chestnut Hybrid Seeds and Seedlings on Surface Mines in West Virginia

Jeff Skousen et al.
West Virginia University

Chestnut

“Redwood of the East”

1 in 4 Trees were Chestnuts

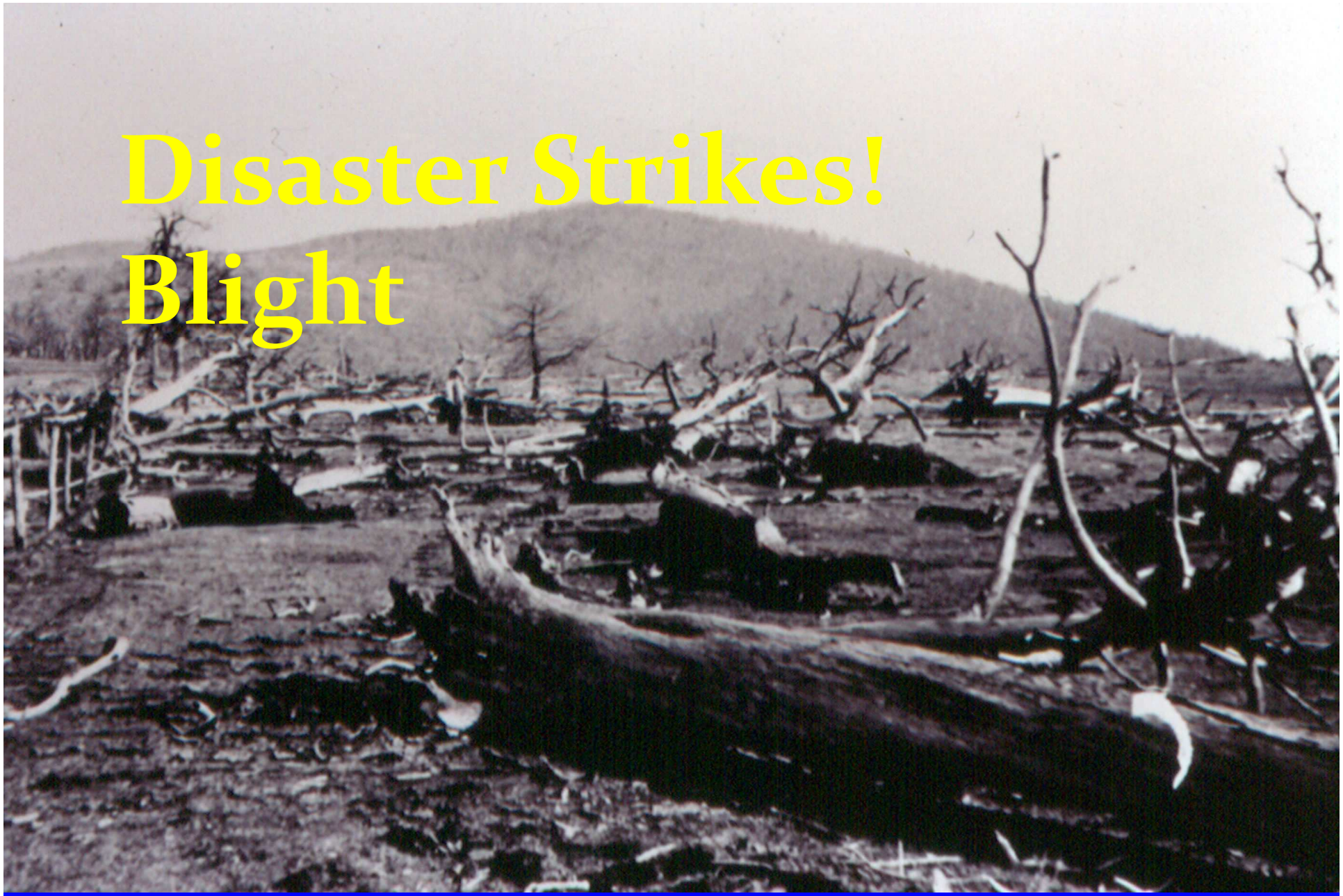
Degradation

Old Stump

Prolific Nut Producer

1904 Discovered
1950 Entire Range
Affected

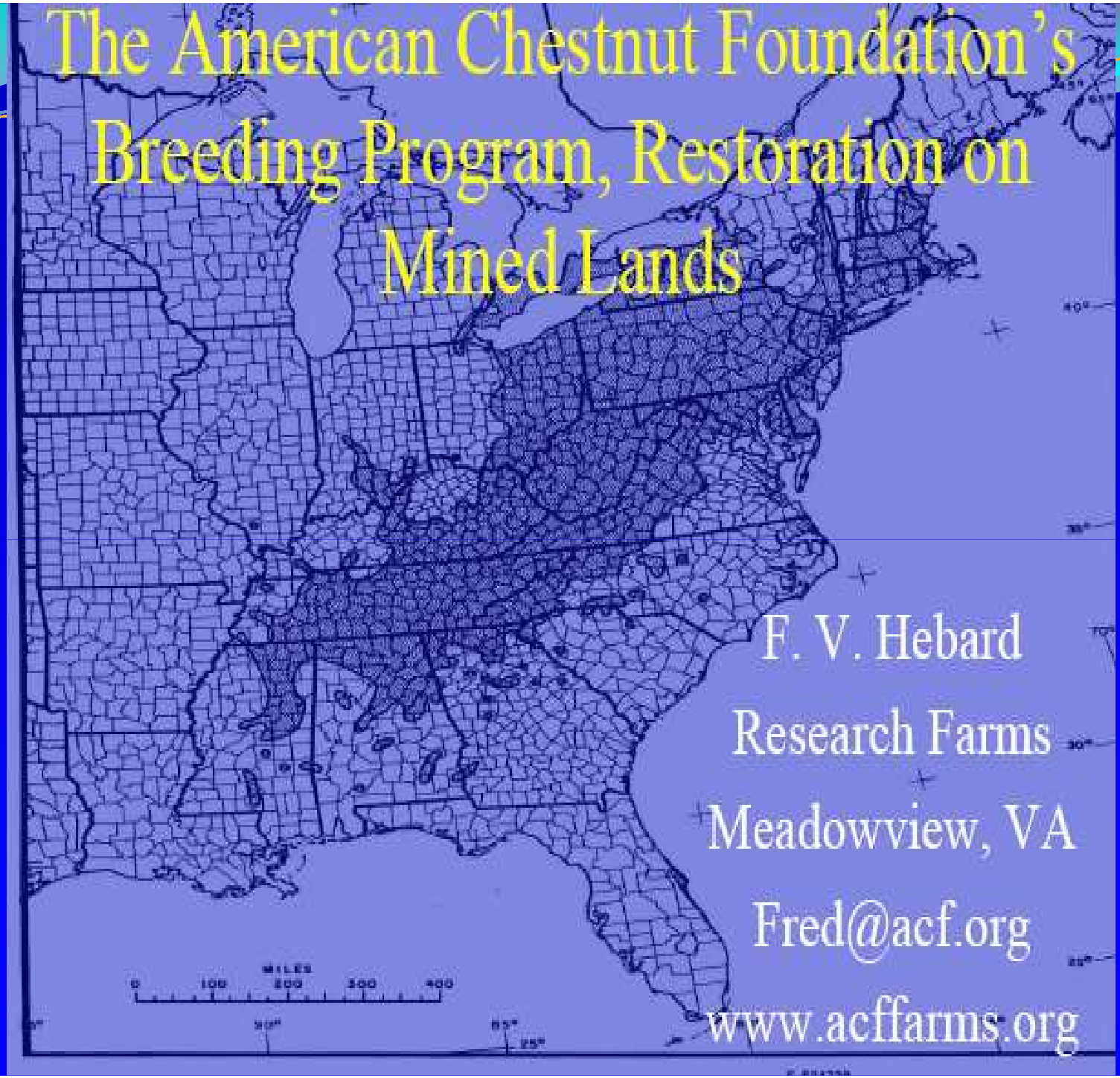
Disaster Strikes!
Blight



Blight fungus infects through wounds in the bark.

The American Chestnut Foundation

The American Chestnut Foundation's Breeding Program, Restoration on Mined Lands



F. V. Hebard
Research Farms
Meadowview, VA
Fred@acf.org
www.acffarms.org

Backcrossing

Expected Proportion Chinese	
	C x A ↓
1/2	F_1 x A ↓
1/4	B_1 x A ↓
1/8	B_2 x A ↓
1/16	B_3 x B_3 ↓
1/16	B_3-F_2 x B_3-F_2 ↓
1/16	B_3-F_3

Blight-resistant Chinese to American B_3-F_2.

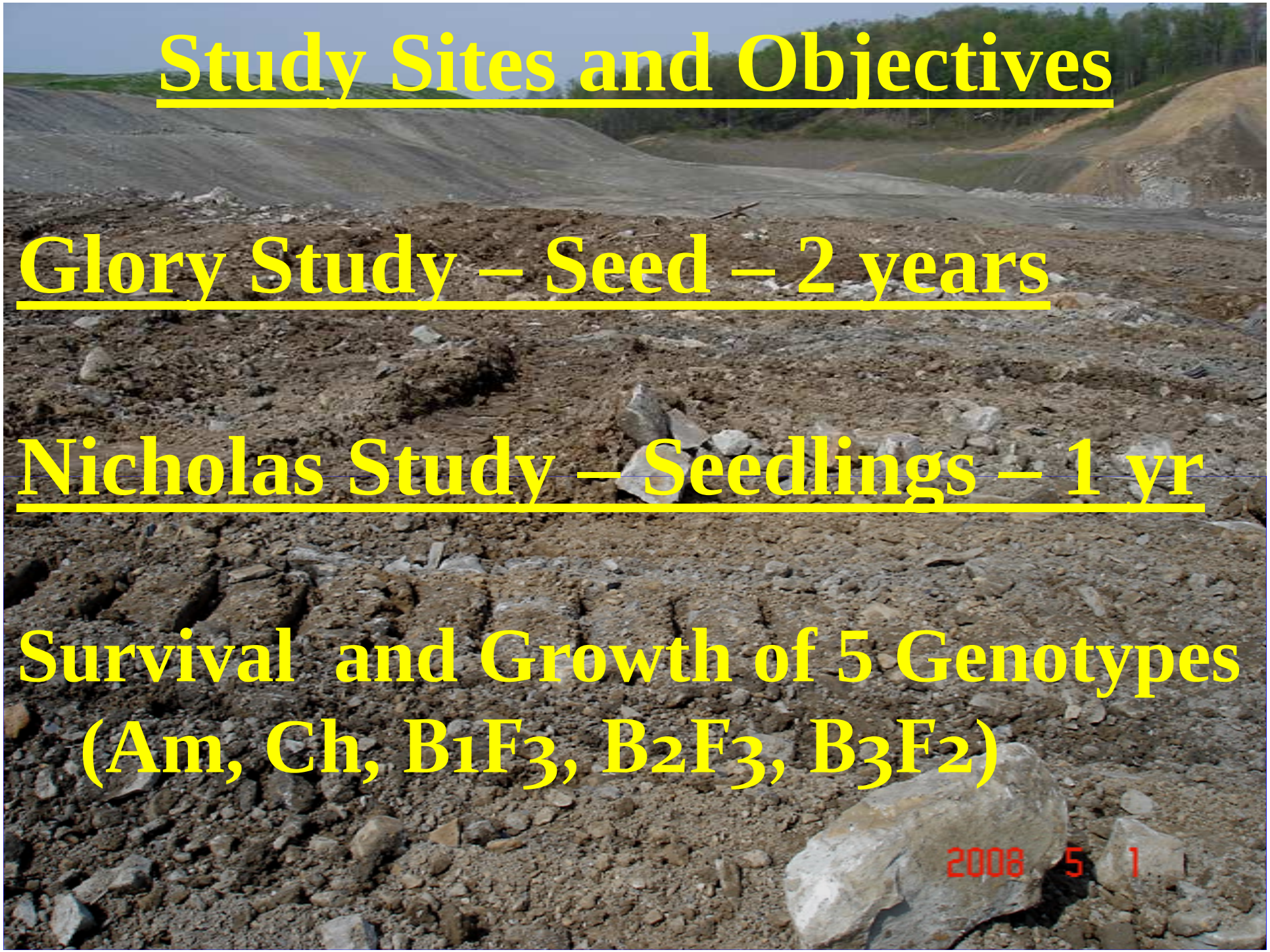
Nuts from this generation will be planted back into the forest and onto mined land.

Operation Springboard

Appalachian Regional Reforestation Initiative (ARRI)

TACF and Several Universities

Chestnut Tree Planting in Kentucky

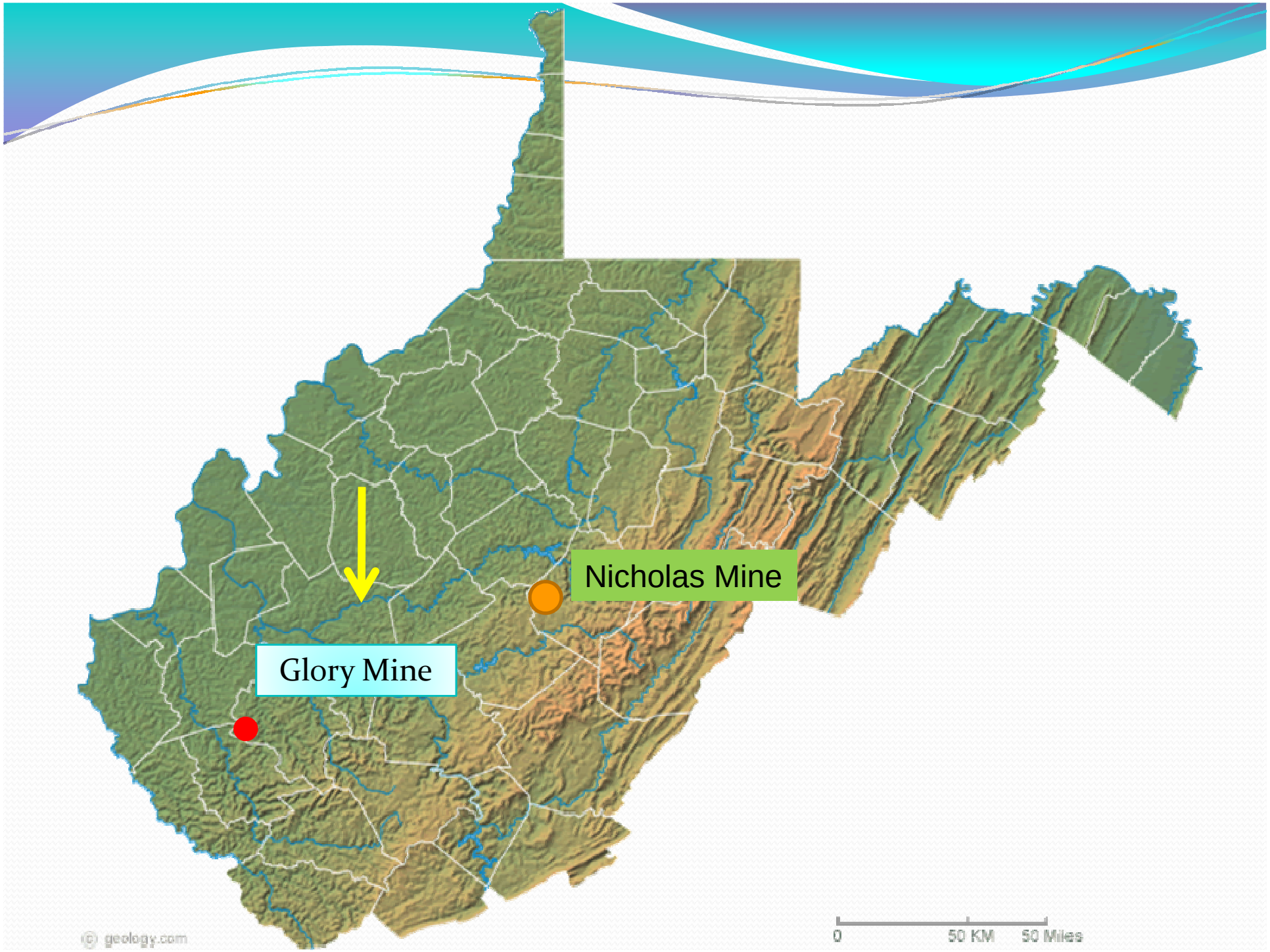


Study Sites and Objectives

Glory Study – Seed – 2 years

Nicholas Study – Seedlings – 1 yr

Survival and Growth of 5 Genotypes (Am, Ch, B1F3, B2F3, B3F2)


Nicholas Mine
Glory Mine
© geology.com
0
50 KM
50 Miles

Glory Mine
Mixed, loosely-graded
Brown-Gray mine soil
2008 5 1

Randomized Complete Block Design – WVU/ACF Chestnut Seed Planting

Glory Mine

BLOCK 1

B2F3	B3F2
AMERICAN	CHINESE
B1F3	B2F3
B3F2	B1F3
CHINESE	AMERICAN
np	p

BLOCK 2

B2F3	B2F3
CHINESE	AMERICAN
AMERICAN	B1F3
B1F3	CHINESE
B3F2	B3F2
p	np

BLOCK 3

CHINESE	AMERICAN
AMERICAN	B1F3
B2F3	CHINESE
B1F3	B3F2
B3F2	B2F3
p	np

BLOCK 4

B2F3	AMERICAN
B3F2	B2F3
B1F3	CHINESE
AMERICAN	B1F3
CHINESE	B3F2
np	p

TUBES

NO TUBES

BLOCK 5

AMERICAN	CHINESE
CHINESE	B2F3
B3F2	AMERICAN
B2F3	B3F2
B1F3	B1F3
np	p

BLOCK 6

B1F3	AMERICAN
AMERICAN	B1F3
CHINESE	B2F3
B2F3	B3F2
B3F2	CHINESE
np	p

BLOCK 7

B2F3	AMERICAN
B1F3	B2F3
B3F2	B3F2
AMERICAN	B1F3
CHINESE	CHINESE
p	np

BLOCK 8

B1F3	CHINESE
B3F2	B3F2
AMERICAN	AMERICAN
CHINESE	B1F3
B2F3	B2F3
np	p

BLOCK 1

B2F3	B3F2
AMERICAN	CHINESE
B1 F3	B2F3
B3F2	B1F3
CHINESE	AMERICAN
np	p

5 seeds of each genotype were seeded in each side with either a Peat or No Peat Treatment

Backcrossing

Expected Proportion Chinese	Chinese American
	C x A
1/2	F_1 x A
1/4	B_1 x A
1/8	B_2 x A
1/16	B_3 x B_3
1/16	B_3-F_2 x B_3-F_2
1/16	B_3-F_3

So we have seeds of:
Chinese
American
B1F3
B2F3
B3F2

2008 5 1

Planting Procedure
2008 5 1

Peat
2008 5 1

Seed in Peat
2008 5 1

Cover Seed

Graduate Students didn't do all the work!

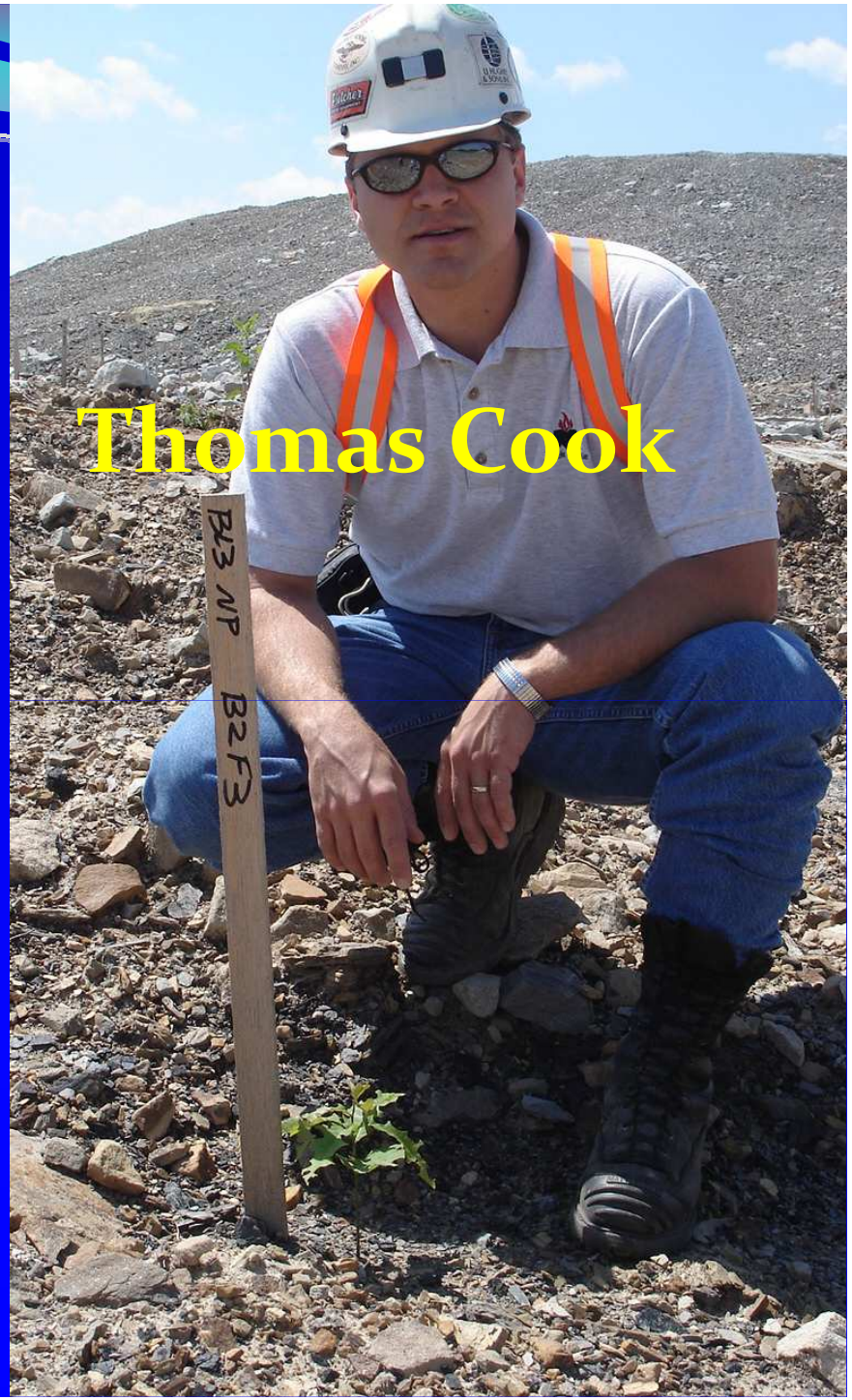
Skousen Planted!
2008 5 2

Tubed Trees

Un-tubed Trees
2008 5 14

Seed ID

7 Weeks later
BL4 P CHEN
2008 6 24


Thomas Cook
BL3 NP B2F3

Statistical Analyses

Survival and Height

T-tests for whole plots

Shelters

Peat

Species within whole plots

Soil Sampling
5 samples from each block
2008 5 1

Chemical Soil Properties

Block	pH	P	K	Ca	Mg	CEC	BS
		mg kg^{-1}	---------	cmol^{+} kg^{-1}		----------	%
1	5.3b	25	0.15	0.76b	1.85	9	30b
2	5.6ab	26	0.17	4.32a	2.08	12	53ab
3	5.2b	23	0.14	0.84b	1.78	12	28b
4	5.5ab	33	0.15	1.05b	2.30	12	40b
5	6.7a	41	0.15	3.86a	2.09	8	79a
6	5.5ab	30	0.12	3.50a	1.70	10	55ab
7	6.2a	35	0.11	2.59ab	1.97	10	64ab
8	6.6a	40	0.15	2.93ab	1.78	7	80a

Soil Chemical Properties between trees with and without shelters

Block	pH	P	K	Ca	Mg	CEC	BS
		mg kg^{-1}	----------	$cmol^{+}$ kg^{-1}		----------	%
Shelters	5.4	27	0.15	1.74	1.93	11	38
No shelters	5.7	36	0.14	3.22	1.96	9	70

Chestnut seed survival with and without tree shelters across all five genotypes at Glory.

*Shelter treatments with different letters are significantly different with an LSD test at $p<0.05$

Treatment	Survival	
	2008	2009
	---------- % ----------	
Shelters	**81 a***	**64 a**
No Shelters	**63 b**	**60 a**

Chestnut seed survival with and without peat treatment across all five genotypes at Glory.

*Shelter treatments with different letters are significantly different with an LSD test at $p<0.05$

Treatment	Survival	
	2008	2009
	---------- % ----------	
Peat	**63 b***	**52 b**
No Peat	**81 a**	**72 a**

Chestnut seed survival for five genotypes across shelter and peat treatments.

*Values with different letters are significantly different with an LSD test at $p<0.05$

Type	Survival	
	2008	2009
	---------- %	----------
American	67 b*	55 b
Chinese	**82 a**	86 a
B1F3	74 b	53 b
B2F3	69 b	53 b
B3F2	72 b	52 b
Totals	72	62

Chestnut seed height for five genotypes across shelter and peat treatments.

*Values with different letters are significantly different with an LSD test at $p<0.05$

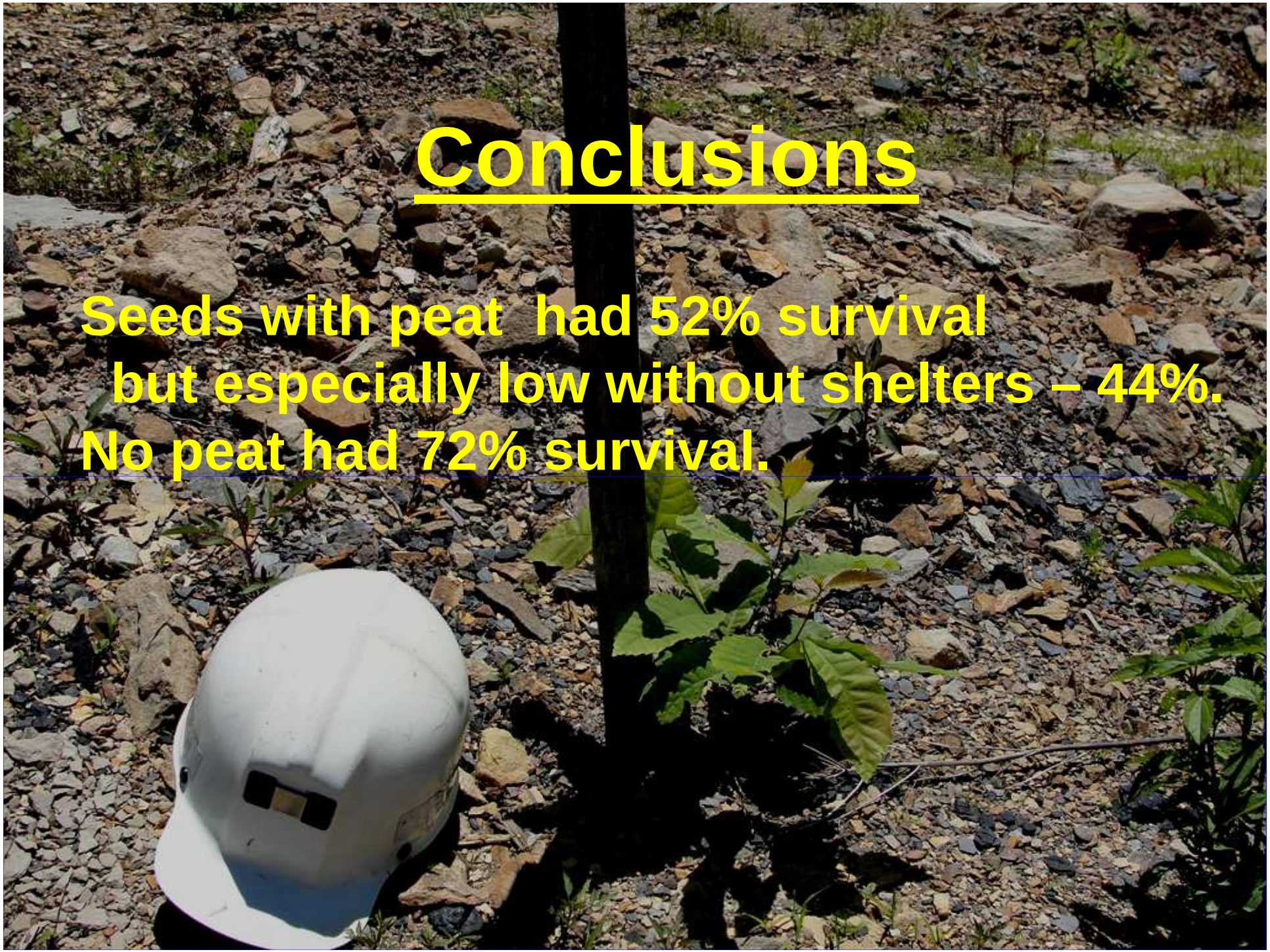
Type	Height	
	2008	2009
	---------- cm ----------	
American	18 c*	10 a
Chinese	**26 a**	13 a
B1F3	22 b	12 a
B2F3	20 b	13 a
B3F2	22 b	11 a
Totals	22	12

Conclusions

Mixed brown/gray sandstone – pH 5.3 to 6.7

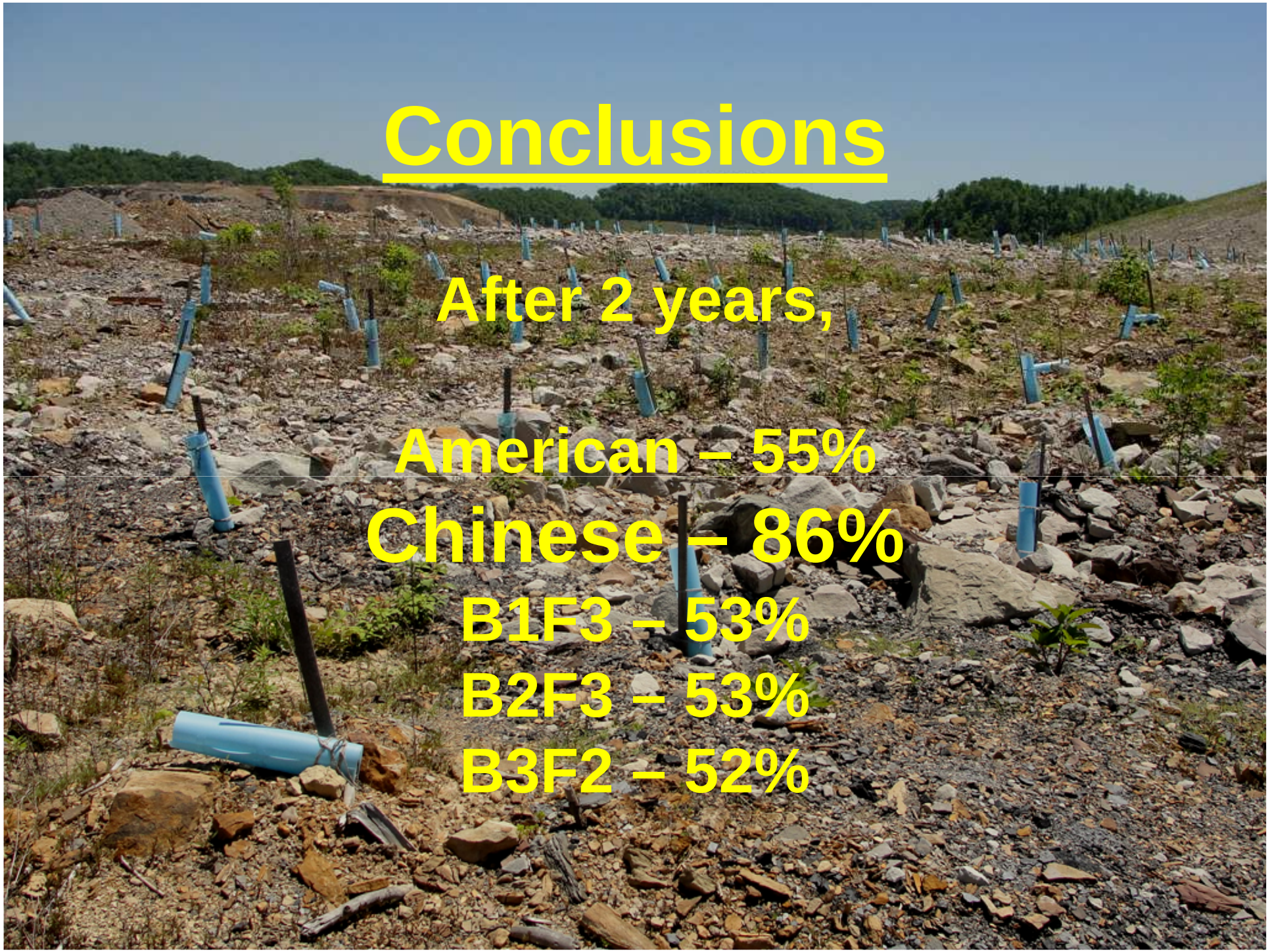
Conclusions

After 2nd year,
Seeds with tree shelters – 64% survival
Seeds without shelters – 60%



Conclusions

Seeds with peat had 52% survival but especially low without shelters – 44%. No peat had 72% survival.



Conclusions

After 2 years,

American – 55%

Chinese – 86%

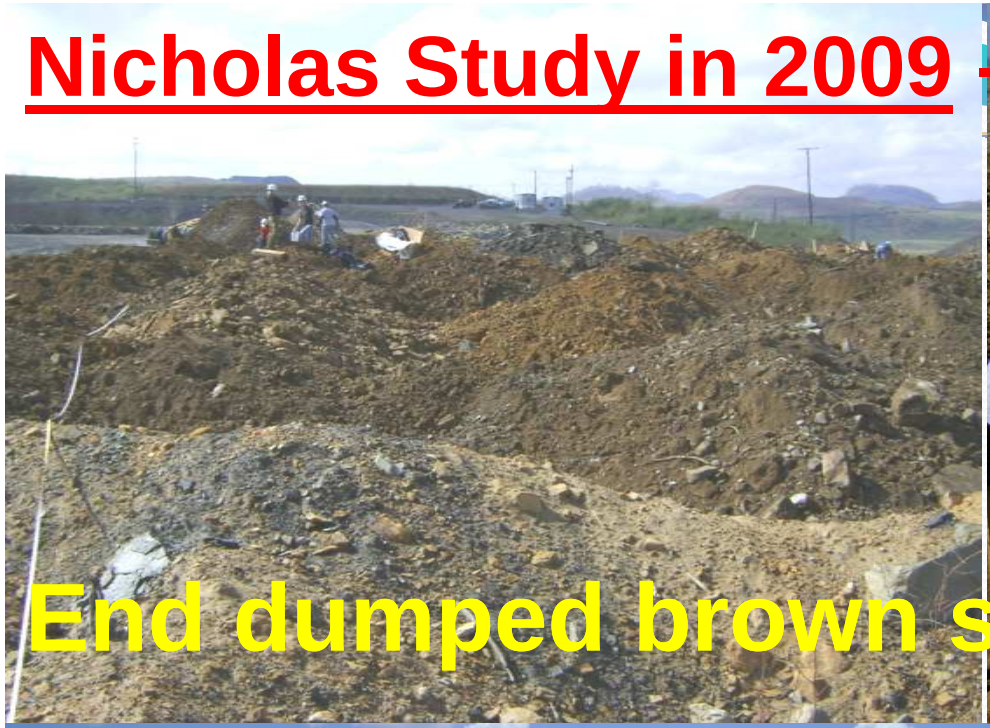
B1F3 – 53%

B2F3 – 53%

B3F2 – 52%

Nicholas Mine
Glory Mine
© geology.com
0
50 KM
50 Miles

Nicholas Study in 2009 – Seedlings and Seeds



End dumped brown sandstone

Heavily graded gray sandstone

Seeds and Seedlings

Hilly, Brown Sandstone – No compact

BLOCK 1	
B_2F_3 - Seed	B_3F_2 - Seedling
AMER - Seed	CHIN - Seedling
B_1F_3 - Seedling	B_2F_3 - Seedling
B_3F_2 - Seed	B_1F_3 - Seed
CHIN - Seed	AMER - Seedling
BLOCK 2	
B_2F_3 - Seed	B_2F_3 - Seedling
CHIN - Seedling	AMER - Seedling
AMER - Seed	B_1F_3 - Seedling
B_1F_3 - Seed	CHIN - Seed
B_3F_2 - Seedling	B_3F_2 - Seed
BLOCK 3	
CHIN - Seed	AMER - Seedling
AMER - Seed	B_1F_3 - Seedling
B_2F_3 - Seed	CHIN - Seedling
B_1F_3 - Seed	B_3F_2 - Seedling
B_3F_2 - Seedling	B_2F_3 - Seed
BLOCK 4	
B_2F_3 - Seedling	AMER - Seedling
B_3F_2 - Seedling	B_2F_3 - Seed
B_1F_3 - Seed	CHIN - Seed
AMER - Seedling	B_1F_3 - Seedling
CHIN - Seedling	B_3F_2 - Seed
BLOCK 5	
B_1F_3 - Seedling	CHIN - Seed
B_2F_3 - Seed	B_1F_3 - Seed
AMER - Seed	B_3F_2 - Seed
B_3F_2 - Seedling	B_2F_3 - Seedling
CHIN - Seedling	AMER - Seedling

Smooth, Gray Sandstone - Compact

BLOCK 1	
AMER - Seed	CHIN - Seed
CHIN - Seedling	B_2F_3 - Seed
B_3F_2 - Seedling	AMER - Seedling
B_2F_3 - Seedling	B_3F_2 - Seed
B_1F_3 - Seed	B_1F_3 - Seedling
BLOCK 2	
B_1F_3 - Seed	AMER - Seed
AMER - Seedling	B_1F_3 - Seedling
CHIN - Seed	B_2F_3 - Seed
B_2F_3 - Seedling	B_3F_2 - Seed
B_3F_2 - Seedling	CHIN - Seedling
BLOCK 3	
B_2F_3 - Seed	AMER - Seedling
B_1F_3 - Seed	B_2F_3 - Seedling
B_3F_2 - Seedling	B_3F_2 - Seed
AMER - Seed	B_1F_3 - Seedling
CHIN - Seedling	CHIN - Seed
BLOCK 4	
B_1F_3 - Seed	CHIN - Seed
B_3F_2 - Seed	B_3F_2 - Seedling
AMER - Seed	AMER - Seedling
CHIN - Seedling	B_1F_3 - Seedling
B_2F_3 - Seed	B_2F_3 - Seedling
BLOCK 5	
AMER - Seedling	B_2F_3 - Seed
CHIN - Seed	AMER - Seed
B_3F_2 - Seedling	B_3F_2 - Seed
B_1F_3 - Seed	CHIN - Seedling
B_2F_3 - Seedling	B_1F_3 - Seedling

BLOCK 1

B_2F_3 - Seed	B_3F_2 - Seedling
AMER - Seed	CHIN - Seedling
B_1F_3 - Seedling	B_2F_3 - Seedling
B_3F_2 - Seed	B_1F_3 - Seed
CHIN - Seed	AMER - Seedling

Backcrossing

Expected Proportion Chinese	Chinese American
	C x A
	↓
1/2	F_1 x A
	↓
1/4	B_1 x A
	↓
1/8	B_2 x A
	↓
1/16	B_3 x B_3
	↓
1/16	B_3-F_2 x B_3-F_2
	↓
1/16	B_3-F_3

So we have Seed and Seedlings

Chinese

Amer

B1F3

B2F3

B3F2

Planting Procedure - Brown

Seeds

Planting Procedure – Gray

Much more difficult

Seeds

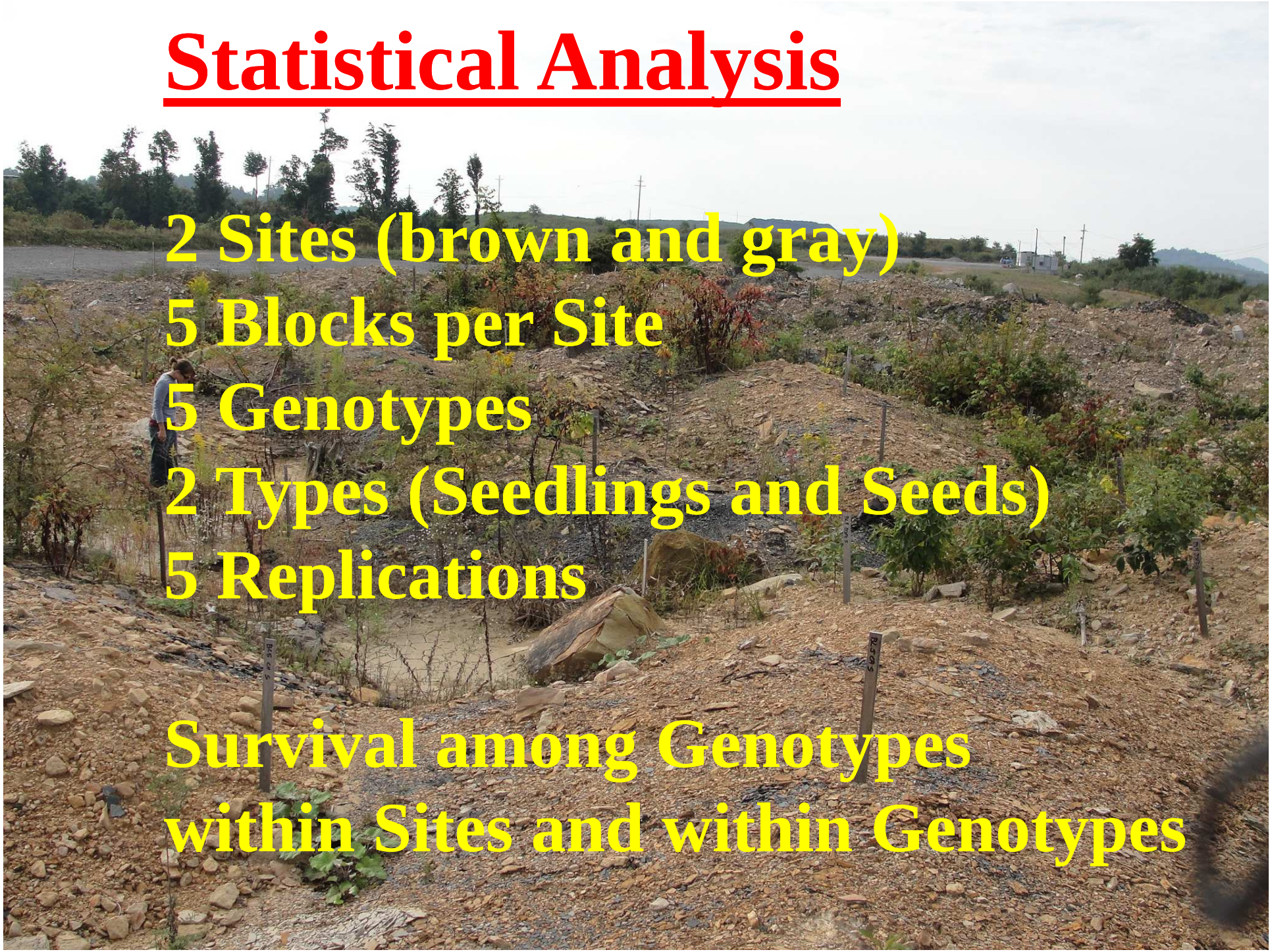
Brown Sandstone End-Dumped Site

Gray Sandstone Compacted Site

B-10 AM +

Rio Bas 1

Statistical Analysis



2 Sites (brown and gray)
5 Blocks per Site
5 Genotypes
2 Types (Seedlings and Seeds)
5 Replications

Survival among Genotypes
within Sites and within Genotypes

Soil Sampling
15 samples from each site

Nicholas Study - Soil Chemical Properties

Substrate	pH	P	K	Ca	Mg	CEC	BS
		mg kg^{-1}	-------	$cmol^{+}$ kg^{-1}		---------	%
Brown	4.5 b	6 b	0.33	2.9 b	3.6 b	13 b	28 b
Gray	6.6 a	56 a	0.40	9.5 a	6.2 a	8 a	100 a

SIX SEEDS
out of 250!

Chestnut seedling survival for five genotypes

*Values with different letters are significantly different with an LSD test at $p<0.05$

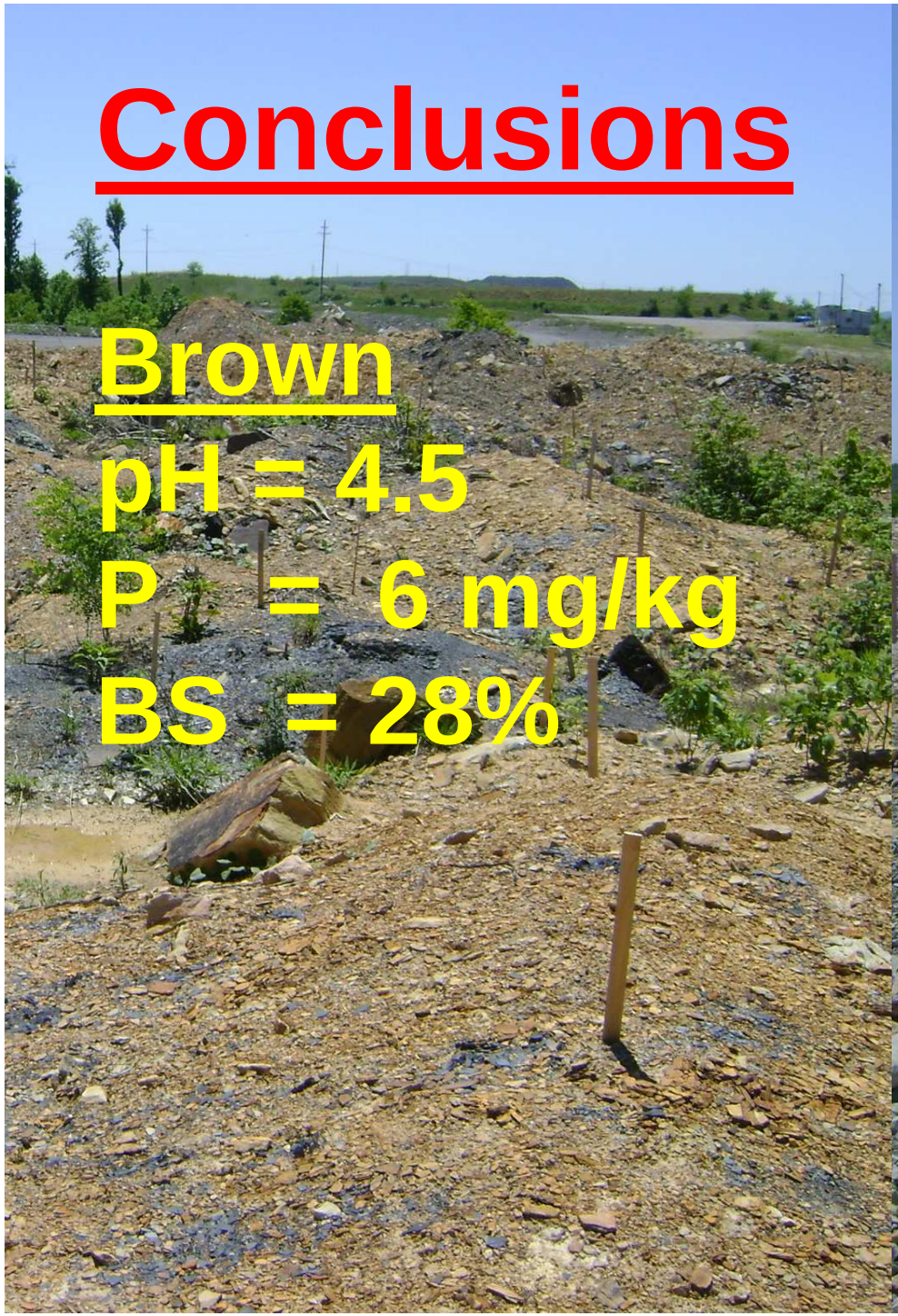
Type	Survival		
	Brown	Gray	Ave
	----------	%	----------
American	93 a	92 a	93
Chinese	100 a	100 a	100
B1F3	96 a	92 a	96
B2F3	85 a*	48 c	67
B3F2	67 b	68 b	67
Totals	88	80	85

Chestnut seedling height for five genotypes

*Values with different letters are significantly different with an LSD test at $p<0.05$

Seed Type	Height		
	Brown	Gray	Ave
	---------- cm ----------		
American	24 b	21 b	23
Chinese	40 a	31 a	36
B1F3	15 c	15 c	15
B2F3	12 cd*	6 d	9
B3F2	9 d	5 d	7
Totals	20	16	18

Conclusions



Brown

pH = 4.5

P = 6 mg/kg

BS = 28%

Gray

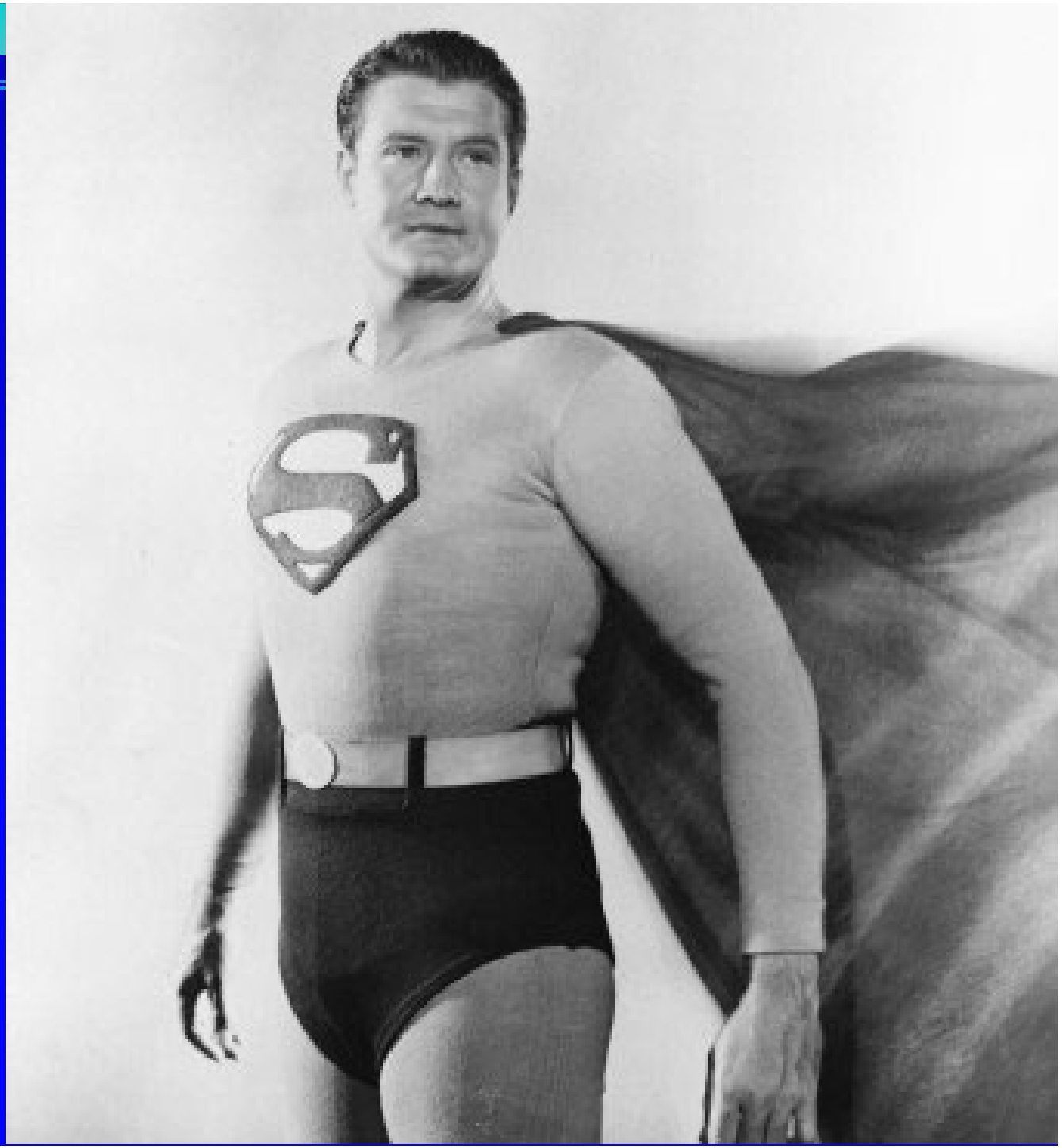
pH = 6.6

P = 56 mg/kg

BS = 100%

Conclusions

Only 6 of 250 seeds germinated!

Ave Survival of Seedlings was >90% for American, Chinese, and B_1F_3

I can answer anything!