SURVIVAL AND GROWTH OF CHESTNUT HYBRID SEEDS AND SEEDLINGS ON MOUNTAINTOP SURFACE MINES IN WEST VIRGINIA[1]

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Abstract: Reforestation of mined lands in Appalachia with chestnut is occurring on a few reclamation projects in West Virginia and is a focus of the Appalachian Regional Reforestation Initiative and the American Chestnut Foundation. In West Virginia, we established two studies to evaluate the survival and growth of chestnuts on mined lands using seeds and seedlings. The first study, initiated in 2008, included planting of five types of chestnut seeds (100% American, 100% Chinese, and three hybrids [B_1F_3, B_2F_3, and B_3F_2]) into loosely-graded minesoils at the Glory surface mine in Boone County, West Virginia. First year survival was Chinese 82%, American 67%, and the hybrids between 69 and 74%. After the 2nd year, survival had declined for all seed types except Chinese: Chinese 86%, American 55%, and hybrids to 59%. Average height after the 2nd season was not significantly different among seed types (mean height of 12 cm). A second study, initiated in 2009, involved planting seeds and seedlings of these same five chestnut types into two substrates (end-dumped brown sandstone/topsoil and into compacted gray sandstone) in a completely randomized block design. Only six seeds (of 250 planted) germinated, which was surprisingly poor after the successful establishment from seed the previous year at the Glory site. However, chestnut seedling survival on brown sandstone and gray sandstone was 100% for Chinese, 93% for American, 96% for the B_1F_3 hybrid, and 68% for the B_3F_2 hybrid. Only the B_2F_3 hybrid showed much lower survival on the gray sandstone (48%) vs the brown sandstone (85%).

Additional Keywords: chestnut hybrids, Forestry Reclamation Approach, minesoils, tree seedlings, tree seeds

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Introduction

Forestry post-mining land uses have gradually emerged during the early 2000s as a preferred post-mining land use option in the Appalachian Region, and especially in West Virginia. To encourage forest re-establishment on mined lands and to optimize the success of tree plantings, the Forestry Reclamation Approach (FRA) of the Appalachian Regional Reforestation Initiative (ARRI) was initiated. ARRI encourages the use of the FRA's five-step process to reclaim coal mined land to forestland:

1. Create a suitable rooting medium for good tree growth that is no less than 1.2 m (4 ft) deep and comprised of topsoil, weathered sandstone, and/or the best available material;
2. Loosely grade the topsoil or topsoil substitutes established in step one to create a non-compacted growth medium;
3. Use ground covers that are compatible with growing trees;
4. Plant two types of trees – 1) early succession species for wildlife and soil stability, and 2) commercially valuable crop trees;
5. Use proper tree planting techniques (Burger et al., 2005).

Recent research has demonstrated the usefulness of the FRA by showing the successful establishment of native hardwood trees when applying this five-step process (Angel et al., 2008; Emerson et al., 2009). Coal operators and regulators are gradually seeing an increase of acreage being reclaimed to forestry post-mining land uses (Angel et al., 2009).

Prior to the 1900s, the eastern hardwood forests of the United States were comprised of an assemblage of 30 or 40 hardwood species. One of the most important species was the American chestnut (*Castanea dentata* (Marsh.) Borkh.), and foresters estimated that this species occupied up to 25% of the forest. American chestnut produced great volumes of timber because it grew straight, fast, and often produced three or four 4-m logs before the first branch was reached.

Chestnut blight, discovered in 1904 in New York, is caused by a fungus (*Cryphonectria parasitica* (Murr.)Barr.), which quickly spread through the eastern US forests. By 1950, about 4 billion trees had perished, nearly one-fourth of the canopy cover of the eastern deciduous forest was gone, and an important wildlife and timber tree was lost. The blight fungus infects American chestnut through wounds in the bark, creating a canker which effectively cuts off circulation to the branches above the canker. The roots, however, remain alive. The ability to sprout has enabled American chestnut to persist in eastern forests, but only as an occasional understory shrub.

The American Chestnut Foundation (TACF), formed in 1983, is crossing surviving American chestnut flowers with blight-resistant Asiatic chestnut. Therefore, these hybrids incorporate Asiatic chestnut's blight resistance while retaining the desirable timber and nut-producing characteristics of the American chestnut. In 2009, TACF produced tree seedlings that are approximately 7/8 American chestnut and 1/8 Chinese chestnut (the B_3F_2 hybrid is the third backcross to American chestnut and the second generation).

The use of reclaimed surface mines for chestnut reestablishment has recently gained momentum (French et al., 2007b). In cooperation with the University of Kentucky, chestnut seeds were planted in 2005 on end-dumped spoil in eastern Kentucky composed of gray sandstone, brown sandstone, and run-of-mill spoil materials. Better growth was found in brown sandstone (Adank et al., 2008; French et al., 2007a). Researchers in Ohio have been examining chestnut direct seeding versus planted seedlings, mycorrhizal inoculation treatments, and protection of seedlings on mine lands (McCarthy et al., 2008). A breeding orchard of hybrid chestnut seedlings on mined land was established in Jefferson County, PA, and it is anticipated that selections and harvesting of nuts will be performed by 2010 (Phelps, 2002).

The objective of this study is to evaluate the survival and growth of chestnuts on mined lands using seeds and seedlings. The first experiment (Glory Study) evaluates establishment and growth of five chestnut seed types planted into a mixed brown/gray sandstone substrate for which we have two years of data. The second experiment, with only one year of data (Nicholas Study), compares the establishment and growth of both seeds and seedlings of five chestnut types in a loosely-dumped brown sandstone material and in a compacted gray sandstone material.

Materials and Methods

Glory Study - 2008

The Glory surface mine is located near Van, in Boone County, West Virginia. Overburden from the Number 5 Block and Clarion coal seams was used to construct a 1-ha plot for this experiment, which was comprised of 75% brown sandstone and 25% gray sandstone. The material was end dumped by trucks and a large bulldozer flattened the tops of the piles to create a rough level surface (Fig. 1). Precipitation is about 112 cm with 60% falling between April and September, the recognized growing season (Wolf, 1994). The average annual temperature during the growing season is 20 degrees C.

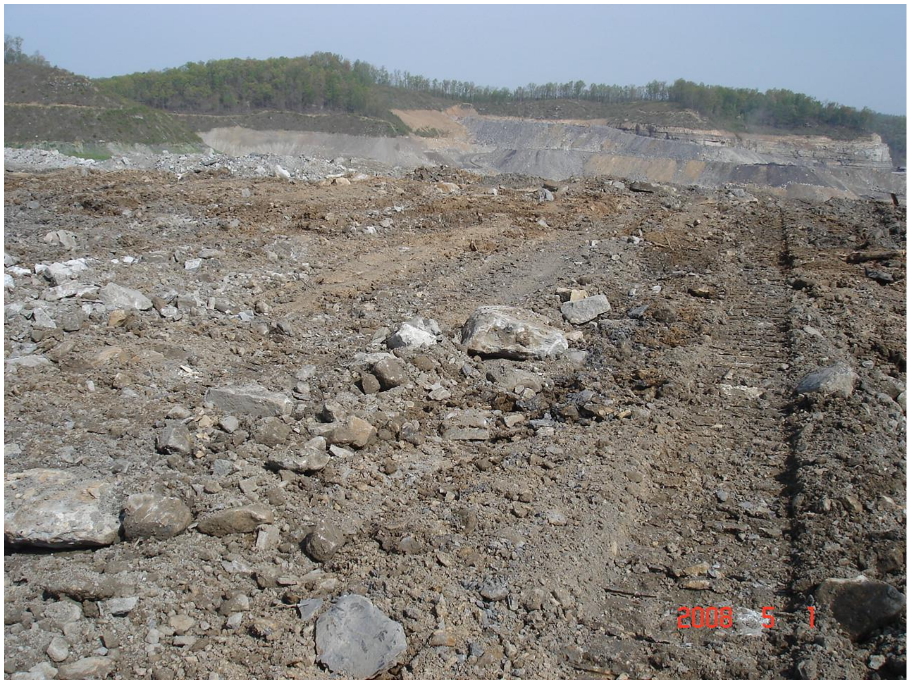

Figure 1. The 1-ha experimental area was constructed with primarily brown sandstone substrate, end-dumped with trucks, and the piles were flattened by one or two passes of a bulldozer.

On this 1-ha site, the experimental setup consisted of two, split plot designs with tree shelters (with or without) being the whole plot factor. Each whole plot (shelters or no shelters) was composed of four blocks. One half of each block was randomly assigned a peat or no peat treatment. In each half block, five subplots were randomly assigned a seed type (Fig. 2). Five seeds of the assigned seed type were planted in each subplot at 2.4 x 2.4 m spacing. The chestnut seeds were provided by Fred Hebard and Bob Paris of the American Chestnut Foundation in Meadowview, VA. Wooden stakes were driven into the soil at each seed location. In total, 80 seeds of each seed type were planted for a total of 400 seeds (5 seed types x 8 blocks x 2 peat treatments x 5 replications = 400 seeds).

Seeds were planted by digging a small 5-cm-deep hole about 5 cm from the base of the wooden stake. Each seed was inoculated with mycorrhizal fungi provided by the American Chestnut Foundation before planting. In peat treatments, about 5 cm^3 of commercial peat from a local gardening store was placed in the hole and the seed was placed on the peat and covered with soil. In the no peat treatment, only soil was used to cover the seed. After planting, 45-cm-tall, plastic tree shelters were placed over each planted seed in blocks 1, 2, 5 and 6 (200 seeds), and no tree shelters were placed on planted seeds in blocks 3, 4, 7 and 8. The tree shelters were secured to the stakes with twine. Shelters were removed during the second growing season in June 2009. No fertilizer was applied at the time of planting or during the 2nd year of growth. Survival was noted and height of each live chestnut seedling was measured in late August 2008 and again in August 2009.

BLOCK 1

B_2F_3	B_3F_2
AMERICAN	CHINESE
B_1F_3	B_2F_3
B_3F_2	B_1F_3
CHINESE	AMERICAN
np	p

BLOCK 5

AMERICAN	CHINESE
CHINESE	B_2F_3
B_3F_2	AMERICAN
B_2F_3	B_3F_2
B_1F_3	B_1F_3
np	p

BLOCK 2

B_2F_3	B_2F_3
CHINESE	AMERICAN
AMERICAN	B_1F_3
B_1F_3	CHINESE
B_3F_2	B_3F_2
p	np

BLOCK 6

B_1F_3	AMERICAN
AMERICAN	B_1F_3
CHINESE	B_2F_3
B_2F_3	B_3F_2
B_3F_2	CHINESE
np	p

BLOCK 3

CHINESE	AMERICAN
AMERICAN	B_1F_3
B_2F_3	CHINESE
B_1F_3	B_3F_2
B_3F_2	B_2F_3
p	np

BLOCK 7

B_2F_3	AMERICAN
B_1F_3	B_2F_3
B_3F_2	B_3F_2
AMERICAN	B_1F_3
CHINESE	CHINESE
p	np

BLOCK 4

B_2F_3	AMERICAN
B_3F_2	B_2F_3
B_1F_3	CHINESE
AMERICAN	B_1F_3
CHINESE	B_3F_2
np	p

BLOCK 8

B_1F_3	CHINESE
B_3F_2	B_3F_2
AMERICAN	AMERICAN
CHINESE	B_1F_3
B_2F_3	B_2F_3
np	p

Figure 2. Split-split plot design experiments in the Glory Study with 1) seeds with shelters (blocks 1, 2, 5 and 6) and 2) seeds with no shelters (blocks 3, 4, 7 and 8) being the whole plot component, and peat treatment being the subplot. Seed types were considered treatments randomly assigned in each peat subplot. Seed types were American, Chinese, B_1F_3, B_2F_3, and B_3F_2.

Statistical analyses were performed using SAS 9.1 software (SAS Institute, 2005). Using Proc GLM means statement, Fisher's t-tests were applied to test for differences in mean chestnut seed survival and growth between whole plots (shelters and no shelters). Significant differences in means were separated by the LSD test at an alpha level of 0.05. Within whole plots, ANOVA was used to evaluate the split plot experimental design and statistically assess the differences in survival and growth between peat treatments and among seed types. An alpha level of 0.05 was considered significant.

<u>Nicholas Study – 2009</u>

A second study was established at the Nicholas Energy site about 15 km west of Summersville, WV. Nicholas produces about 2.4 million metric tons of high quality coal per year using large shovels, trucks, and dozers. The brown sandstone and topsoil materials came from the surface which overlies the No. 5 Block coal seam, and was end dumped by trucks with no striking off or flattening of the piles (Fig. 3). The gray sandstone substitute material came from the overburden above the Clarion coal seam. This gray sandstone was placed and compacted for typical reclamation and hydroseeding of forages, and was composed of coarse-textured materials and rocks (Fig. 4). Precipitation at the site is about 118 cm with 55% falling between April and September, the recognized growing season (Carpenter, 1992).

Chestnut seedlings and seeds were provided by Fred Hebard and Bob Paris of the American Chestnut Foundation in Meadowview, VA. Seedlings and seeds of five chestnut types (three hybrids and American and Chinese) were randomly planted in subplots of each of five blocks in both substrates (Fig. 5). Five replications were planted in each subplot. A total of 250 seeds and seedlings were planted in each substrate (5 chestnut types x 2 seed or seedling x 5 blocks x 5 replications = 250) and 500 for the entire experiment. Wooden stakes were driven in at the point where seedlings or seeds were planted on 2.4 by 2.4 m spacing and labeled. The planting procedure involved digging holes large enough to place the roots of the seedlings into, while the seeds were planted approximately 3-4 cm deep in the soil and covered. Survival was noted and height of each live chestnut seedling was measured in August 2009. For this study, we instituted a vigor rating to evaluate the quality of the seedlings (Table 1).

Figure 3. The 0.3-ha experimental area of brown sandstone and topsoil at Nicholas was constructed in 2009 and simply end-dumped with trucks.

Figure 4. The 0.3-ha experimental area of gray sandstone at Nicholas was constructed in 2009 with primarily gray sandstone and some brown material, and graded and compacted by bulldozers.

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

Figure 5. Completely randomized block design for seeds and seedlings at the Nicholas Study site. Each seed and seedling of each chestnut type were randomly planted in each of five blocks in both substrates.

Table 1. Vigor ratings were assigned based on the health and vigor of each tree according to the criteria below.

Vigor Rating	Description
1	>75% leaves discolored; extensive dieback
2	50%-75% discoloration; dieback present
3	25 - 50% leaves discolored; dieback present
4	25 - 50% leaves discolored; no dieback present
5	< 25% leaf discolored; no dieback present

Statistical analyses were performed using SAS 9.1 software (SAS Institute, 2005). Using ANOVA, significant differences for chestnut seedling survival and growth among hybrid types were evaluated at an alpha level of 0.05. Substrates were also compared by Fisher's t-tests.

Soil Sampling

For the Glory study site, soil samples were extracted at five locations in each block (at the four corners and center) to a depth of 15 cm to evaluate chemical properties. At the Nicholas site, soil samples were extracted from five locations across each substrate type to a depth of 15 cm. Samples were analyzed for pH (1:1 soil:water) with a Beckman 43 pH meter and elemental content by the West Virginia University Soil Testing Laboratory by extracting each sample with a Mehlich 1 extract, which is composed of approximately 0.05*N* HCl and 0.025*N* H_2SO_4. The extract was analyzed with a Perkin Elmer Plasma 400 emission spectrometer for H, Al, P, K, Ca, and Mg. Cation exchange capacity was calculated by summing the above elements and base saturation was calculated as the sum of base cations divided by total cations.

At Glory, statistical analysis for soil means was performed with ANOVA (completely randomized block design) to determine significant differences among blocks for soil parameters and the LSD test was used to separate means when significant (SAS Institute, 2005). At Nicholas, t-tests were used to determine significant differences among substrates for soil parameters.

Results and Discussion

Glory Study

At Glory, soil analysis revealed a pH range from 5.2 to 6.7 across blocks (Table 2). Blocks 5, 7 and 8 had pH above 6.2, while the others were pH < 5.6. Blocks 2, 5 and 6 had significantly higher Ca content than other blocks, which translated into higher base saturation values. No other soil parameters we measured were significantly different among blocks. When comparing Blocks 1, 2, 5 and 6 (where tree shelters were placed) to Blocks 3, 4, 7 and 8 (no tree shelters), the tree shelter blocks appear to be slightly more acidic than the no tree shelter blocks based on pH, Ca, and base saturation. Soil differences between blocks were not significantly different between sheltered and unsheltered trees. We expected some variation in soils among blocks and these values are within anticipated ranges of soil chemical values.

Table 2. Chemical properties of soils in 2008 where five chestnut seed types were planted at the Glory surface mine in West Virginia. Seeds were planted in 8 blocks and tree shelters were placed on blocks 1, 2, 5 and 6 and no shelters were placed on blocks 3, 4, 7 and 8.

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
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 seeds established and survived at a significantly higher rate where tree shelters were placed on top of the seed compared to those that did not have a tree shelter (81 vs 63%) during the first year, but that difference disappeared after the second year (Table 3). The tree shelters may have originally protected the seed from predators but there was no evidence that small mammals or deer had visited the plot. Tree shelters may have also slightly changed the climate and environment within the shelter during the first year. We noticed some heat stress and burning of leaves at the end of the first growing season, plus many of the seedlings were crowded in the shelters. Therefore, in June of 2009, in the middle of the second growing season, we decided to remove the tree shelters, which may have eliminated some of the moisture stress and high temperature within the tube.

Table 3. Chestnut seed survival with and without tree shelters across all five seed types and peat treatments in the Glory Study in 2008 and 2009.

Treatment	Survival		
	2008	2009	Total
	----------------------	%	----------------------
Shelters	81 a*	64 a	72
No Shelters	63 b	60 a	62
Ave	72	62	67

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

Seeds planted with peat showed lower seed survival and establishment than seeds without peat (63% vs 81%, Table 3) during the first year. Survival was reduced an additional 10% regardless of peat treatment by the end of the second growing season. As previously reported (Skousen et al., 2009), we do not know the reason why peat treatment negatively affected seed germination during the first year.

Table 4. Chestnut seed survival with and without peat treatment across all five seed types and tree shelter treatments in the Glory Study in 2008 and 2009.

Treatment	Survival		
	2008	2009	Total
	----------------------	%	----------------------
Peat	63 b*	52 b	58
No Peat	81 a	72 a	77
Ave	72	62	67

*Peat treatment values within years with different letters are significantly different with an LSD test at p<0.05

Chinese seeds showed significantly higher establishment than the other seed types (Table 5). While four of the seed types had a 10 to 20% reduction in survival from the first to the second year, Chinese slightly increased from 82 to 86%.

Tree height was significantly greater on seeds with shelters compared to no shelters (Table 6), while peat treatment was not significant for height (Table 7). During the first year, Chinese seeds grew to significantly greater height than the other types (Table 8), and the hybrids were significantly greater than American. By the second year, the height differences among seed types had disappeared. We will continue to monitor survival and height growth of these seedlings during subsequent years.

Table 5. Chestnut seed survival for five seed types across shelter and peat treatments in the Glory Study in 2008 and 2009.

Seed Type	Survival		
	2008	2009	Ave
	------------------------ % ----------------		
American	67b*	55 b	61
Chinese	82a	86 a	84
B_1F_3	74b	53 b	63
B_2F_3	69b	53 b	61
B_3F_2	72b	62 b	67
Ave	72	62	67

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

Table 6. Chestnut seed height with and without tree shelters across all five seed types and peat treatments in the Glory Study in 2008 and 2009.

Treatment	Height		
	2008	2009	Total
	--------------------- cm ----------------------		
Shelters	27 a*	15 a	21
No Shelters	16 b	9 b	13
Ave	22	12	17

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

Table 7. Chestnut seed height with and without peat treatment across all five seed types and tree shelter treatments in the Glory Study in 2008 and 2009.

Treatment	Height		
	2008	2009	Total
	--------------------- cm ----------------------		
Peat	21 a*	11 a	16
No Peat	23 a	13 a	18
Ave	22	12	17

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

Table 8. Chestnut seed height for five seed types across shelter and peat treatments in the Glory Study.

Seed Type	Height		
	2008	2009	Ave
	---------------- cm ---------------		
American	18 c*	10 a	14
Chinese	26 a	13 a	20
B_1F_3	22 b	12 a	17
B_2F_3	20 b	13 a	16
B_3F_2	22 b	11 a	16
Ave	22	12	17

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

Nicholas Study

Soil chemical properties were significantly different for most parameters between brown and gray substrates (Table 9). Soil pH was much lower at 4.5 for the brown and much higher, 6.6, for the gray. In many cases with gray sandstone soil substitute in West Virginia, the pH is generally much higher at nearly 8.0 (Emerson et al., 2009), so the pH of this gray material is more optimum for tree growth. The almost 10-fold greater P in the gray vs the brown materials has also been documented in other studies. Significantly greater quantities of Ca and Mg are found in gray vs brown sandstone, which then gives much higher base saturation.

Table 9. Chemical properties in 2009 of the two substrate types in the Nicholas Study, where five chestnut seed and seedling types were planted into brown and gray sandstone plots in West Virginia.

Substrate	pH	P	K	Ca	Mg	CEC	BS
		mg kg^{-1}	--------------	$cmol^+ kg^{-1}$	----------------		%
Brown	4.5 b	6.0 b	0.33 a	2.90 b	3.60 b	13 b	28 b
Gray	6.6 a	56.1 a	0.40 a	9.50 a	6.20 a	8 a	100 a

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

A surprising finding during this study of seeds and seedlings during this first year was that only a handful of the 250 seeds planted on either substrate germinated and established. So few, in fact, that there was no apparent trend or reason why the six seeds germinated; four were on gray sandstone while the other two were in brown. These six germinated seeds were also not just one seed type; two were B_1F_3, two were Chinese, and one was B_2F_3, and the other B_3F_2. Therefore, no other information could be gathered about seed germination and establishment during this first year.

For seedlings, high survival of planted seedlings occurred for American (93%), Chinese (100%), and B_1F_3 (96%) chestnut types (Table 10). For B_2F_3, seedling survival was significantly lower on the gray sandstone substrate at 48% compared to almost twice the seedling survival on the brown (85%). B_3F_2 had similarly low survival on both substrates (67%). We found hardwood tree survival to be similar on brown and gray topsoil materials in other studies in West Virginia (Emerson et al., 2009; DeLong and Skousen, 2009), so this result is not surprising.

Table 10. Chestnut seed survival for five seed types in the Nicholas Study in 2009. Statistically significant differences were assessed using split plot ANOVA analysis (α=0.05).

Seed Type	Substrate		
	Brown	Gray	Ave
	---------------	%	---------------
American	93a	92a	93
Chinese	100a	100a	100
B_1F_3	96a	92a	96
B_2F_3	85a*	48c	67
B_3F_2	67b	68b	67
Ave	88	80	85

*Values between brown and gray are significantly different with LSD test at an alpha level of 0.05.

For height, Chinese seedlings were significantly greater in height than the other chestnut seedling types but these height differences were largely due to the initial differences in the size of the seedlings when planted (Table 11). During the second growing season and subsequently during later years of measurement, height differences will be due to growing conditions and growth media differences. We have seen in some of our other studies that tree growth is much greater on the brown sandstone materials vs the gray materials, even though survival might be very similar.

Table 11. Chestnut seed height for five seed types in the Nicholas Study in 2009. Statistically significant differences were assessed using split plot ANOVA analysis (α=0.05).

Seed Type	Substrate Brown	 Gray	 Ave
	--------------	**cm**	--------------
American	24b	21b	23
Chinese	40a	31a	36
B_1F_3	15c	15c	15
B_2F_3	12cd*	6d	9
B_3F_2	9 d	5 d	7
Ave	20	16	18

*Significant difference between Brown and Gray with LSD test at an alpha level of 0.05.

Overall, vigor ratings were similar between brown and gray substrate materials for American (2.8), Chinese (2.9), B_1F_3 (2.9), and B_3F_2 at (2.0). The B_2F_3 seedlings on the gray material were lower than on brown material. Again, these values are beginning values to which we can compare subsequent vigor ratings.

Table 12. Chestnut seed vigor for five seed types in the Nicholas Study in 2009. Statistically significant differences were assessed using split plot ANOVA analysis (α=0.05).

Seed Type	Substrate Brown	 Gray	 Ave
	-----------	rating	-----------
American	2.9a	2.7a	2.8
Chinese	2.9a	2.9a	2.9
B_1F_3	3.0a	2.8a	2.9
B_2F_3	2.5a*	1.4b	1.9
B_3F_2	2.1a	1.9a	2.0
Ave	2.7	2.3	2.5

*Values between brown and gray with different letters are significantly different with LSD test at an alpha level of 0.05.

Summary and Conclusions

In the Glory study, five chestnut seed types (American, Chinese, B_1F_3, B_2F_3, and B_3F_2 hybrids) were planted into a mixed brown sandstone substrate material in 8 blocks with and without peat and with and without tree shelters on a surface mine in southern West Virginia. The mixed brown sandstone soil material had a pH that varied across the blocks from pH 5.3 to 6.7, with the tree sheltered blocks being slightly more acidic than the non-sheltered blocks. Germination and survival after the first year was 72% across all treatments and survival dropped to 62% after the second year. By the second year, Chinese seeds had significantly higher survival at 86% compared to around 53 to 62% survival for American and hybrid seeds. Height of trees showed a similar pattern as that of survival. In the Nicholas Study, only six seeds (of 250 planted) germinated, which was quite surprising compared to the good success we had with seeds the previous year at the Glory site. Planted seedling survival was >90% with Chinese, American and the B_1F_3 hybrid on both brown and gray substrates. Overall, we have seen good establishment success on mined lands with both chestnut seeds in the Glory Study and chestnut seedlings in the Nicholas Study. Long-term establishment and survival will be evident as time passes and we continue monitoring these tree studies.

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References

Adank, K.M., C.D. Barton, M.E. French, and P.B. de Sa. 2008. Occurrence of Phytophthora on reforested loose-graded spoils in eastern Kentucky. Proceedings America Society of Mining and Reclamation, 2008 pp 1-13. http://dx.doi.org/10.21000/JASMR08010001.

Angel, P., J. Burger, V. Davis, C. Barton, M. Bower, S. Eggerud, and P. Rothman. 2009. The forestry reclamation approach and the measure of its success in Appalachia. Proceedings America Society of Mining and Reclamation, 2009 pp 18-36 http://dx.doi.org/10.21000/JASMR09010018

Burger, J., D. Graves, P. Angel, V. Davis, and C. Zipper. 2005. The forestry reclamation approach. US Office of Surface Mining, ARRI, Forest Reclamation Advisory No. 2.

Carpenter, S. 1992. Soil survey of Nicholas County, West Virginia. US Dept. of Agriculture and the WVU Agricultural Experiment Station, Morgantown, WV. 138 p.

DeLong, C., and J. Skousen. 2009. Amendments for reforestation on a reclaimed West Virginia surface mine. In: Abstracts, American Society of Agronomy, Nov. 1-5, 2009. Pittsburgh, PA.

Emerson, P., J. Skousen, and P. Ziemkiewicz. 2009. Survival and growth of hardwoods in brown versus gray sandstone on a surface mine in West Virginia. J. Environ. Qual. 38: 1821-1829. http://dx.doi.org/10.2134/jeq2008.0479.

French, M.E., C.D. Barton, D. Graves, P.N. Angel, and F.V. Hebard. 2007a. Evaluation of mine spoil suitability for the introduction of American chestnut hybrids in the Cumberland Plateau. Proceedings America Society of Mining and Reclamation, 2007 pp 249-258 http://dx.doi.org/10.21000/JASMR07010249

French, M.E., C.D. Barton, F.V. Hebard, D. Graves, S. Fei, and K. Adank. 2007b. Can surface mine reclamation help to restore American chestnut. p. 24-28. In: Reclamation Matters, Issue 2, Fall 2007. American Society of Mining and Reclamation, Lexington, KY.

McCarthy, B.C., J.M. Bauman, and C.H. Keiffer. 2008. Mine land reclamation strategies for the restoration of American chestnut. Ecological Rest. 26: 292-294. http://dx.doi.org/10.3368/er.26.4.292.

Phelps, T. 2002. Fifth generation orchard initiated at Penn State Arboretum. In: Chestnut Tree, Newsletter of the Pennsylvania Chapter of the American Chestnut Foundation. http://www.patacf.org/

SAS Institute. 2005. SAS user's guide. Cary, NC.

Skousen, J., T. Keene, C. DeLong, E. Pena-Yewtukhiw, and T. Cook. 2009b. Survival and growth of five chestnut seed types on mountaintop surface mines in West Virginia Proceedings America Society of Mining and Reclamation, 2009 pp 1276-1291
https://doi.org/10.21000/jasmr09011276

Wolf, B. 1994. Soil survey of Boone County, West Virginia. US Dept. of Agriculture and the WVU Agricultural Experiment Station, Morgantown, WV. 115 p.