

## Evaluation of *Alnus* Species and Hybrids

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### ABSTRACT

*Trials of a common set of seed lots representing 39 parents and five species of Alnus have been started in four countries: Belgium, Canada, the UK, and the US. Initial results indicate that cold hardiness is a problem in using A. acuminata but that sufficiently hardy A. rubra sources are available. A. glutinosa had the best growth in the nursery, and A. cordata had the best survival under severe moisture-stress conditions. A summary also is given of a workshop on alder improvement that further demonstrates the potential for developing the genus for biomass energy production.*

*Key words: Alnus acuminata, A. cordata, A. glutinosa, A. incana, A. rubra, seed source, cold hardiness, moisture stress tolerance.*

### INTRODUCTION

Depending on one's taxonomic viewpoint, the genus *Alnus* consists of  $20 + ^{1,2}$  or  $35^3$  species that are native to many parts of the northern hemisphere and to a limited area of Central and South America. The alders have been ranked behind such genera as *Populus* and *Salix* in their ability to produce rapid biomass growth in energy plantations.<sup>4</sup> However, the ability of alders to symbiotically fix nitrogen at high rates in

TABLE 1  
*Alnus* Entries for the 1987 IEA Evaluation Trial

Species	Identification code	Origin	Belgium <sup>a</sup>	Canada <sup>a</sup>	UK <sup>a</sup>	US <sup>a</sup>
<i>A. acuminata</i>	A.a. 1	Costa Rica	No	Yes	No	Yes
<i>A. cordata</i>	A.c. 48	La Retuzière, France	Yes	Yes	Yes	Yes
	A.c. 49	Beaucouzé, France	Yes	Yes	Yes	Yes
	A.c. 16	Avellino Campania, Italy	No	No	Yes	Yes
	A.c. 17	Potenza, Basilicata, Italy	No	No	Yes	Yes
	A.c. 52	Alta valle del Tevere, Italy	No	No	Yes	Yes
	A.c. 26	Prato, Corsica	No	Yes	Yes	Yes
	A.c. 73	Bocca di Pelza, Corsica	No	No	Yes	Yes
	A.c. 82	Barghiana, Corsica	No	Yes	Yes	Yes
<i>A. glutinosa</i>	A.g. 322	Sulechów, Poland	Yes	No	No	Yes
	A.g. 325	Sulechów, Poland	Yes	No	No	Yes
	A.g. 330	Sulechów, Poland	Yes	No	No	Yes
	A.g. 170	Podébrady, Poland	Yes	Yes	Yes	Yes
	A.g. 171	Podébrady, Poland	Yes	Yes	Yes	Yes
	A.g. 177	Podébrady, Poland	Yes	Yes	Yes	Yes
	A.g. 62	Seed Orchard Source DS-1, West Germany	Yes	Yes	Yes	Yes
	A.g. 66	Seed Orchard Source WA-18, West Germany	Yes	Yes	Yes	Yes
	A.g. 70	Seed Orchard Source ROTT-3, West Germany	Yes	Yes	Yes	Yes

A.g. 154	Ötvöskónyi, Hungary	Yes	Yes	Yes	Yes
A.g. 161	Nagkorpád, Hungary	Yes	Yes	Yes	Yes
A.g. 162	Homokszentgy, Hungary	Yes	Yes	Yes	Yes
A.g. 340	Durdevac, Yugoslavia	Yes	Yes	Yes	Yes
A.g. 341	Popovaca, Yugoslavia	Yes	Yes	Yes	Yes
A.g. 342	Durdevac, Yugoslavia	Yes	Yes	Yes	Yes
A.g. 223	Sperchios River, Greece	Yes	Yes	Yes	Yes
A.g. 224	Sperchios River, Greece	Yes	No	Yes	Yes
A.g. 225	Sperchios River, Greece	Yes	No	No	Yes
A.g. 234	Mesudiye, Ordu, Turkey	No	No	No	Yes
A.g. 236	Kesap, Giresum, Turkey	No	Yes	Yes	Yes
A.g. 239	Maçka, Trabzon, Turkey	No	Yes	Yes	Yes
A.i. 25	Försterei Hulske, Poland	Yes	Yes	Yes	Yes
A.i. 26	Dubrava Forest, Lithuania	Yes	Yes	Yes	Yes
A.i. 31	Loughgall, Co. Armagh, N. Ireland	Yes	No	No	Yes
A.r. 38	Auke Bay, Alaska, USA	No	No	No	Yes
A.r. 42	Eagle River, Alaska, USA	No	Yes	Yes	Yes
A.r. 45	Glacier Highway, Alaska, USA	No	Yes	Yes	Yes
A.r. 46	Sandpoint, Idaho, USA	Yes	No	No	Yes
A.r. 47	Sandpoint, Idaho, USA	Yes	No	No	Yes
A.r. 48	Sandpoint, Idaho, USA	Yes	Yes	Yes	Yes

<sup>a</sup>Included in evaluation trials.

soil has made them important trees for use in energy plantation systems in which frequent harvests will place a significant drain on site nutrient status. Hence, alders have been one of the genera focused on by the International Energy Agency (IEA) Forestry Energy Agreement. A directory of tree improvement programs in *Alnus* has been published,<sup>5</sup> a breeding strategy was proposed for working with the genus,<sup>6</sup> and an IEA joint evaluation trial including 30 entries of *Alnus* was established in 1985.<sup>7</sup> As the IEA Agreement entered its second stage in 1986, the following problems/objectives were identified: (1) a limited number of *Alnus* species and provenances had been evaluated in international trials; (2) the availability of selected seed sources of *Alnus* and especially for hybrids within the genus was limited; and (3) there was a need for better exchange of information among groups that were working on *Alnus* to promote the advantages the genus offers and to solve the problems it presents. Therefore, the IEA sponsored 3 years of effort to address these three areas. This report will deal primarily with the early results related to the first objective. The results from objectives (2) and (3) will only be summarized, because a separate proceedings is being devoted to them based on a workshop held in August 1988.<sup>8</sup>

## MATERIALS AND METHODS

### Alder seed lots, seedling production, and test designs

A total of 39 seed lots representing five species was chosen as the most interesting of materials available for testing (Table 1). To the extent possible, three single parent tree seed lots were used for each of several regions within the range of a species that might be expected to perform well in participating countries. Unfortunately, it was not possible to represent *A. incana* as adequately as we would have liked because of the unavailability of seed from desired locations. The choice of *A. cordata* seed lots was based on the recommendations of Steenackers in Belgium and Teissier du Cros in France, who have some experience with the species. The choice of *A. glutinosa* seed lots was based on the results of provenance tests with the species in North America.<sup>9</sup> It was known from a previous IEA test<sup>7</sup> that only interior and very northern provenances of *A. rubra* would survive continental winters, so those were the sources emphasized in this study. One seed lot of *A. acuminata* was included because it is a promising species on the basis of its taxonomic relationship to the other species and its growth in its native habitat.<sup>1</sup>

The *A. acuminata* seed came from the seed supplies of R. B. Hall; all other seed lots were supplied from the IEA alder seed bank maintained by V. Steenackers in Belgium. Identical seed lots were distributed to all four participating countries: Belgium, Canada, the UK, and the US.

Seeds were sown in small containers (approximately 350 cm<sup>3</sup> in soil volume) and grown under greenhouse conditions. In the Iowa trial, the seedlings were inoculated with a crushed nodule inoculum of local *Frankia* strains when they reached a stage at which they had several primary leaves.<sup>10</sup> When the seedlings reached a height of approximately 15 cm, they were hardened off and then planted in nursery beds (Belgium, Canada, UK) or directly in a field site (US). The nursery planting design used three replications with six to eight trees per replication. The field planting design was a randomized block design with five replications and four-tree row-plots of each seed lot in each replication. Observations were then taken on bud set, survival, and height growth. Trees were judged to have completed bud set when the terminal bud showed no evidence of the production of new leaves and the bud felt hard when pressed between one's fingers.

To better incorporate this international joint evaluation trial with the testing of other *Alnus* germplasm in national programs, a system of interlocking blocks (Fig. 1) was followed in the field planting established in the US. This interlocking design allows for direct comparisons between the different entries in two different studies.

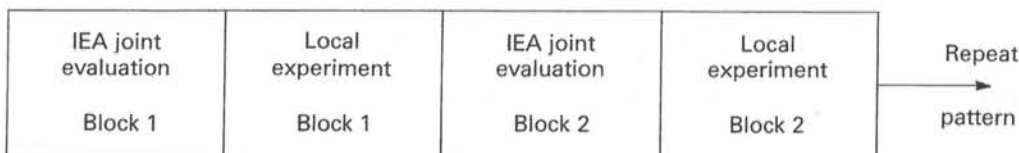


Fig. 1. An interlocking design for incorporating the IEA evaluation trial with national program trials of other *Alnus* germplasm sources.

## Workshop

The *Alnus* tree improvement workshop was held on 8 and 9 August 1988. It began in Tacoma, Washington, USA, after a general IEA meeting. The first day was a field trip that included an inspection of an *A. rubra* provenance test at Puyallup, Washington, a tour of a very efficient red alder sawmill operated by Northwest Hardwoods (Arlington, WA), and travel across an elevation transect of the natural range of *A. rubra* along the Stillaguamish and Skagit Rivers in Washington. The second

day was devoted to the presentation and discussion of seven papers on various aspects of genetic variation and applied improvement efforts in *Alnus*. The paper session was hosted by Dr D. T. Lester and the Faculty of Forestry at the University of British Columbia, Vancouver, Canada.

## RESULTS AND DISCUSSION

### Seedling production

Several seed lots exhibited much lower than expected germination and this resulted in insufficient seedlings for testing of all seed lots at each location (Table 1). In addition, the *A. acuminata* seed lot did not prove cold hardy and was quickly lost from most of the trials. Although all cooperators started with the same seed lots, unanticipated problems were encountered in the germination and early growth of several seed lots. A 'No' in Table 1 indicates that insufficient seedlings were produced for field testing of that seed lot at that location. Field tests were established in all four countries in 1987 and 1988, but data are available for only two of the test sites at this time.

### Fall dormancy in the United States

Table 2 gives the results of observations on bud set in all 39 seed lots at latitude 42°N at Ames, Iowa in the Fall of 1987. At that location, previous work has shown that the first week in October is a significant time for judging dormancy.<sup>11</sup> Killing frosts typically occur by 10 October. The percentage of seedlings that were dormant for each test entry ranged from 91% to zero. Only 20 of the 39 entries had 50% or more of their seedlings in a dormant condition. This is much less than expected from our previous field experiments in which a similar range of genetic materials had shown 80% of the first-year trees were dormant by 1 October. It seems that the onset of dormancy was delayed in these trees growing in containers in a hardening-off bed. Whether the relative rates and patterns of bud set between entries were the same as they would be under field conditions remains to be answered by correlation with a field test. Unexpectedly, one of the *A. rubra* entries from Idaho had the highest percentage of bud set. Indeed, all of the entries for *A. rubra*, except *A.r.* 48, set bud at a higher frequency than 50%. A general clinal trend of increasing bud set is shown in going from southern to northern entries of *A. glutinosa*. Entry *A.g.* 325 from Poland is a notable exception, displaying no bud set by 1 October. As a group, the *A. incana*

**TABLE 2**  
Relative Bud Set of *Alnus* Entries in the IEA Test in the Fall of 1987 at Ames, Iowa

Entry	Bud set <sup>a</sup> (%)	Entry	Bud set <sup>a</sup> (%)
<i>A.r.</i> 46	91	<i>A.i.</i> 31	48
<i>A.g.</i> 66	87	<i>A.g.</i> 341	45
<i>A.r.</i> 47	86	<i>A.g.</i> 234	34
<i>A.g.</i> 70	76	<i>A.g.</i> 225	33
<i>A.g.</i> 161	75	<i>A.g.</i> 330	33
<i>A.c.</i> 73	74	<i>A.g.</i> 171	31
<i>A.g.</i> 62	71	<i>A.g.</i> 236	31
<i>A.g.</i> 170	71	<i>A.i.</i> 25	30
<i>A.g.</i> 340	70	<i>A.r.</i> 48	19
<i>A.g.</i> 342	69	<i>A.c.</i> 48	16
<i>A.g.</i> 162	68	<i>A.g.</i> 223	15
<i>A.g.</i> 322	67	<i>A.c.</i> 52	13
<i>A.r.</i> 42	67	<i>A.g.</i> 224	13
<i>A.g.</i> 177	66	<i>A.c.</i> 16	10
<i>A.r.</i> 38	64	<i>A.c.</i> 17	10
<i>A.i.</i> 26	64	<i>A.c.</i> 49	09
<i>A.g.</i> 154	55	<i>A.c.</i> 26	06
<i>A.r.</i> 45	54	<i>A.a.</i> 1	00
<i>A.g.</i> 239	53	<i>A.g.</i> 325	00
<i>A.c.</i> 82	50		

<sup>a</sup>Percentage of trees with bud set on 1 October 1987.

entries were somewhat less advanced in developing dormancy than *A. glutinosa* and *A. rubra*. The *A. cordata* entries lagged still further behind in dormancy as might be expected because of the Mediterranean origin of the species. The *A. acuminata* entry showed no bud set and, indeed, was dead at the end of October when the containerized seedlings were moved to cold storage for the winter.

### Nursery performance in Canada

Table 3 summarizes the survival and height growth measurements made over two seasons of growth in a nursery bed at Petawawa, Ontario. The 1988 growing season was particularly severe, exceptionally hot and dry. Due to those conditions, there was additional mortality of 39 trees and growth averaged only about 5 cm. Typical growth under field conditions can be expected to be at least 1.0 m.<sup>9</sup> As a group, the *A. glutinosa* had relatively good survival and the best growth; *A. incana* had the best



**TABLE 3**  
Survival and Height of Alder Seedlings after the First and Second Growing Seasons in  
the Nursery at Petawawa, Ontario

1987 Results				1988 Results			
Entry	N	% Survival	Total height (cm) <sup>a</sup>	Entry	N	% Survival	Total height (cm) <sup>a</sup>
A.g. 70	18	100	52.5 a	A.g. 70	18	100	55.4 a
A.g. 223	18	100	51.3 ab	A.g. 223	18	100	55.1 a
A.g. 341	18	100	49.8 abc	A.g. 62	18	100	52.5 ab
A.g. 154	18	100	46.6 abcd	A.g. 340	13	72	52.3 ab
A.g. 62	18	100	43.5 abcde	A.g. 171	17	94	51.5 ab
A.g. 170	18	100	43.2 abcdef	A.g. 341	16	89	50.8 abc
A.g. 340	16	89	43.1 abcdef	A.g. 170	16	89	49.9 abcd
A.g. 171	18	100	40.1 bcdefg	A.i. 45	17	94	49.7 abcde
A.g. 161	18	100	40.0 bcdefgh	A.g. 154	18	100	49.2 abcde
A.g. 239	18	100	39.8 cdefgh	A.g. 66	15	83	48.6 abcde
A.r. 48	16	89	39.4 cdefgh	A.g. 161	18	100	46.6 abcde
A.r. 45	18	100	37.5 defgh	A.i. 25	18	100	45.9 abcde
A.i. 25	18	100	35.1 defgh	A.g. 239	16	89	43.5 abcdef
A.g. 66	18	100	35.1 defgh	A.g. 162	13	72	42.9 abcdef
A.g. 177	18	100	34.9 defgh	A.g. 171	12	67	42.4 bcdef
A.g. 162	18	100	32.6 efgh	A.r. 45	16	89	38.6 cdef
A.g. 236	18	100	32.4 efgh	A.r. 48	11	61	38.5 cdef
A.c. 48	18	100	32.2 efgh	A.g. 342	13	72	38.2 defg
A.g. 342	18	100	31.7 efgh	A.i. 26	18	100	38.0 defg
A.i. 26	18	100	31.5 fgh	A.g. 236	17	94	37.6 defg
A.r. 42	18	100	31.0 gh	A.r. 42	10	56	37.2 egh
A.c. 49	18	100	28.3 h	A.c. 48	17	94	31.8 fgh
A.c. 82	18	94	17.2 i	A.c. 49	18	67	30.6 gh
A.c. 26	18	100	15.8 i	A.c. 82	11	61	18.4 i
A.a. 1	0	0	—	A.c. 26	15	83	18.4 i

<sup>a</sup>Values followed by the same letter are not significantly different at the 5% level of significance.

survival and was second in growth; *A. rubra* still showed respectable survival (68.5%) after the second year. It is encouraging that these *A. rubra* entries seem to be much more winter hardy than the typical members of this species. The growth of *A. rubra* was relatively poor, however. The *A. cordata* entries in the test were also surviving as well as could be expected at the end of the second year, but their growth was the poorest of all the surviving species.

These early nursery results must be viewed with some caution. Our previous experience suggests that only a portion of the *A. rubra* and *A. cordata* entries will continue to be winter hardy when they grow above



the height of the protecting, winter snow cover. Furthermore, in a previous field study at Petawawa, *A. incana* outgrew *A. glutinosa* over the first 4 years.<sup>12</sup> In longer-term field studies in the Netherlands, both *A. cordata* and *A. incana* entries were able to outgrow *A. glutinosa*.<sup>13</sup> The one *A. acuminata* entry in this test did not survive at all under the growing conditions in the nursery at Petawawa. The failure of this species in all the tests is not surprising because the species originates from the tropics. However, it might be worthwhile to do additional trials with *A. acuminata* sources from high elevations in the tropics. Growth and form of the species are very good under greenhouse conditions and it would be useful to find some trees of the species that could be used in interspecific breeding.

### Field survival in the United States

After the study was field-planted in central Iowa, we experienced one of the worst growing-season droughts on record. Only 207 mm of rainfall was recorded at the planting site from the time of planting until the end of September 1988. Total precipitation for 1988 was 495 mm, compared with the long-term average of 816 mm for central Iowa. Consequently, extensive mortality occurred in the new plantation, as illustrated in Tables 4 and 5. To statistically analyze the mortality occurring in the four-tree plots of the field design, the data were transformed to account for the small sample size on each plot and the binomial nature of survival observations.<sup>14</sup> Two of the blocks in the plantation showed nearly complete mortality — eight trees survived of 288 planted. Those two blocks were planted 1 week later than the others and were on a Vesser silt loam soil type, as compared with a Colo silty clay loam soil type<sup>15</sup> for the blocks with better survival.

Considered as groups by species, the *A. cordata* entries survived the best, and the *A. rubra* had the poorest survival under these strong moisture-stress conditions. The hot, dry Mediterranean climate where the *A. cordata* species originates probably has selected for trees that have more drought tolerance. The thick, shiny leaves of *A. cordata* are one observable trait that probably is a part of this drought tolerance. In contrast, the cool, moist climates where *A. rubra* originates would not have been likely to select for much drought tolerance in that species. One of the entries, *A.r.* 48, from the continental interior does show a reasonable level of survival that might be worthwhile under more normal growing conditions.

With the predictions of global warming trends, it may be very important to place a premium on selecting heat- and drought-tolerant plants for our energy plantations. Both *A. cordata* and *A. glutinosa* show

**TABLE 4**  
First-Year Drought Hardiness of Individual Entries in Iowa During the 1988 Growing Season

<i>Entry</i>	<i>Number of 4-tree plots (observed)</i>	<i>Transformed % survival (mean)<sup>a</sup></i>
A.g. 70	3	77 a
A.c. 26	3	77 a
A.c. 73	3	70 ab
A.c. 48	3	70 ab
A.g. 322	3	70 ab
A.c. 49	3	66 abc
A.c. 52	3	62 abcd
A.g. 171	3	61 abcd
A.g. 223	2	57 abcde
A.c. 16	2	57 abcde
A.g. 162	3	57 abcde
A.c. 82	3	56 abcdef
A.c. 17	3	56 abcdef
A.g. 342	3	49 abcdefg
A.r. 48	1	45 abcdefgh
A.i. 26	3	45 abcdefgh
A.i. 25	3	45 abcdefgh
A.g. 234	3	45 abcdefgh
A.g. 224	3	41 bcdefgh
A.g. 154	3	41 bcdefgh
A.g. 62	3	38 bcdefgh
A.g. 161	3	38 bcdefgh
A.g. 177	3	34 cdefgh
A.g. 170	3	34 cdefgh
A.g. 341	3	34 cdefgh
A.g. 236	3	34 cdefgh
A.i. 31	3	34 cdefgh
A.g. 325	3	34 cdefgh
A.g. 330	3	33 defgh
A.r. 45	3	30 defgh
A.g. 225	3	30 defgh
A.g. 340	3	30 defgh
A.g. 239	3	26 efgh
A.r. 38	3	26 efgh
A.r. 47	2	23 fgh
A.r. 46	2	23 fgh
A.r. 42	3	20 gh
A.g. 66	3	13 h

<sup>a</sup>Values followed by the same letter are not significantly different at the 5% level of significance.

**TABLE 5**  
First-Year Drought Hardiness by Species in Iowa During the 1988 Growing Season

<i>Species</i>	<i>Number of plots</i>	<i>Transformed % survival (mean)<sup>a</sup></i>
<i>A. cordata</i>	23	65 a
<i>A. glutinosa</i>	62	42 b
<i>A. incana</i>	9	41 b
<i>A. rubra</i>	14	26 c

<sup>a</sup>Values followed by the same letter are not significantly different at the 5% level of significance.

some promising entries in this regard. The variation in survival for *A. glutinosa* does not follow the geographic trend that one might expect. Several entries of northern European origin survived the best; some, but not all, southern sources showed the poorest survival. As an upland species, *A. incana* might also be expected to have good drought tolerance, but the limited number of entries of that species in this study averaged no better than *A. glutinosa*.

#### Summary of workshop papers<sup>8</sup>

L. Bouvarel and E. Tessier du Cros of the French INRA are testing 158 provenances of *A. glutinosa*, 7 *A. incana*, 45 *A. cordata*, 81 *A. rubra*, and 1 each of *A. hirsuta*, *A. inokumai*, and *A. rhombifolia*. Results to date include the following observations on growth:

**TABLE 6**

	<i>Dry weight yield</i>	<i>Height</i>
<i>A. glutinosa</i>	0.8–2.6 Mg ha <sup>-1</sup> year <sup>-1</sup>	5–6.4 m at 6 years
<i>A. cordata</i>	2–7 Mg ha <sup>-1</sup> year <sup>-1</sup>	6–7.8 m at 6 years
<i>A. rubra</i>	1–2.6 Mg ha <sup>-1</sup> year <sup>-1</sup>	3–5 m at 4 years

Some *A. rubra* have survived –20°C temperatures.

According to a provenance test conducted by A. Ager of the University of Washington, genetic variation in *A. rubra* is predictable by river drainage and elevation. Growth and dormancy patterns relate closely to heat sums and frost dates at origins, but 'cue' changes may be

large in moving trees long distances. Most of the variation in *A. rubra* is due to geographic origin with only small parent tree effects. The species has very little variation in specific gravity. Sources from poor sites with gravel soils allocate more growth to roots and nodules. However, the best means of selecting for improved nitrogen fixation rates is to select for aboveground biomass production. There is a close correlation between the results for *A. rubra* grown in Washington and France.

R. B. Hall reported that fast-growing, early-flowering hybrids of *A. glutinosa* have been produced through the  $F_2$  generation. In addition, *A. incana*  $\times$  *glutinosa* and *A. glutinosa*  $\times$  *rubra* hybrids have been mass-produced in provenance test plantings. The *A. glutinosa*  $\times$  *rubra* hybrids show hybrid vigor for growth under greenhouse conditions. These hybrids are also much easier to vegetatively propagate than *A. glutinosa*. In joint entomological studies with E. R. Hart (Iowa State University), Hall has found no satisfactory levels of resistance to the European alder leafminer, *Fenusa dohrnii*, in *A. glutinosa*. *A. incana* is moderately resistant. *A. cordata* and *A. rubra* are essentially immune to the insect. *A. glutinosa*  $\times$  *rubra* hybrids show a range of resistance spanning the behavior of the two parental species.

R. N. Nyong'o (Iowa State University) has studied pollen dispersal in *A. glutinosa* and has found that the pollen travels farther than experience with other species would have suggested. At least a 400-m isolation strip is needed around seed orchards to exclude at least 91% of the alder pollen from non-orchard sources.

## CONCLUSION

The alder evaluation trials are just entering the establishment phase, but they do highlight some important points. We need to continue to improve the techniques for growing alder seedlings and getting good field establishment. The relatively poor tolerance of *Alnus* as a genus to moisture stress will be a limitation to its use in biomass plantations, but we probably can improve drought hardiness by working with species like *A. cordata*, by making selections within other species, and possibly through species hybridization. Improvement in biomass production will depend on similar procedures.

Three serious biological limitations in the present study need to be corrected in the future: (1) we need more *A. incana* germplasm from the central and southern portion of the species range to get a legitimate evaluation of its potential in biomass systems; (2) the value of hybrids needs much more study — none was available in sufficient quantity to

include in this joint evaluation at the time it started; and (3) we need to integrate the testing of alder species and hybrids with the evaluation of pure culture isolates of *Frankia* as they become available.<sup>12,16</sup>

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