SELECTION AND RELEASE OF INDIGENOUS PLANT MATERIALS FOR THE ANACONDA SMELTER SUPERFUND SITE - 'OPPORTUNITY' GERMPLASM NEVADA BLUEGRASS[1]

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Abstract: The Development of Acid/Heavy Metal Tolerant Releases (DATR) project [formerly the Development of Acid/Heavy Metal Tolerant Cultivars (DATC) project], was initiated in 1995 to address a critical need for native plants adapted to the edaphic and climatic conditions found in the Anaconda – Butte area. The goal of the project was to select plant materials demonstrating superior tolerance of low pH and/or heavy metal contaminated soils that are also adapted to the severe environmental conditions characterized by the intermountain foothills and mountains of Montana and Wyoming. Whereas most seeds and plants used for mine land reclamation have been cultivar releases from universities, USDA Plant Materials Centers, USDA Agricultural Research Service, or private plant breeders, pre-varietal release is a streamlined process encompassing a series of release classes determined by incremental levels of specified testing. The time interval to release and commercial availability of selected material can be reduced by as much as half by using pre-varietal release versus the traditional cultivar.

To date, the DATR project has assembled 130 seed collections of 72 native species of grasses, forbs, shrubs, and trees within the Upper Clark Fork River Basin (UCFRB). These collections have been planted at various study sites in comparison with non-local native and introduced plant species. Five species indigenous to the UCFRB have demonstrated superior survivability and vigor on amended acidic, heavy metal-contaminated soils at varying elevations in western Montana, and have been released through the DATR project using the Pre-Varietal release mechanism.

Opportunity Germplasm Nevada bluegrass is one of three selections made from indigenous material collected near Anaconda, Montana and tested at Stucky Ridge. In 2006, this selection proved significantly (ANOVA; Tukey; p=0.05) superior to four other seed sources (2 indigenous; 2 cultivars) for percentage stand cover, vigor rating, and biomass production on a lime and fertilizer amended site.

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Introduction

In 1983, an approximate 777-square kilometer (300-square mile) area in southern Deer Lodge valley and the surrounding foothills was designated the Anaconda Smelter NPL Site under the 1980 Comprehensive Environmental Response Compensation and Liability Act. In the Clark Fork River Basin alone, there are more than 453 square kilometers (175 square miles) of soils and vegetation contaminated by emission fallout from smelting operations. Low pH in the surface soils, as well as elevated levels of heavy metals including As, Cd, Cu, Pb and Zn, have impacted plant communities, leaving the region either barren or sparcely vegetated.

Since the extent of the injured area is so large, problems inherent to the reclamation of the site must be addressed while realizing resource limitations and cost effectiveness. The level of intervention planned as part of the restoration effort must succeed in establishing a self-sustaining community of plant species capable of stabilizing the soil, maximizing water usage, providing wildlife habitat and accelerating successional processes.

Techniques for selecting metal and acid tolerant vegetation was developed in the 1970's in the United Kingdom to stabilize acidic mine wastes. British researchers found that plants collected from mine sites possessed tolerance to the metals present in their native substrate (Marty October 2003).

To address the need for adapted native plants, the Development of Acid/Heavy Metal-Tolerant Cultivars (DATC), now referred to as the Development of Acid/Heavy Metal Tolerant Releases (DATR) project, was initiated in 1995. The DATR project is currently funded by the Montana Department of Justice – Natural Resource Damage Program**,** awarded to the Deer Lodge Valley Conservation District (DLVCD), in cooperation with the USDA-NRCS Bridger Plant Materials Center (PMC), a 57-hectare (140-acre) research facility dedicated to the selection and release of native plant materials, primarily for use in Montana and Wyoming.

Opportunity Germplasm Nevada bluegrass, released to the commercial seed industry in 2007, is an excellent example of a local ecotype that was tested as part of the DATR project, and shows great promise for use in mineland restoration. As a seed source found growing naturally in the northern Rocky Mountains, it is assumed this selection will perform well in other mountainous regions of the Intermountain West with similar environments and climates.

Role of the NRCS Plant Materials Center – Bridger, Montana

The PMC at Bridger, Montana, has been testing, selecting, and releasing superior seed sources since 1959. Technologies for their propagation, production, and establishment are also developed. The process begins with identification of a priority conservation problem. A list of potential plant species that may be used to solve this problem is generated, and is used as the basis for a seed collection list. In a typical scenario, seeds from multiple locations are collected for 2 to 4 years and then sown at the PMC, or at other locations in an Initial Evaluation Planting (IEP). In some cases, if inadequate seed is not available, the establishment of small seed production fields may be necessary to generate enough seed for testing – a step that may delay IEP establishment for 2 or more years. For grass and forb species, data is collected on critical performance criteria (plant height, percentage stand cover, vigor, and biomass) for 2 to 5 years and then selections of superior seed sources made. The selection may or may not be tested further in field trials to determine range of adaptation. Large scale field increase then occurs at the PMC so that adequate seed is available on-hand for testing and release to the commercial seed production market. Extensive documentation of plant performance is used to support official release of the selection to growers, including an evaluation of the potential invasiveness of the plant.

The national network of 27 Plant Materials Centers is the primary source of native plants developed specifically for reclamation and conservation use. To date, the DATR project has resulted in more than 130 seed collections of native grasses, forbs, shrubs, and trees from within the Upper Clark Fork River Basin and abandoned mine sites throughout western Montana. These collections have been planted at various study sites in comparison with nonlocal native and introduced plant species. The Atlantic Richfield Company (ARCO) has provided land access for seed collection and sites for experimental plots.

The Conservation Problem at Anaconda

One of the most impacted areas is the ~47 km^2 (~18 mi^2) of uplands within the Anaconda Smelter Superfund Site. The uplands are commonly derived from the weathering of bedrock and are typically thin, clay-rich alfisols. Soils are characterized by elevated heavy metal concentrations and low pH. Prior to treatment three metals, As, Cu, and Zn were detected at concentrations in soils that exceeded published phytotoxicity levels for plants (see Table 1).

Without an established plant community, these soils are susceptible to erosion by wind and water. Original vegetation in the uplands consisted primarily of shrub lands with coniferous forests above approximately 1,768 meters (5,800 feet). Annual precipitation at the site ranges from 254 to 356 mm (10 to 14 inches), with most of the precipitation occurring in the spring. In an effort to stem the transport of contaminants and restore these injured areas, state and federal regulatory agencies have developed several reclamation alternatives, many of which include the planting of shrubs and trees in the uplands (Marty, October 2003).

Pre-Varietal Release Mechanism

Historically, the seed and plants available to reclamationists are usually cultivar (cultivated varietal) releases from universities, USDA-NRCS (PMCs), USDA-Agricultural Research Service, or private plant breeders. To be released as a cultivar, the germplasm must be extensively tested, reviewing primary traits through multiple generations, and field testing to determine range of adaptability. The process takes at least 10 years with herbaceous plant material and can take 20 or more years for woody plants. Other sources of native plants are wildland collections and pre-damage plant salvage.

In recent years, the demand for native, indigenous plant material has resulted in the development of an alternate, more expedient mechanism for the release of plant materials known as Pre-Varietal Release. Through this process, plant propagules can be released to the commercial seed and nursery industries more expeditiously, but at the expense of extensive field testing. Germplasm identity and quality is still assured via seed certification agencies, maintaining the same quality, purity, and germination standards of Cultivar releases. There are three categories of Pre-Varietal releases:

Source-Identified

With the Source-Identified classification, a collector can locate and collect seed from a specific site, and then have the seed certified as to source (location) alone. A representative from a seed certification agency must inspect the collection site prior to harvest, documenting the identity of the species, elevation, latitude/longitude, and associated species. The collector now has the ability to guarantee the origin of the seed, assure it to quality standards, and can sell the seed directly to a customer. The collector can also take that seed and establish seed production fields, producing up to two generations beyond the original collection. The subsequent product

must also be included in a seed certification program to qualify as 'Source Identified' germplasm. Through this process, seed can be certified the year of collection or in 2 or more years later when the increase fields begin to produce seed. For the DATR project, staff elected to establish stands of Source-Identified releases at the PMC, an additional step taken to guarantee germplasm preservation (wildland stands can be lost through a variety of causes) and availability to the commercial market.

Selected

The Selected category is for plant breeders who assemble and evaluate multiple collections of a species, making a selection of the superior accession, or bulking or cross-pollinating superior accessions. The release process can take as few as 5 years, but can claim only one accession or bulk of accessions has been found to be superior for the conditions under which it was tested. No field testing or the testing of progeny is required.

Tested

The Tested category is the highest class in the Pre-Varietal Release process. If the progeny of a superior germplasm is tested to make sure the desired traits continue to manifest themselves in subsequent generations, the germplasm qualifies to be released as a Tested germplasm. The process can take 6-8 years for herbaceous plant material and considerably longer for woody plants. The only difference between Tested and Cultivar releases is the extensive field testing required of a Cultivar (Majerus, 2007).

To date, there have been five official germplasm releases by the DATR project: Washoe Germplasm Selected Class basin wildrye (*Leymus cinereus*), Old Works Germplasm Source-Identified Class fuzzytongue penstemon (*Penstemon eriantherus*), Prospectors Germplasm Selected Class common snowberry (*Symphoricarpos albus*), Copperhead Germplasm Selected Class slender wheatgrass (*Elymus trachycaulus*), and Opportunity Germplasm Selected Class Nevada bluegrass (*Poa secunda* ssp. *Nevadensis).*

Opportunity Germplasm Nevada Bluegrass

Opportunity Germplasm Selected Class Nevada bluegrass (NRCS accession number 9081633) is an example of the testing, selection, and pre-varietal release process for the DATR project (Fig. 1).

Figure 1. Opportunity Germplasm Nevada Bluegrass at PMC

Collection Site Information:

The original seed collection was made in 1998 near the Wisdom Junction along Highway 1, 5 km (3.1 miles) east of Anaconda, Montana (Section 5 Township 4N Range 10W) Seed was collected from at least 10 plants. The site was severely impacted by smelter fallout and surface wind and water transported contaminants, as well as historic overflow from the canal transporting waste material to the Opportunity Sediment Ponds. The area exhibited low pH and the presence of heavy metals consistent with conditions throughout the smelter fallout area. This bluegrass was found growing in association with slender wheatgrass *Elymus trachycaulus,* redtop *Agrostis gigantea,* scarlet globemallow *Sphaeralcea coccinea*, and western wheatgrass

Pascopyrum smithii. Reshaping and replanting of the original site has occurred since the collection was made, resulting in the destruction of the original population.

Description

The present nomenclature for Sandberg bluegrass *Poa secunda* combines several previously distinct species; both the *P. sandbergii/P. canbyii* type (short stature, early maturing, primarily basal leaves) and the *P. ampla/P. juncifolia/P. nevadensis* type (taller stature, late maturing, primarily cauline leaves). Although under the present nomenclature Opportunity Germplasm Nevada bluegrass is classified as *P. secunda*, it has the same general botanical (floral, foliage, and seed) and phenological attributes as the species previously classified as *P. nevadensis*. Opportunity Germplasm Nevada bluegrass is late maturing (unlike Sandberg and Canby bluegrass), has numerous basal and folded leaves (unlike big bluegrass type *P. ampla)*, and long ligules (unlike the alkali bluegrass type *P. juncifolia)*. This germplasm release has been given the common name of Nevada bluegrass to distinguish it from Sandberg bluegrass. In the northern Great Plains and northern Rocky Mountain foothills and valleys the Nevada bluegrass type is more robust and later maturing than the Sandberg bluegrass type and is therefore a more desirable reclamation species.

It is assumed 'Opportunity' traits are heritable and the progeny from the selection will appear and perform in a similar manner. Nevada bluegrass is a long-lived perennial bunchgrass. The donor plants of this selection reached 61 to 122 cm (2 to 4 feet) in height by early July. Opportunity Germplasm Nevada bluegrass reached a mean plant height of 62.4 cm (24.5 inches) on the pH adjusted test site (see Table 2) in a 355-millimeter (14-inch) precipitation zone four years after planting. Nevada bluegrass is a medium-stature, cool season grass with basal leaves reaching 15.2 to 30.5 cm (6 to 12 inches) in length and cauline leaves about half that length. Nevada bluegrass leaves are smooth, deep blue-green and folded with keel-shaped tips typical of bluegrasses. The species inflorescence is a narrow panicle up to 20.3 cm (8 inches) long.

Method of Selection

Opportunity Germplasm Nevada bluegrass was released as a 'Natural–Track' germplasm, that is, it was selected with no intent to genetically manipulate the population, ie. no genetic engineering or restricted/controlled pollination. This accession was compared to two other collections from acid/heavy-metal impacted sites (9081635)—Stucky Ridge north of Anaconda and 9081322—from Marysville, Montana, and two released cultivars; 'Canbar' (Washington),

and ‘Sherman’ (Oregon). The five accessions are all listed as *Poa secunda* but they were originally described as different species. Opportunity and ‘Sherman’ were originally *Poa ampla*, 9081635 and ‘Canbar’ were originally *Poa canbyii*, and 9081322 was originally *Poa sandbergii*. This material has been field tested at three impacted sites (two deep plowed, one deep plowed and amended) in the Anaconda area, as well as in a controlled greenhouse environment utilizing contaminated soil. This selection of bluegrass exhibited superior emergence, survival, and biomass production on amended acid/heavy-metal impacted soil under the ambient climatic condition of the Upper Clark Fork Watershed (Deer Lodge County, Montana).

Materials and Methods

The test site occupies 0.61 hectares (1.5 acres) and is located on Stucky Ridge, approximately 3.2 km (2 miles) northeast of Anaconda, Montana, in Deer Lodge County. The study plot is known as the Stucky Ridge Comparative Evaluation Planting (CEP) and is situated on a stream terrace above Lost Creek at an elevation of 1,618 meters (5,308 feet) above sea level, and is sited on relatively flat ground on the east end of Stucky Ridge. Sparse vegetation cover includes scattered groves of quaking aspen (*Populus tremuloides)*, shrublands dominated by Wood’s rose (*Rosa woodsii)*, currant (*Ribes* species), rubber rabbitbrush (*Chamerion nauseosa)*, horsebrush (*Tetradymia canescens)*; and grasslands dominated by redtop (*Agrostis gigantean)* and basin wildrye (*Leymus cinereus)*. Annual precipitation at the site ranges from 254 to 356 mm (10 to 14 inches)**,** with most of the precipitation occurring in the spring. The parent material is alluvium. The soil has a gravelly loam texture and is well drained. Slope at the plot site averages approximately 5 to 10 percent. Contamination identified in the Stucky Ridge RDU includes elevated As concentrations in surface soils, barren or sparsely vegetated areas due to low pH and elevated contaminant concentrations, and steep slopes with high erosion potentials (ARCO 2002, May) (Table 1). Current and historic use of the area consists primarily of agricultural grazing, recreational, open space, and wildlife habitat.

Table 1. Pre-treatment soils analyses, Stucky Ridge CEP area, Anaconda, MT (ARCO 2002).

Soil Sample Station	Depth	As	Cu	Zn	Sat. Paste pH
	inches	*mg/kg*	*mg/kg*	*mg/kg*	*s.u.*
94S-SR-71	0-2	-----	-----	-----	4.70
94S-SR-71	2-8	-----	-----	-----	4.90
94S-SR-73	0-2	-----	-----	-----	4.30
94S-SR-73	2-8	-----	-----	-----	4.60
99-098A	0-2	495.0	1660.0	419.0	-----
99-098B	2-6	163.0	1320.0	276.0	-----
99-098C	0-6	-----	-----	-----	4.20
99-098D	6-12	-----	-----	-----	7.60
99-098E	12-18	-----	-----	-----	7.80
99-099A	0-2	489.0	1370.0	303.0	-----
99-099B	2-6	95.8	1020.0	245.0	-----
99-099C	0-6	-----	-----	-----	4.00
99-099D	6-12	-----	-----	-----	7.30
99-099E	12-18	-----	-----	-----	7.70
99-123A	0-2	656.0	1530.0	425.0	-----
99-123B	2-6	167.0	1530.0	332.0	-----
99-123C	0-6	-----	-----	-----	4.40
99-123D	6-12	-----	-----	-----	4.80
99-123E	12-18	-----	-----	-----	6.30
99-163A	0-2	537.0	2180.0	493.0	-----
99-163B	2-6	256.0	1430.0	365.0	-----
99-163C	0-6	-----	-----	-----	4.00
99-163D	6-12	-----	-----	-----	6.20
99-163E	12-18	-----	-----	-----	-----
Phytotoxicity Range+		136-315	236-750	196-240	<5

+ Phytotoxicity ranges as stated in CDM Federal, 1997, "Final Baseline Ecological Risk Assessment"

The entire Stucky Ridge CEP was graded then tilled to a 30.5-cm (12-inch) depth in mid-September 2002, followed by amendment with approximately 20 metric tons per hectare (~22 tons per acre) of lime. The lime was incorporated into the soil by disking (four passes) the surface 30 cm (12 inches) of the soil profile in November 2002. Pre- and post-treatment soil analyses, as well as phytotoxicity standards for pH, As, Cd, Cu, Pb, and Zn appear in Table 2. Although soil contaminant level and pH varied by depth and location pre-treatment soil analyses suggested potentially phytotoxic levels of As, Cu, and Zn, as well as pH levels unacceptable for growth. Post-treatment soil analyses suggest generally acceptable plant growth levels of As, Cd, and Pb, as well as acceptable soil pH values. Copper levels, however, were potentially phytotoxic at all sample locations. Soil Zn levels ranged from acceptable to potentially phytotoxic, depending on sample location.

Table 2. Summary mean and composite pre- and post-treatment soil analyses and phytotoxicity standards for the Stucky Ridge CEP, Anaconda, MT.

Mean Values	pH	As	Cd	Cu	Pb	Zn
		mg/kg	*mg/kg*	*mg/kg*	*mg/kg*	*mg/kg*
Pre-treatment (0 - 2 in)	4.5	544	NA	1685	NA	410
Pre-treatment (2 - 6 in)	4.4	170	NA	1325	NA	305
Pre-treatment (0 - 6 composite)	4.4	357	NA	1505	NA	358
Post-treatment (0 - 6 composite)	7.8	145	1.3	832	43	194
Phytotoxicity Range	<5	136-315	5.1-20.0	236-750	94-250	196-240

In April 2003, fertilizer was applied and incorporated at a rate of 67 kilograms per hectare (60 pounds per acre) nitrogen, 90 kilograms per hectare (80 pounds per acre) P_2O_5, and 280 kilograms per hectare (250 pounds per acre) K_2O. In May 2003, the test planting was installed comparing the performance of 36 accessions of grasses (11 species), 16 accessions of forbs (11 species), and four grass/forb mixtures. The CEP included three collections of indigenous and two cultivated releases of bluegrass. The planting was installed with a four-row

cone-seeder equipped with double-disk openers and adjustable depth bands. Eight, 7.6-meter (25-foot) long rows of each accession were planted 12.7 millimeters (0.5 inches) deep, with 30.5-centimeter (1-foot) space between rows.

Seedling density was the only growth response variable used to assess performance during the first growing season (2003). Data was collected from within a 30- x 50-cm (11.8- x 19.7-inch) sampling frame randomly placed at five sample locations within each 2.4- x 7.6-meter (8- x 25-foot) treatment block. Sampling frames were located along row numbers 2 and 3, as well as rows 6 and 7 to prevent edge-effect error. The sampling frame was situated with its long axis perpendicular to the seeded rows so that each sampling measurement included two rows. Only seedlings rooted within the sampling frame were counted and used to calculate seedling density. Seeded and non-seeded seedlings (of the same accession) were counted and recorded separately. Seedling density data was collected in June 2003, in order to assess emergence and initial establishment, and then again in August to determine subsequent establishment and survival.

Test plots were evaluated in mid- to late-summer or fall each year from 2004 through 2006. A 30- x 50-cm (11.8- x 19.7-inch) sampling frame was randomly placed in four locations within each test plot as previously described. For summer evaluations, plants rooted within the sampling frame were evaluated for percentage stand cover (visual estimate), plant vigor rating, and mean plant height. Vigor rating is a subjective, visual assessment of the overall health and performance of the plant relative to other plants of the same accession. For fall evaluations, the same criteria were measured, as well as sampling for total biomass production. If the total weight of the four biomass samples did not yield approximately 10 grams (0.35 ounces) of material, additional plant tissue was sampled to assure enough biomass for tissue analysis. All biomass samples were oven dried at 60^0C (140^0F) for 24 hours and then weighed. Grab samples were taken from each sample, processed into small pieces, packaged in plastic zip-lock bags, and then delivered to an analytical laboratory for heavy metal analysis. All tissue samples were unwashed, so the heavy-metal results represent a combination of intercellular and surface tissue accumulations.

Plots were evaluated once per year, late summer/early fall, with the exception of the first two years of establishment, where evaluations were conducted twice during the growing season.

Results

2003

Seedling density data for all *Poa secunda* accessions tested at the Stucky Ridge CEP in June 2003 appear in Table 3. Opportunity Germplasm Nevada bluegrass (9081633) had significantly greater seedling density (7.08 seedlings per square foot) than all other *Poa secunda* accessions evaluated in June. By comparison, the grand mean density of all grass species tested in the Stucky Ridge CEP was 58.1 seedlings per square meter, ranging from 160.11 ('Pryor' slender wheatgrass) to 3.66 ('Gruening' alpine bluegrass) seedlings per square meter. Different letters in the separation column signify a statistically significant difference in density at $p<0.05$)

Table 3. Seedling density of *Poa secunda* accessions, Stucky Ridge CEP, June 24, 2003, Anaconda, MT.

	Accession Number		Seedling	Mean
Species	Or Cultivar	Test ID	Density	Separation
			seedlings/m^2	
Poa secunda	**9081633 (Opportunity)**	29	76.18	def
Poa secunda (*P. ampla*)	'Sherman'	30	33.46	hijk
Poa secunda	9081635	32	20.01	ijk
Poa secunda	9081322	33	14.1	ijk
Poa secunda	'Canbar'	31	13.02	ijk

By the August 2003 evaluation, seedling density for Opportunity Germplasm declined to 55.09 seedlings per square meter, but was still significantly higher than all other *Poa secunda* accessions tested on that date. The grand mean seedling density of all grasses on the August 25 evaluation was 46.27 seedlings per square meter, ranging from 154.73 (slender wheatgrass 9081620) to 3.01 (Indian ricegrass 9081629) seedlings per square meter.

2004

Percentage stand cover, vigor rating, and mean plant height data for all *Poa secunda* accessions tested at the Stucky Ridge CEP in June 2004 appears in Table 4 Opportunity Germplasm Nevada bluegrass had significantly higher percentage stand cover (31.3 percent) than

all other *Poa secunda* accessions tested. Similarly, Opportunity Germplasm had a vigor rating (3.3) as good, or better, than all other *Poa secunda* accessions tested, and significantly greater mean plant height (26.3 centimeters) than all other *Poa secunda* accessions tested.

Table 4. Percentage stand cover, vigor rating, and mean plant height of *Poa secunda* accessions, Stucky Ridge CEP, June 30, 2004, Anaconda, MT.

	Accession Number	Percentage	Vigor	Mean Plant
Species	Or Cultivar	Stand Cover [1]	Rating	Height
		%	(1-9)[2]	*cm*
Poa secunda	**9081633 (Opportunity)**	31.3 b	3.3	26.3 bcd
Poa secunda	9081635	15.0 efghij	3.3	23.3 cde
Poa secunda (*P. ampla*)	'Sherman'	6.2 ijklm	4.8	16.5 efghi
Poa secunda	9081322	3.6 klm	4.3	17.6 defgh
Poa secunda	'Canbar'	0.4 m	6.2	6.3 klmn

[1] – Percentage stand cover indicates the amount of cover within the seeded row, not the entire plot or stand.
[2] – A vigor rating of "1" is best, "4" is average, and "9" is worst or near death.

Percentage stand cover, and vigor rating data for all *Poa secunda* accessions tested at the Stucky Ridge CEP was evaluated in September 2004. Opportunity Germplasm Nevada bluegrass had significantly higher percentage stand cover (37.2 percent) than all other *Poa secunda* accessions tested. Similarly, Opportunity Germplasm had the best vigor rating (2.4) of all *Poa secunda* accessions tested.

Biomass production data for all *Poa secunda* accessions tested at the Stucky Ridge CEP in September 2004 appears in Table 5. Opportunity Germplasm Nevada bluegrass had significantly greater biomass production (408 kilograms per hectare; 364 pounds per acre) than all other *Poa secunda* accessions tested.

2005

Percentage stand cover, vigor rating, and mean plant height data for all *Poa secunda* accessions tested at the Stucky Ridge CEP in August 2005 appears in Table 6. Opportunity Germplasm Nevada bluegrass had significantly higher percentage stand cover (43.4 percent) than all other *Poa secunda* accessions tested. Similarly, Opportunity Germplasm had the best vigor rating (2.1) of all *Poa secunda* accessions tested, and the greatest mean plant height (59.1 cm; 23.0 inches), although not significantly greater than accessions 9081635 or 9081322.

Table 5. Biomass production of *Poa secunda*, Stucky Ridge CEP, September 22, 2004, Anaconda, MT.

	Accession Number	Biomass
Species	Or Cultivar	Production
		kg/ha
Poa secunda	**9081633 (Opportunity)**	408 bcd
Poa secunda	9081635	216 cdef
Poa secunda (*P. ampla*)	'Sherman'	183 def
Poa secunda	9081322	24 f
Poa secunda	'Canbar'	0 f

Table 6. Percentage stand cover, vigor rating, and mean plant height of *Poa secunda* accessions, Stucky Ridge CEP, August 30, 2005, Anaconda, MT.

	Accession Number	Percentage	Vigor	Mean Plant
Species	Or Cultivar	Stand Cover [1]	Rating	Height
		%	(1-9) [2]	*cm*
Poa secunda	**9081633 (Opportunity)**	43.4 b	2.1	59.1 b
Poa secunda	9081635	25.9 cd	3.3	45.6 bcde
Poa secunda (*P. ampla*)	'Sherman'	12.5 efghijkl	4.0	34.9 cdefghi
Poa secunda	9081322	10.0 ghijklm	4.2	45.4 bcdef
Poa secunda	'Canbar'	0.1 m	8.0	0 m

[1] – Percentage stand cover indicates the amount of cover within the seeded row, not the entire plot or stand.

[2] – A vigor rating of "1" is best, "4" is average, and "9" is worst or near death.

Biomass production data for all *Poa secunda* accessions tested at the Stucky Ridge CEP on August 2005 was analyzed. Opportunity Germplasm Nevada bluegrass had significantly greater biomass production (2,506 kilograms per hectare; 2,235 pounds per acre) than all other *Poa secunda* accessions tested.

2006

Percentage stand cover, vigor rating, and mean plant height data for all *Poa secunda* accessions tested at the Stucky Ridge CEP in August 2006 appears in Table 7. Opportunity Germplasm Nevada bluegrass had significantly higher percentage stand cover (63.1 percent) than all other *Poa secunda* accessions tested. Similarly, Opportunity Germplasm had the best vigor rating (2.9) of all *Poa secunda* accessions tested, although mean plant height (62.4 centimeters; 24.5 inches) was not significantly greater than 'Sherman' or 'Canbar'.

Biomass production data for all *Poa secunda* accessions tested at the Stucky Ridge CEP in August 2006 appears in Table 8. Opportunity Germplasm Nevada bluegrass had significantly greater biomass production (2,311 kilograms per hectare; 2,061 pounds per acre) than all other *Poa secunda* accessions tested.

Table 7. Percentage stand cover, vigor rating, and mean plant height of *Poa secunda* accessions, Stucky Ridge CEP, August 29, 2006, Anaconda, MT.

	Accession Number	Percentage	Vigor	Mean Plant
Species	Or Cultivar	Stand Cover [1]	Rating	Height
		%	(1-9) [2]	*cm*
Poa secunda	**9081633 (Opportunity)**	63.1 a	2.9	62.4 abcdefg
Poa secunda	9081635	23.7 bcdefg	4.1	0.8 defghij
Poa secunda (*P. ampla*)	'Sherman'	13.4 cdefghij	4.4	61.9 abcdefgh
Poa secunda	9081322	4.1 hij	4.0	7.1 fghijklm
Poa secunda	'Canbar'	2.5 j	5.0	60.8 abcdefgh

[1] – Percentage stand cover indicates the amount of cover within the seeded row, not the entire plot or stand.
[2] – A vigor rating of "1" is best, "4" is average, and "9" is worst or near death.

Table 8. Biomass production of *Poa secunda* accessions, Stucky Ridge CEP, August 29, 2006, Anaconda, MT.

	Accession Number	Biomass
Species	Or Cultivar	Production
		kg/ha
Poa secunda	**9081633 (Opportunity)**	2311.1 bcd
Poa secunda	9081635	1150.0 cdefgh
Poa secunda	9081322	444.4 fgh
Poa secunda (*P. ampla*)	'Sherman'	305.6 gh
Poa secunda	'Canbar'	122.2 gh

Conclusions

The Pre-Varietal Release process has proven to be highly effective in expediting the selection, release, and commercial availability of native seed sources for the DATR Project. In many cases, native seed sources out perform commercially available selections by significant margins. It is likely that a combination of adaptation to local environmental conditions, as well as potential tolerance to acidic and heavy metal contaminated soil, is involved.

Opportunity Germplasm Nevada bluegrass originated in the upper Clark Fork River basin of western Montana where a native stand was found growing on acidic soil impacted by acid/heavy metal contamination. Based upon the performance data in the Stucky Ridge CEP, other non-reported field trials, and in its native range, Opportunity Germplasm Nevada bluegrass is best adapted to elevations of 2,000 to 6,000 feet, performing more favorably on lower elevation (valley) sites. This selection should prove well adapted for use on drastically disturbed acidic and heavy metal impacted areas of low to mid-mountain elevations in the northern Rocky Mountain region, and especially well given soil amendment and other favorable climatic conditions.

Restorationists may assume that local plant materials such as Opportunity, released via the studies from the DATR project, are specialized and will have limited application. No doubt the acid-tolerant releases will hopefully play a significant role in revegetation associated with the Anaconda Smelter Superfund Site. Consideration in plant development beyond DATR might involve the use of these plants for broader uses in other conservation practices and geographic areas.

Continued comparative evaluations between local seed sources and commercially available material could potentially yield numerous additional releases. Some possible future releases through DATR include bluebunch wheatgrass (*Pseudoroegneria spicata*), silverleaf phacelia (*Phacelia hastata*), woolly cinquefoil (*Potentilla hippiana*), silver buffaloberry (*Shepherdia argentea*), Woods' rose (*Rosa woodsii*), and horizontal juniper (*Juniperus horizontalis*).

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