Plant Variety Release Proposal

Crop name: Barley
Variety: Successor
Experimental designation of variety: DH190481
Inventor/breeder: Patrick Hayes

Introduction

Eastern Oregon and Washington are major small grain production areas within the United States (US). These areas are primarily cultivated under dryland conditions and many growers use conservation-till or no-till practices. Wheat is the dominant small grain crop in the region with just over 1.2 million hectares planted across both states (USDA NASS, 2022). While barley is a smaller crop than wheat, it still plays an important role in each state, with approximately 29,000 hectares in Washington and 15,000 hectares in Oregon in 2022, both within the top-ten states in the US (USDA NASS, 2022). Barley has historically been part of a seasonal rotation of wheat, oil seed crops, and legumes and will commonly follow winter wheat (Schillinger et al., 2011). Barley produced in this region under dryland conditions is primarily grown for the feed market as the limited rainfall does not typically produce grain suitable for the malting stream.

Imidazolinone (IMI) herbicides are popular and effective broad-spectrum chemicals as they target an enzymatic pathway common in many plant species but have minimal to no animal toxicity. However, their mode of action also targets many commodity crops. Wheat cultivars with genetic resistance to IMI-herbicides were developed using mutagenesis and can confer complete tolerance to herbicide exposure (Pozniak & Hucl, 2004; Tan et al., 2005). These cultivars have since become part of the trademarked Clearfield® system and thus allow for IMI application to control economically important grass weeds such as cheat-grass and jointed goat-grass and a variety of broad leaf weeds. A challenge to the Clearfield system is herbicide residue in the soil and/or on stubble in no-till systems limiting rotation options with crops without any herbicide tolerance (Alister & Kogan, 2005). This has contributed to a steady decline in barley acreage in Oregon and Washington due to declining performance related to IMI residues (Rustgi, 2013). Development of suitable barley varieties with tolerance to herbicide residue is important to maintain a successful rotation and return barley to its valuable role in the regional agricultural system. Currently there is one barley variety on the market suitable for rotation into a Clearfield system – Survivor, a spring-habit, two-row variety with tolerance to IMI-herbicides released by Washington State University (Murphy & Ullrich, 2018). A tolerance allele (AHAS) was identified on chromosome 6H and this variety was developed via a back-crossing scheme with a mutant parent (Rustgi et al., 2014).

The aim of this variety development was to create an agronomically successful, spring-planted variety that is well-adapted to Oregon and Washington dryland environments and tolerant to soil residue from IMI-herbicides, while exhibiting suitable grain quality for the barley feed market.

Plant Description and Performance

Successor was selected from a set of doubled haploids developed for the purposes of introgressing IMI tolerance into OSU barley germplasm. The doubled haploids were derived in 2018 from two crosses: Survivor/Lightning and RCS124/07WA201. Lightning [SHORT11-7 (TC6W265)/HERZ 29494/2991] is a facultative, two-row variety released by Oregon State University (Hayes et al., 2021). Survivor, as noted earlier, is a variety released from Washington State University – it is an induced mutant in the variety "Bob". RCSL 124 is an introgression line developed from the cross of *H. vulgare subsp. spontaneum* x Harrington (Matus et al., 2003) and 07WA201 is a sister selection of Survivor. From these F1 crosses, a population of 411 doubled haploids was produced in 2019 via anther culture following the methods of Cistué et al. (2003). This population of germplasm is herein referred to as the successor population. These DHs were screened for IMI tolerance in the greenhouse by spraying the foliage of plants at the 2-leaf stage with the IMI-herbicide Imazamox at 79 g a.i. ha^{-1} (1.5x highest recommended dose) with 0.25% (v/v) non-ionic surfactant and 1% (v/v) ammonium sulfate. Plants that were actively growing after the herbicide application were considered tolerant and were advanced to field trials.

From the original 411 lines, 174 were found to be IMI-tolerant and 171 were advanced to an off-season seed increase in New Zealand and then to field trials in 2020 (three of the 174 had limited seed production). For the 2021 trials, 20 lines were selected based on agronomic performance for a multi-environment trial in Oregon. Finally, two selections (both from the cross of Lightning/Survivor) - DH190346 and DH190481 (Successor) – were selected for an additional round of off-season increase in Chile and into another multi-environment trial in the same locations as well as an on-farm trial with a commercial grower, both in 2022.

a. Field trials

The 171 IMI tolerant lines were evaluated in 2020 in field trials conducted at the OSU Hyslop Crop Science Field Research Lab (Corvallis, OR, USA) and at the OSU-CBARC (Pendleton, OR, USA). At Hyslop, IMI-herbicide tolerance was confirmed with a post-planting, pre-emergence, application of Imazamox at 53 g a.i. ha^{-1} (1x highest recommended dose) with 0.25% (v/v) non-ionic surfactant and 1% (v/v) ammonium sulfate. In both locations, the 171 lines were planted in an augmented randomized block design and were screened for several agronomic characteristics.

Of the 171 lines, 20 advanced on to replicated trials in 2021 and 2022; four replicates were planted in each year. At Hyslop, IMI-herbicide tolerance was again confirmed with a post-planting, pre-emergence, application of Imazamox as described for the 2020 field trial. Trials in Eastern Oregon were coordinated and managed by the OSU-CBARC in 2021 and 2022. In 2021 Successor (DH190481), Survivor, and DH190346 were included in CBARC's annual yield trials in three locations: Ione, OR; Kent, OR; and Pendleton, OR. The 2022 trials included the same three entries and was performed in the same locations with the addition of La Grande, OR. However, the 2022 Kent, OR trial was not analyzed or reported due to substantial elk damage affecting all entries. All field sites were managed according to local agricultural practices under dryland conditions. The exception was the La Grande field site which received irrigation.

b. On-farm trial

The top two accessions from the successor population - DH190346 and DH190481 (Successor) – and Survivor were evaluated in a large-scale field trial at a commercial grower – Emerson Dell Farm, The Dalles, OR, USA (45.54°N, 120.98°W). Grain was planted in approximately 0.40ha strips with the experimental lines planted as a single replicate and Survivor planted in two strips on each side of the experimental lines respectively: two for sample evaluation and two as borders to mitigate border effect. The field was previously planted to sunflowers in 2021, left as no-till fallow in 2020, and planted to soft white winter wheat in 2019. Weeds were managed with two applications using a tank mix of Affinity® at 51mL/ha (active ingredient: carfentrazone-ethyl) and Huskie® at 1096mL/ha (active ingredients: pyrasulfotole, bromoxynil octanoate, and bromoxynil heptanoate). A propiconazole fungicide was added to the tank mix at 292mL/ha for disease control. Grain was harvested on August 27, 2022 using the farm's equipment.

c. Statistical analysis

Data were assessed using ANOVA and mean comparisons were performed using Fisher's LSD. Data for the multi-environment trials presented herein are subsets of the complete trials and statistical analysis incorporates the entire dataset. Access to all data is available online here: https://cropandsoil.oregonstate.edu/wheat/osu-wheat-variety-trials.

2. Characteristics

The cross between Survivor and Lightning was made to combine the IMI-tolerance and yield potential of Survivor with the superior disease resistance qualities of Lightning. While Survivor displays resistance to IMI via a similar molecular mechanism to fully resistant wheat cultivars, it does not confer complete tolerance to IMI exposure, rather Survivor is suitable for growth in soil and/or stubble with residual IMI (Rustgi et al., 2014). Successor, gaining its AHAS allele from Survivor, thus has similar tolerance. Greenhouse screening for IMI-tolerance was performed on the complete population of 411 DHs at a rate above the recommend application dosage. Herbicide screening found that 174 lines were IMI tolerant and 171 were advanced to further evaluation trials in two locations in Oregon in 2020.

Initial agronomic evaluation of selections was performed in 2020 in Corvallis and Pendleton, OR for yield, grain quality, and disease resistance. A subset of the complete 171 selection dataset from Corvallis is shown in Table 1 (the complete dataset is available upon request) and includes the 20 selections advanced to 2021 and the two parents. The data is not reported for the 2020 Pendleton field trial as a field effect negatively impacted a large portion of the trial, including nearly half of the 20 lines selected for advancement. At Corvallis, Successor showed strong overall agronomics, with moderate yields, high test weight, and minimal susceptibility to scald (incited by *Rhynchosporium commune*) and barley stripe rust (incited by *Puccinia striiformis* f. sp. *hordei*). While scald impact was minimal across the experiment as a whole, Successor outperformed the trial average for barley stripe rust susceptibility (30.2%). Successor had an earlier heading date than Survivor and was eight days earlier than the complete experimental mean.

In the 2021 Corvallis yield trials, Successor performed well with an average yield of over 4,000kg/ha. However, there was minimal separation in yield between all entries with all but two entries falling into one group as shown in Table 2 and Successor, Lightning, Survivor, and 18 of the other DHs did not yield significantly different from each other. Successor had moderately plump grain (88.1%), but did have the second highest overall TW (682.9 g/L). It also had the lowest protein (11.3%) in the trial. There is no premium paid for grain protein in the feed barley market, so this is not an issue. It should be noted that Corvallis, a high-rainfall environment with available spring irrigation, is not a target environment for this variety in particular, or spring feed barley in general.

A multi-environment trial was performed in 2021 and 2022 to evaluate the variety in a selection of locations within the current growing region under dryland conditions. Results for the 2021 and 2022 trials are shown in Tables 3 and 4 respectively. In the 2021 trials, Successor performed similar to Survivor for most metrics at all locations. The exception was TW at Pendleton where Successor and DH190346 were both significantly higher than Survivor. The 2022 trials saw greater separation for yield and TW at the three locations analyzed. Successor saw the highest yields in La Grande and Pendleton, but did not yield significantly different than Survivor. Yields overall were much lower in Ione than the other locations and Survivor yielded significantly greater there than both Successor and DH190346. At all three locations Successor had the highest TW of the three entries, but again did not differ significantly than Survivor. Plant height of Successor was shorter than Survivor and taller than DH190346 in all locations, but this effect was only significant in La Grande, which may be a result of the available irrigation at this site. Grain protein did not differ significantly between entries at any location or year, which is most likely an indicator that protein synthesis in these lines is driven primarily by environmental conditions such as water availability (Gous et al., 2015).

The on-farm trial in Eastern Oregon was performed to evaluate the final two accessions against Survivor in a commercial setting as well as to assess grower interest in the new variety. Results from this trial are shown in Table 5. The two experimental lines were only planted as single replicates and thus no statistical analysis was performed. At this location Successor had the earliest heading date of the three entries but was similar to DH190346 (data not shown), which aligns with the Corvallis field trial. Successor was the highest yielding entry, out yielding Survivor by 300kg/ha. Protein was similar between Successor and DH190346, which were both 0.5-0.6% lower than Survivor. The largest range was found for plump grains and Successor was the lowest of the three, but interestingly had similar thin kernels to Survivor. Weather data for the location is shown in Figure 1 and shows that while temperature followed close to the 30-year mean, rainfall came in a much different pattern. The area saw much lower winter precipitation, receiving 109mm less over January and February than average. However, the spring was quite wet, with 139mm more rain than usual throughout April, May, and June. As it appears Successor heads and matures earlier than Survivor, this may indicate that Successor would be even better suited for the environment during a normal rainfall year as it would be more apt to take advantage of the typical precipitation patterns.

3. Conclusion

Successor barley is a novel variety that exhibits tolerance to residual IMI herbicide in soil and crop residue. It is well suited to dryland and conservation/no-tillage agricultural systems in Eastern Oregon and Washington. In a multi-environment trial, Successor met or exceeded the yield of Survivor in all years in all but one location year. Additionally, it had similar protein levels and test weights as Survivor. An on-farm trial showed its potential under standard management conditions in a rotation following winter wheat and was perceived favorably by the grower. Successor provides a new option for growers to add barley into a rotation with Clearfield crops, being only the second released variety with known tolerance to IMI residues.

Seed/plant production

The production of certified classes of seed is proceeding as follows. Breeder seed was produced from head row purification blocks at Hyslop Farm, near Corvallis, Oregon in 2021. Approximately one quarter of an acre of this seed was harvested in summer of 2022 in Othello, Washington by Washington State Crop Improvement Association to produce Breeder Seed. Seed for one acre (~ 50 kg) will be saved for planting ~ 1 – 2 acres of Foundation seed increase in the spring of 2023. Orders for Foundation Seed (to be harvested summer, 2023) will be taken one year in advance (in spring 2023) for planting spring of 2024.

Successor is proposed for release with a non-exclusive license, per previous OSU malting barley varieties. There will be a one-time application fee of $250 for each non-exclusive license. Those interested in a license should contact Denis Sather at the OSU Office of Commercialization and Corporate Development (denis.d.sather@oregonstate.edu). Successor seed, for planting purposes, can only be sold as a class of certified seed with a royalty of $0.02/lb (approximately $0.067/kg). The $0.02/lb royalty will be paid on sale of this seed. All grain harvested from the certified production must be disposed of by malting or feeding, unless permission is obtained - in writing - from OSU to use the seed for other purposes, including re-planting.

Plant Variety Protection will not be sought for Successor because this variety will meet an immediate short-term need for IMI-tolerant barley. Long-term, the reduced use of IMI herbicides and generally low price for feed barley do not justify the cost of PVP. The variety will be protected by Federal Seed Law and OSU recognized as the owner of the variety. Furthermore, Oregon, Idaho and Washington state trademarks will specify that the variety can only be sold under the name of “Successor”.

Acknowledgements

We would like to thank the following for contributions to this work.

We would like to thank the following for their assistance and support of this work:

- Dr. Ivan Matus, Instituto de Investigaciones Agropecuarias, for coordination of the final offseason seed increase in Chile.
- Dr. Javier Hernandez, Oregon State University, for work on genetic mapping of IMI-tolerance.
- Dr. Caio Brunharo, Pennsylvania State University (formerly at OSU), for assistance with the herbicide assays.
- Mr. David Brewers for the on-farm trial
- The Oregon Wheat Commission for critical funding.

References

Alister, C., & Kogan, M. (2005). Efficacy of imidazolinone herbicides applied to imidazolinone-resistant maize and their carryover effect on rotational crops. *Crop Protection*, *24*(4), 375–379. https://doi.org/10.1016/j.cropro.2004.09.011

Cistué, L., Vallés, M., Echávarri, B., Sanz, J., & Castillo, A. (2003). Barley Anther Culture. In I. Maluszynski, M., Kasha, K.J., Forster, B.P., Szarejko (Ed.), *Barley anther culture*. Springer Science & Business Media. https://doi.org/https://doi.org/10.1007/978-94-017-1293-4_5

Gous, P. W., Gilbert, R. G., & Fox, G. P. (2015). Drought-proofing barley (Hordeum vulgare) and its impact on grain quality: A review. *Journal of the Institute of Brewing*, *121*(1), 19–27. https://doi.org/10.1002/jib.187

Hayes, P., Carrijo, D. R., Filichkin, T., Fisk, S., Helgerson, L., Hernandez, J., Meints, B., & Sorrells, M. E. (2021). Registration of ‘Lightning’ barley. *Journal of Plant Registrations*, *15*(3), 407–414. https://doi.org/10.1002/plr2.20129

Matus, I., Corey, A., Filichkin, T., Hayes, P. M., Kling, J., Powell, W., Riera-Lizarazu, O., Sato, K., Vales, M. I., & Waugh, R. (2003). Development and characterization of recombinant chromosome substitution lines (RCSLs) using Hordeum vulgare subsp. spontaneum as a source of donor alleles in a Hordeum vulgare subsp. vulgare background. *Genome*, *46*, 1010–1023.

Murphy, K., & Ullrich, S. E. (2018). *Barley Variety "Survivor" (07M-203), Two-row Spring Barley*.

Pozniak, C. J., & Hucl, P. J. (2004). Genetic Analysis of Imidazolinone Resistance in Mutation-Derived Lines of Common Wheat. *Crop Science*, *44*(1), 23–30. https://doi.org/10.2135/cropsci2004.2300

PRISM Climate Group. (2022). *Time Series Values for Individual Locations*. Oregon State University. https://www.prism.oregonstate.edu/

Rustgi, S. (2013). Medicinal & Aromatic Plants Bringing Barley Back in Crop Rotation by Breeding for Imidazolinone Resistance. *Mecicinal and Aromatic Plants*. https://doi.org/10.4172/2167-0412.1000e148

Rustgi, S., Matanguihan, J., Mejías, J. H., Gemini, R., Brew-Appiah, R. A. T., Wen, N., Osorio, C., Ankrah, N., Murphy, K. M., & Von Wettstein, D. (2014). Assessment of genetic diversity among barley cultivars and breeding lines adapted to the US Pacific Northwest, and its implications in breeding barley for imidazolinone-resistance. *PLoS ONE*, *9*(6), 1–13. https://doi.org/10.1371/journal.pone.0100998

Schillinger, W., McKenzie, R. H., & Tanaka, D. (2011). Cultural Practices: Focus on Major Barley-Producing Regions - North America. In S. E. Ullrich (Ed.), *Barley: Production, Improvement, and Uses*.

Tan, S., Evans, R. R., Dahmer, M. L., Singh, B. K., & Shaner, D. L. (2005). Imidazolinone-tolerant crops: History, current status and future. *Pest Management Science*, *61*(3), 246–257. https://doi.org/10.1002/ps.993

USDA NASS. (2022). *Small Grains Summary - September 2022*.

Table 1. Disease screening and agronomic data from a subset of the initial Successor population from 2020 at the Corvallis, OR field site.

Line	Heading Date (DOY)	Scald (%)	Stripe Rust (%)	Plant Height (cm)	Yield (kg/ha)	Protein (%)	Plump (>6/64")	Test Weight (g/L)
Survivor	148	22	0	87	5477.4	12.6	87.5	700.2
Successor	140	0	5	77	5703.9	11.9	88.8	710.6
Lightning	151	0	0	80	3347.3	14.0	87.4	680.1
DH190675	146	5	60	90	3993.1	13.2	62.6	589.8
DH190674	140	5	70	90	3370.7	13.6	41.2	574.6
DH190634	142	0	0	80	5250.5	14.5	95.2	703.8
DH190575	142	0	0	95	6045.3	12.6	95.4	727.1
DH190541	139	0	0	85	6141.0	12.4	93.2	729.0
DH190526	146	0	0	80	4671.9	13.0	91.3	700.0
DH190523	150	1	13	100	5691.2	13.6	88.7	719.5
DH190495	148	0	0	85	5383.1	13.4	87.9	684.3
DH190435	148	0	1	95	5891.1	12.8	91.5	714.5
DH190424	148	0	1	90	6167.0	13.3	88.0	680.6
DH190417	146	2	0	93	5462.7	13.2	89.0	696.7
DH190404	145	2	1	85	5235.7	11.9	90.6	680.9
DH190366	140	5	10	85	5209.7	13.5	93.1	708.2
DH190358	150	0	2	90	5139.7	12.7	76.2	627.8
DH190346	138	0	5	83	5736.3	11.4	91.8	695.0
DH190319	142	1	1	90	5050.5	14.3	92.8	707.0
DH190317	153	0	0	90	4302.4	13.4	78.5	672.0
DH190305	142	10	0	85	6204.2	12.2	90.6	722.2
DH190287	146	0	5	80	5035.1	12.1	88.9	677.6

DOY, day of year.

Table 2. Spring yield trials from 2021 at the Corvallis, OR field site. This assessment includes 20 lines selected from the original Successor population and the two parents – Survivor and Lightning.

Line	Yield (kg/ha)		Protein (%)		Plump (>6/64")		TW (g/L)	
Survivor	4452.0	a	11.39	ab	88.0	bcd	674.5	defg
Successor	4117.4	ab	11.33	a	88.1	bcd	682.9	fg
Lightning	3028.3	ab	14.44	g	92.2	cde	649.4	bc
DH190675	3911.2	ab	12.46	abcde	82.1	b	643.3	ab
DH190674	3576.6	ab	11.81	ab	73.2	a	625.6	a
DH190634	3384.1	ab	13.77	fg	93.6	cde	655.8	bcde
DH190575	3685.1	ab	12.42	abcde	96.2	e	682.5	fg
DH190541	4106.8	ab	12.12	abcd	93.2	cde	685.7	g
DH190526	2960.2	ab	13.24	defg	95.5	de	655.8	bcde
DH190523	3631.3	ab	12.59	bcdef	92.0	cde	685.1	g
DH190495	3866.1	ab	12.51	abcde	93.1	cde	661.0	bcdef
DH190435	3695.7	ab	12.23	abcd	92.9	cde	674.5	defg
DH190424	4056.9	ab	11.92	abc	91.7	cde	653.6	bcd
DH190417	3298.9	ab	12.48	abcde	92.0	cde	651.0	bc
DH190404	3789.3	ab	11.37	a	93.3	cde	657.1	bcde
DH190366	3432.6	ab	13.10	cdef	93.8	cde	670.9	cdefg
DH190358	3738.0	ab	12.19	abcd	93.2	cde	642.3	ab
DH190346	4136.2	ab	11.67	ab	89.6	bcde	652.9	bcd
DH190319	3227.8	ab	13.46	efg	86.0	bc	650.0	bc
DH190317	2579.9	b	14.37	g	89.0	bcde	642.3	ab
DH190305	4069.5	ab	11.63	ab	88.0	bcd	676.4	efg
DH190287	3822.1	ab	11.69	ab	94.1	de	659.7	bcde

Significant differences for each metric were found at the <0.05 level**.** Letters in superscript annotate mean separation within groups; entries with the same letter are not significantly different using LSD.

Table 3. 2021 results from field trials in Eastern Oregon and Washington at three locations: Ione, OR; Kent, OR; and Pendleton, OR. Significant differences were found only for TW at the Pendleton location.

Location	Line	Yield (kg/ha)	Protein (%)	TW (g/L)
Ione	DH190346	1442.5	15.3	686.1
	Successor	1697.0	14.3	696.4
	Survivor	1550.1	15.1	677.1
Kent	DH190346	1194.8	14.4	576.7
	Successor	1690.2	12.1	620.4
	Survivor	1175.8	13.3	567.7
Pendleton	DH190346	2711.3	8.9	708.0[a]
	Successor	2541.0	8.8	718.3[a]
	Survivor	2464.8	8.4	665.5[b]

Mean separation was performed at each location and annotated results for metrics at locations were found to be significant at the <0.05 level. Letters in superscript annotate mean separation within groups; entries with the same letter are not significantly different using LSD.

Table 4. 2022 results from field trials in Eastern Oregon and Washington at three locations: Ione, OR; La Grande, OR; and Pendleton, OR. Data from Kent, OR was not included in 2022 due to elk damage at the field site.

Location	Line	Yield (kg/ha)	Protein (%)	TW (g/L)	Plant Height (cm)
Ione	DH190346	3436.5[b]	10.4	691.2[b]	62.7
	Successor	3759.3[b]	10.4	710.5[a]	64.0
	Survivor	4582.0[a]	10.1	702.8[ab]	66.8
La Grande	DH190346	5553.8[b]	11.8	705.4[b]	77.2[c]
	Successor	6680.3[a]	11.5	728.5[a]	82.3[b]
	Survivor	5877.7[ab]	11.5	728.5[a]	92.5[a]
Pendleton	DH190346	5568.4[b]	9.7	727.3[b]	79.2
	Successor	6141.1[a]	9.6	751.7[a]	87.6
	Survivor	5810.5[ab]	9.6	746.6[a]	90.9

Mean separation was performed at each location and annotated results for metrics at locations were found to be significant at the <0.05 level. Letters in superscript annotate mean separation within locations; entries with the same letter are not significantly different using LSD.

Table 5. Results of on farm trials at Emerson Dell Farm, The Dalles, OR, U.S.A. Data is the mean of two sub-samples from the same strip.

Line	Yield (kg/ha)	Protein (%)	Plump (>6/64")	Thin (<5.64")	TW (g/L)	Moisture (%)
DH190346	3921.3	12.8	98.2	2.25	654.5	10.6
Successor	3926.7	12.9	85.6	5.4	652.6	10.5
Survivor	3501.3	13.4	92.4	4.85	647.5	10.5

Figure 1. Weather data for Emerson Dell Farm comparing 2022 to the 30-year average rainfall and temperature. Data was aggregated using the Prism Climate Group tool https://www.prism.oregonstate.edu/ (PRISM Climate Group, 2022).