9-16-2020

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Received: 28 January 2020 | Accepted: 2 April 2020 | Published online: 16 September 2020

DOI: 10.1002/plr2.20044

Journal of Plant Registrations

REGISTRATION

Cultivar

Registration of ‘Purple Bounty’ and ‘Purple Prosperity’ hairy vetch

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Registration by CSSA.

Abstract

The hairy vetch (*Vicia villosa* Roth) cultivars ‘Purple Bounty’ (Reg. no. CV-12, PI 648342) and ‘Purple Prosperity’ (Reg. no. CV-11, PI 654047) were released in 2007 and 2008, respectively, by the USDA–ARS in collaboration with the Rodale Institute and the agricultural experiment stations of Pennsylvania State University and Cornell University. Hairy vetch is a commonly used annual legume cover crop grown for its cold tolerance, fast growth, large biomass production, and ability to fix N_2. However, this species has not been selected for the traits needed to optimize its use as a cover crop. Our breeding program focused on developing a cultivar that was both early flowering and had adequate winter survival and therefore adapted to mechanical termination in organic no-till production in the U.S. Northeast and Mid-Atlantic. Purple Bounty and Purple Prosperity were developed between 1998 and 2005 using recurrent selection at nurseries in Beltsville and Keedysville, MD. In 2005–2006, selections were evaluated against commercial checks for flowering time in Maryland and Pennsylvania, and in the 2006–2007 and 2007–2008 seasons they were evaluated in 10 locations (12 total site-years) across the United States for winter survival. Purple Bounty and Purple Prosperity both flowered earlier than the commercial material against which they were tested (significance depended on the date and site); Purple Bounty was the earlier flowering of the two cultivars. Purple Bounty and Purple Prosperity also had equivalent or improved winter survival compared with ‘AU Early Cover’, an early-maturing cultivar developed in the southern United States, at all test locations. Purple Prosperity is no longer commercially available, but Purple Bounty is currently licensed and distributed by Allied Seed (Nampa, ID).

1 | INTRODUCTION

Hairy vetch (*Vicia villosa* Roth; Leguminaceae) is a winter annual legume with historic use as a cover crop in the eastern and mid-Atlantic regions of the United States. Hairy vetch is one of the few annual legumes with the combination of winterhardiness, fast growth, large biomass production, and large N_2 fixation capacity necessary for it to be successful as a winter annual legume cover crop. However, much of the hairy vetch grown as a cover crop has not been selected for adaptation to specific regions or cropping

Abbreviations: AFLP, amplified fragment length polymorphism.

system needs (Brainard, Henshaw, & Snapp, 2012; Mirsky et al., 2017; Wilke & Snapp, 2008).

In organic no-till systems, cover crops are terminated mechanically. Mechanical termination is more effective when hairy vetch reaches at least 50% flowering, and preferably at early pod-set. In order to produce adequate biomass and reach flowering or pod-set before early-season cash crop planting, a cultivar with early vigor and flowering is required. Early-flowering hairy vetch cultivars (e.g., 'AU Early Cover', PI 575701; 'AU Merit', PI 619630) were developed in the southern United States and do not have adequate winter survival for the Northeast and Mid-Atlantic regions (Maul, Mirsky, Emche, & Devine, 2011; Mischler, Duiker, Curran, & Wilson, 2010). The cultivars 'Purple Bounty' (Reg. no. CV-12, PI 648342) and 'Purple Prosperity' (Reg. no. CV-11, PI 654047) were developed to fill this gap, being early flowering and having adequate winter survival in the region.

2 | METHODS

2.1 | Development of Purple Bounty

Purple Bounty was developed using recurrent phenotypic selection for winter survival and vigorous growth at Beltsville and Keedysville, MD, from a hairy vetch population (PI 561947) developed at Auburn, AL, and Americus, GA. Beltsville is located on the Coastal Plain of Maryland; Keedysville is located at a higher elevation in the Maryland Piedmont. Winter temperatures are typically colder and of longer duration at Keedysville than at Beltsville.

Screening of parental material for the breeding program was initiated with a screening of 47 hairy vetch accessions from the USDA Germplasm Resources Information Network (GRIN) collection (Supplemental Table S1). The accessions, derived from Afghanistan, Australia, Belgium, Canada, China, Germany, Greece, Hungary, Iran, Italy, Macedonia, Mexico, Portugal, the former Soviet Union, Spain, Turkey, and the U.S. states of Alabama, Georgia, and Washington, were planted at the USDA farm at Beltsville, MD, for evaluation in the 1998–1999 growing season. Endemic vetch plants were also present in the area. In addition to wild insect pollinators, hives of bumble bees were introduced to facilitate pollination. No attempt was made to constrain pollination. PI 561947 was identified as a potential winter-hardy parent, and open-pollinated seeds were collected from individual winter-surviving plants.

Selection for winter survival and early flowering continued in open-pollinated nurseries at Beltsville for three successive cycles (during the 1999–2000, 2000–2001, and 2001–2002 growing seasons). Another three cycles of selection for winterhardiness and early flowering then took place in open-pollinated nurseries at Keedysville (during the 2002–2003, 2003–2004, and 2004–2005 growing seasons).

2.2 | Development of Purple Prosperity

Purple Prosperity was selected from a cross between AU Early Cover and PI 561947. After initial population development (1998–1999) and the first cycle of selection (1999–2000) of the PI 561947-derived population as described above, seed of the PI 561947-derived population was planted in a row adjacent to AU Early Cover, and the two populations were intermated in pollination cages (2000–2001 growing season). This population then underwent four additional cycles of recurrent selection (2001–2002, 2002–2003, 2003–2004, and 2004–2005 growing seasons).

2.3 | Genomic analysis

In 2008, Purple Bounty, Purple Prosperity, AU Early Cover, 'Groff' (a farmer-selected population derived from AU Early Cover; Moyer, 2011), a variety-not-stated (VNS) population from Nebraska (Kaup Seed), a VNS population from Pennsylvania (Kings Agriseed), and Population 26, another promising selection from the breeding program, were planted in the field at Beltsville at 3-m row spacing using 100 seeds per row in a single-row, randomized design. Genomic DNA was extracted from 50 mg of dried shoot tissue from each of the vetch accessions using the PowerPlant DNA Isolation Kit (MoBio Laboratories). DNA concentration and purity were determined by measuring absorbance at 260 and 280 nm. The Amplified Fragment Length Polymorphism (AFLP) Plant Mapping Kit (Applied Biosystems) was used to process samples for AFLP analysis using restriction enzymes MseI and EcoRI. The AFLP samples were analyzed on an ABI PRISM 3100 Genetic Analyzer (Applied Biosystems) using POP-4 polymer and a 36-cm capillary; results were analyzed using GeneScan Analysis and Genotyper software (Applied Biosystems). Differentiation among the accessions was determined using a set of 217 possible validated markers (Maul et al., 2011). A distance matrix was created using Euclid's algorithm, and then hierarchical clustering was conducted using the Ward procedure (Ward, 1963) (JMP version 7.0.2, SAS Institute).

2.4 | Field evaluation

In the 2005–2006 growing season, Purple Bounty (experimental designation K-12) and Purple Prosperity (experimental designation B-35) were evaluated against commercial checks and other experimental lines for flowering time in Beltsville, MD, and State College, PA. The lines were compared with two commercial checks: (a) a VNS population sourced from Nebraska, and (b) Groff, a population selected by Steven Groff, a farmer and seed producer in Lancaster County, PA. Groff is a population of AU Early Cover that has been grown for several years in Pennsylvania and is more hardy than the AU Early Cover populations produced in the southern United States (Moyer, 2011).

The trial was planted as a randomized complete block design with four replications. Each plot was composed of a single row, 3 m in length, planted at a seeding rate of 10 seeds m^{-1}. Flowering time was rated on three dates as flowering progressed at each site (27 April, 11 May, and 15 May in Beltsville, MD, and 12 May, 18 May, and 8 June in State College, PA). Flowering time was rated as the percentage of plants flowering within the plot.

In the 2006–2007 and 2007–2008 growing seasons, Purple Bounty and Purple Prosperity were evaluated against commercial checks for winter survival at a wider range of sites across the United States, including Cornell University (Ithaca, NY), Pennsylvania State University (State College, PA), the Rodale Institute (Kutztown, PA), and the USDA–Natural Resources Conservation Service Plant Materials Centers in Aberdeen, ID; Alderson, WV; Bismarck, ND; Bridger, MT; Lansing, MI; Manhattan, KS; and Elsberry, MO. The commercial checks included AU Early Cover, Groff, and the Nebraska VNS population. The trial was planted as a randomized complete block design with three replications. Each plot was composed of a single row, 3 m in length. In 2006–2007, the seeding rate was 30 seeds per plot (10 seed m^{-1}) for all genotypes. In 2007–2008, the seeding rate was increased to 47 to 65 seeds per plot, determined by the germination percentage of each population. The number of seedlings within each plot was counted approximately 2 mo after planting and again in the spring after risk of frost had passed. Winter survival was determined based on both planted seeds and emerged plants.

2.5 | Data analysis

The flowering data were analyzed using a beta regression. Due to the inability of beta regression to incorporate 0 and 1 values, observations of 0% flowering were treated as 0.000001 and observations with 100% flowering were treated as 0.999999. Each of the three dates at the two sites were treated as a separate "environment," and line, site-date, and line × site-date interaction were treated as fixed effects; replication within environment was a random effect. Pairwise comparisons of treatment effects were conducted using Tukey's honest significant difference test.

The winter survival data were analyzed using two different dependent variables: (a) survival percentage with reference to the number of seeds planted, and (b) survival percentage with reference to the number of plants that emerged within approximately 2 mo of planting. The data were analyzed using a generalized linear mixed-effect model from a binomial distribution with a logit link, where line, site-year, and line × site-year interaction were treated as fixed effects, and replication within environment was a random effect. Pairwise comparison of treatment effects was conducted using Tukey's honest significant difference. The data were analyzed using RStudio Version 1.2.1335. Packages included agricolae (Mendiburu, 2019), car (Fox & Weisberg, 2018), emmeans (Lenth, 2019), glmmTMB (Brooks et al., 2017), lme4 (Bates, Mächler, Bolker, & Walker, 2015), nlme (Pinheiro, Bates, DebRoy, & Sarkar, 2019), and tidyverse (Wickham, 2017).

3 | CHARACTERISTICS

3.1 | Genetic relatedness

The genetic relatedness of seven accessions of vetch was determined using AFLP analysis. A set of 217 possible markers (Maul et al., 2011) was used to create a distance matrix for Purple Bounty, Purple Prosperity, Population 26, AU Early Cover, Groff, and the Nebraska and Pennsylvania VNS populations (Table 1). Distances ranged from a low of 0.005 (Pennsylvania and Nebraska VNS populations) to a high of 0.55 (Purple Bounty and Population 26). The distances among accessions largely corresponded to the expected relatedness based on known lineage and progenitor lines. For example, distance is low among Purple Bounty, Purple Prosperity, and Population 26, which all have a significant genetic contribution from PI 561947. Likewise, Groff, a selection of AU Early Cover with improved winter survival, was more related (less distance) to AU Early Cover. Surprisingly, Purple Prosperity had a relatively low relatedness to AU Early Cover, which is a parent of the population from which Purple Prosperity was selected.

3.2 | Flowering time

There were significant effects of line, site-date, and line × site-date interaction (Table 2). The mean flowering percentage at a given site-date ranged from 17 to 75%. The line × site-date interaction was due primarily to the third flowering measurement at State College, PA, which was taken after all genotypes had reached nearly full flowering; there were no observed differences among genotypes

TABLE 1 Distance matrix of genetic relatedness developed using amplified fragment length polymorphism (AFLP) analysis

	Purple Bounty	Purple Prosperity	Population 26	AU Early Cover	Groff	Nebraska VNS	Pennsylvania VNS
Purple Bounty	1.00						
Purple Prosperity	0.54	1.00					
Population 26	0.55	0.30	1.00				
AU Early Cover	0.35	0.18	0.39	1.00			
Groff	0.35	0.29	0.25	0.43	1.00		
Nebraska VNS	0.18	0.13	0.05	0.09	0.01	1.00	
Pennsylvania VNS	0.24	0.27	0.10	0.13	0.31	0.01	1.00

Note. VNS, variety not stated.

TABLE 2 Analysis of deviance using Type II Wald's χ^2 test to determine the effect of genotype and site-date on flowering time. Data were fit to a beta regression with a logit link

	χ^2	df	Pr (> χ^2)
Genotype	147.77	3	$<2.2 \times 10^{-16}$ ***
Site-date	266.01	5	$<2.2 \times 10^{-16}$ ***
Genotype × site-date	99.82	15	1.41×10^{-14}***

***Significant at $P \leq .001$.

for this set of observations. At all other site-dates, we noted similar relationships among the lines—Purple Bounty was the earliest flowering (always significantly earlier than the Nebraska VNS and sometimes significantly earlier than Groff and/or Purple Prosperity as well), followed by Purple Prosperity (sometimes significantly earlier than the Nebraska VNS), Groff, and the Nebraska VNS population (Table 3).

3.3 | Winter survival

For both winter survival calculated based on survivors per seeds planted and survivors per fall-emerged plants, there was a significant genotype × environment interaction (Tables 4 and 5). Several sites had nearly complete winter survival or nearly complete winterkill; no differences in winter survival were detected among genotypes for either metric. Among the sites that *did* have significant differences in winter survival among genotypes, there was no cultivar that consistently performed best. Two of the commercial checks (Groff and the Nebraska VNS population) sometimes performed better than Purple Bounty and Purple Prosperity in terms of winter survival. However, in all cases Purple Bounty and Purple Prosperity had winter survival equal to or better than AU Early Cover (a southern cultivar not bred for winter survival) (Table 6). These results indicate that Purple Prosperity and Purple Bounty have winter survival comparable to or better than AU Early Cover; while they did not consistently outperform Groff or the Nebraska VNS population for winter survival, they were consistently earlier flowering than those two lines.

3.4 | Additional phenotype information

Purple Bounty has purple flowers and glabrous foliage and stems. The seed has a spherical shape, blue-black testa, brown hilum, and yellow cotyledons. Seed weigh 24.5 g per 1,000 seed. Purple Prosperity has purple flowers and glabrous foliage and stems. The seed has a spherical shape, dark brown testa and hilum, and yellow cotyledons. Seed weigh 24.5 g per 1,000 seed.

4 | AVAILABILITY

Purple Bounty and Purple Prosperity were released jointly by the Rodale Institute, the Penn State University Agricultural Experiment Station, the Cornell University Experiment Station, and USDA-ARS in April 2007. In 2013, protection was granted for both cultivars under the Plant Variety Protection Act of 1994; Purple Bounty was registered under PVP no. 200700280, and Purple Prosperity was registered under PVP no. 200800302. Germplasm of both cultivars is maintained by the Sustainable Agricultural Systems Laboratory, USDA-ARS (Beltsville, MD). Accessions were also deposited in the USDA-ARS National Plant Germplasm System, where they are available for breeding, education, and research purposes.

TABLE 3 Mean flowering percentage in Beltsville, MD, and State College, PA, for three cultivars (Purple Bounty, Purple Prosperity, and Groff) and a variety-not-stated (VNS) population from Nebraska on three observation dates in 2006

Site	Date	Cultivar or population	Flowering
			%
Beltsville, MD	27 Apr.	Nebraska VNS	0.0 A
		Groff	13.1 AB
		Purple Prosperity	18.1 BCD
		Purple Bounty	38.8 DEF
	11 May	Nebraska VNS	28.3 CDE
		Groff	43.3 DEFG
		Purple Prosperity	63.5 EFG
		Purple Bounty	65.3 FG
	15 May	Nebraska VNS	42.5 DEFG
		Groff	57.5 EFG
		Purple Prosperity	72.5 FG
		Purple Bounty	80.0 H
State College, PA	12 May	Nebraska VNS	0.0 A
		Groff	4.0 ABC
		Purple Prosperity	21.3 BCD
		Purple Bounty	42.5 DEFG
	18 May	Nebraska VNS	0.3 A
		Groff	16.3 BCD
		Purple Prosperity	41.3 DEFG
		Purple Bounty	57.5 EFG
	8 June	Nebraska VNS	77.5 GH
		Groff	71.3 FG
		Purple Prosperity	75.0 FG
		Purple Bounty	75.0 FG

Note. Means followed by the same letter are not significantly different according to Tukey's honest significant difference test ($P < .05$).

TABLE 4 Analysis of deviance using Type II Wald's χ^2 test to determine the effect of genotype and site-year on winter survival, calculated on the basis of winter survivors relative to seeds planted. Data were fit to a generalized linear mixed-effect model from a binomial distribution with a logit link

	χ^2	df	Pr (> χ^2)
Genotype	129.23	4	$<2.2 \times 10^{-16}$ ***
Site-year	421.56	11	$<2.2 \times 10^{-16}$ ***
Genotype × site-year	371.04	43	$<2.2 \times 10^{-16}$ ***

***Significant at $P \leq .001$.

TABLE 5 Analysis of deviance using Type II Wald's χ^2 test to determine the effect of genotype and site-year on winter survival, calculated on the basis of winter survivors relative to fall-emerged plants. Data were fit to a generalized linear mixed-effect model from a binomial distribution with a logit link

	χ^2	df	Pr (> χ^2)
Genotype	45.87	4	2.62×10^{-9} ***
Site-year	188.74	7	$<2.2 \times 10^{-16}$ ***
Genotype × site-year	187.67	24	$<2.2 \times 10^{-16}$ ***

***Significant at $P \leq .001$.

TABLE 6 Mean winter survival for each of 12 site-years for four tested cultivars (Purple Bounty, Purple Prosperity, Groff, and AU Early Cover) and a variety-not-stated (VNS) population from Nebraska

Site-year	Cultivar or population	Winter survival per seeds planted	Winter survival per emerged plant
		%	
Aberdeen, ID (2006–2007)	Groff	77.8 A	84.9 A
	Purple Bounty	31.1 B	32.4 B
	Purple Prosperity	11.1 BC	16.0 BC
	Nebraska VNS	3.3 C	3.4 C
	AU Early Cover	3.3 C	4.0 C
Bismarck, ND (2006–2007)	Nebraska VNS	75.6 A	NA[a]
	Groff	13.3 B	NA
	Purple Prosperity	7.8 B	NA
	AU Early Cover	6.7 B	NA
	Purple Bounty	2.2 B	NA
Kutztown, PA (2006–2007)	Nebraska VNS	97.8 A	NA
	Groff	76.7 A	NA
	Purple Bounty	72.2 A	NA
	Purple Prosperity	67.8 AB	NA
	AU Early Cover	36.7 B	NA
Manhattan, KS (2006–2007)	Groff	86.7 A	100.0[b]
	Nebraska VNS	63.3 AB	88.3 A
	Purple Prosperity	50.0 BC	88.1 A
	Purple Bounty	26.7 C	72.9 AB
	AU Early Cover	23.3 C	36.2 B
State College, PA (2006–2007)	Nebraska VNS	94.4 A	NA
	Groff	66.7 B	NA
	Purple Prosperity	61.1 B	NA
	Purple Bounty	56.7 B	NA
	AU Early Cover	40.0 B	NA

Note. Means followed by the same letter within a given site are not significantly different according to Tukey's honest significant difference test ($P < .05$).
[a]NA, not available due to lack of fall emergence data. [b]Groff had no variation (100% survival in all plots) at this site, so a confidence interval could not be calculated.

Both cultivars were licensed by Allied Seed (Nampa, ID) and King's AgriSeeds (Lancaster, PA) starting in 2008. King's AgriSeeds has since discontinued production and distribution of both cultivars, but Allied Seed continues to produce and distribute Purple Bounty. Purple Bounty continues to be distributed throughout the United States, with highest sales volumes in the Northeast, mid-South, Ohio Valley, Midwest, and Pacific Northwest.

ACKNOWLEDGMENTS

We would like to thank Mr. Timothy Ellis of the University of Maryland Western Maryland Research and Education Center (Keedysville, MD) and the NRCS Plant Materials Centers for their assistance with winter survival evaluations.

CONFLICT OF INTEREST

The authors declare no conflict of interest.

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SUPPORTING INFORMATION

Additional supporting information may be found online in the Supporting Information section at the end of the article.

How to cite this article: Moore VM, Maul JE, Wilson D, et al. Registration of 'Purple Bounty' and 'Purple Prosperity' hairy vetch. *J. Plant Regist*. 2020;14:340–346. https://doi.org/10.1002/plr2.20044