

The Naked Barley Doubled Haploid Composite

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The Naked Barley Doubled Haploid Composite (NBDHC) is a resource for research and plant breeding. It consists of 909 doubled haploids derived from 35 crosses involving 28 different parental lines and 40 different grandparental lines (Table 1). Grandparents originated from the following breeding programs/germplasm collections: CIMMYT/ICARDA-Mexico (Calicuchima-sib); Crop Development Center, University of Saskatchewan (CDC Alamo); Getreidezuechtungsforschung Darzau (DZ100289, DZ100341); KWS (Fridericus, KW2-8499); Martonvasar Research Institute of the Hungarian Academy of Sciences (Luca); Oregon State University (Alba, BISON1,4,5, Full Pint, Maja, Orca, Oregon Wolfe Barley Dominant, Strider, StabBC42 ((Strider/88Ab536//Strider), Kab51 (Kold/88Ab536//Kold)); Research Institute of Bioresources, Okayama University Germplasm collection (Chame 14 Nepal, Tibet Black 8, Tibet Violet 1); University of California, Davis (Tamalpais); University of Minnesota (Legacy); University of Nebraska, Lincoln (NB3437f, P713); USDA-ARS Idaho (Charles, Transit, 2Ab09-X05W048-378HL, 2Ab11-W06Bg60-18); USDA National Small Grains Germplasm Collection (Karma = PI 60205); Virginia Polytechnic and State University (Doyce); and WestBred/Monsanto/Highland Specialty Seeds (Merlin, Waxbar). The only common denominator of the parental lines is that all are naked (hull-less) and therefore homozygous recessive at the *Nud* locus (Taketa et al., 2004). The grandparental germplasm is genetically diverse and includes known alternative alleles for growth habit (facultative, spring, and winter), inflorescence type (2-row, 6-row), plant height (semi-dwarf, standard), seed color (blue, brown, purple, white), seed starch type (normal, waxy). This diverse germplasm is also likely to have contributed allelic variation for uncharacterized traits including aroma, disease resistance, drought tolerance, flavor, malting quality, nutritional factors, and yield.

The doubled haploids were produced at Oregon State University, following the protocol of Cistue et al. (2011). The numbers of doubled haploids produced per cross combination are shown in Table 1. In addition to maintaining the set of 909 doubled haploids as a genetics resource, also harvested all doubled haploids *en masse* as a composite. The objectives of this report are to inform the research and breeding communities of this germplasm resource, to document the development of the NBDHC,

and provide a snapshot of the diversity in morphological and phenological traits present in the NBDHC.

Materials and Methods

Seed for this experiment was obtained from a 10 kg sample of Naked Barley Composite seed harvested in 2016 at the OSU Hyslop Agronomy Farm (GPS XXXXX). Seed for the 2016 plots was harvested from plots harvested at the same location in 2015, which traced to the original composite made in 2014 from plots grown at the same site. From the 2016 harvest 10 kg sample, a random sample of 305 seeds was taken and seeds were visually sorted for color for color as white, brown, brown and white, blue, and black. The distribution of seed colors is shown in Figure 1. Subsequently an additional four random samples of 450, 455, 377 and 408 seeds each were taken from the same seed lot. In order to facilitate scoring of seed color, seeds were imbibed for 24 hours and scored for color as white, brown, brown and white, blue, black, grey and purple. Results from the second color scoring experiment are shown in Figure 2. In addition, the number of seeds with adhering hulls (as measured by complete hull retention after squeezing between thumb and forefinger) was determined on the 408 seeds of the sample four used in the second assessment of seed color.

From the sample of 305 seeds, 224 seeds representing the proportion of each of the five seed color classes were selected at random from each color group and two seeds from each color group were planted in a single Cone-tainerTM. Each Cone-tainer contained approximately 156 ml of soil mix. The soil mix consisted of 1 bag (56 L) Metro-Mix[®] 840 Professional, 0.95 L of Turface (soil conditioner), 245 g of Osmocote (NPK 14-14-14), 300 g of Lime (raise soil pH), 6.7 L of Pumice (aeration and retain moisture of the soil) and 8.5 L of Perlite (aeration of the soil). The Cone-tainers were maintained in a hydroponic system, with spacing of 8.9 cm x 7.6 cm between the 120 Cone-tainers. The hydroponic solution consisted of tap water, which was maintained at a depth of 10.0 cm for the base of each container. Cone-tainers were fertilized weekly with a dilute fertilizer solution NPK 20-20-20. The plants in Cone-tainers were grown under greenhouse conditions at Oregon State University (OSU), Corvallis, Oregon, USA. The greenhouse was maintained at a constant 16⁰C with a 16h/24h photoperiod

regime supplemented with high intensity lights. Neither seeds nor plants were vernalized.

Morphological and phenological traits were scored as follows:

Phenotype	Descriptor(s)/Units	Protocol
Anthesis date	Days	Days from planting until appearance of inflorescence
Plant height	Centimeters	Distance from soil surface to tip of spike, exclusive of awns
Spike type	2-row/6-row	Visual assessment of lateral floret fertility
Spike length	Centimeters	Distance from base of first spikelet to tip of terminal spikelet
Lemma color	Black/white	Visual assessment of color
Vernalization sensitivity	Sensitive/Insensitive	Flowered or not flowered by end of experiment

Table 1 – Numbers of doubled haploids, doubled haploid pedigrees and parents of the naked barley doubled haploid composite

Number of DH	Pedigree	Female	Male
1	09OR-55/Color-F4-SSD-034	Strider/Merlin, F1//Strider	Tibet Violet 1///Luca/Merlin//Luca
4	09OR-55/DZ100341	Strider/Merlin, F1//Strider	Astrid//BGRC5110/BGRC5126//Billa
28	09OR-56/Color-F4-SSD-092	Strider/Merlin, F1//Strider	Tibet Black 8/// Luca/Merlin//Luca
16	09OR-59/2Ab09-X05W048-378HL	Strider/Merlin, F1//Strider	93Ab428/Orca//CDC Alamo
39	09OR-59/Color-F4-SSD-148	Strider/Merlin, F1//Strider	Oregon Wolfe Barley Dominant/Calicuchima sib//Luca/Merlin//Luca
31	2Ab09-X05W048-378HL/DZ100289	93Ab428/Orca//CDC Alamo	BGRC5110/BGRC5126//Tiffany///Carrero
18	2Ab09-X05W048-378HL/OR101	93Ab428/Orca//CDC Alamo	StabBC 42///Kab 51/Legacy//Kab 51
20	2Ab09-X05W048-378HL/Tamalpais	93Ab428/Orca//CDC Alamo	Tamalpais
6	2Ab11-W06Bg60-18/09OR-56	93Ab428/Orca//Azhul/Thuringia	Strider/Merlin, F1//Strider
5	Alba/Color-F4-SSD-151	Alba	Oregon Wolfe Barley Dominant/Calicuchima sib//Luca/Merlin//Luca
45	Alba/DZ100341	Alba	Astrid//BGRC5110/BGRC5126//Billa
16	Chame 14 Nepal///Luca/Merlin//Luca	Chame 14 Nepal	Luca/Merlin//Luca
12	Charles/Color-F4-SSD-092	Charles	Tibet Black 8/// Luca/Merlin//Luca
25	Color-F4-SSD-002/10.0655	Chame 14///Luca/Merlin//Luca	KW2-849/Luca/Waxbar//Luca
9	Color-F4-SSD-092/Full Pint	Tibet Black 8/// Luca/Merlin//Luca	Full Pint
5	Color-F4-SSD-092/Bison 1,4,5	Tibet Black 8/// Luca/Merlin//Luca	Bison 1,4,5
50	Color-F4-SSD-092/PO71DH-87	Tibet Black 8/// Luca/Merlin//Luca	P713/OR71
17	Color-F4-SSD-148/OBADV10-13	Oregon Wolfe Barley Dominant/Calicuchima sib//Luca/Merlin//Luca	Strider/Doyce
10	DH 10.0655/09OR-55	KW2-849/Luca/Waxbar//Luca	Strider/Merlin, F1//Strider
37	DH 10.0655/Color-F4-SSD-034	KW2-849/Luca/Waxbar//Luca	Tibet Violet 1///Luca/Merlin//Luca
29	DH 10.0655/Color-F4-SSD-043	KW2-849/Luca/Waxbar//Luca	Tibet Violet 1///Luca/Waxbar//Luca
3	DH 10.0655/Karma	KW2-849/Luca/Waxbar//Luca	Karma
12	DH 10.0655/Tamalpais	KW2-849/Luca/Waxbar//Luca	Tamalpais
14	DH 10.0969/Color-F4-SSD-138	KW2-849/Luca/Waxbar//Luca	Tibet Black 8///Luca/Merlin//Luca
11	NO71DH-86/Color-F4-SSD-092	NB3437f/OR71	Tibet Black 8/// Luca/Merlin//Luca
18	OBADV10-13/Color-F4-SSD-092	Strider/Doyce	Tibet Black 8/// Luca/Merlin//Luca
3	OBADV10-13/Tamalpais	Strider/Doyce	Tamalpais
1	OR101/Color-F4-SSD-017	StabBC 42///Kab 51/Legacy//Kab 51	Chame 14///Luca/Merlin//Luca
14	OR101/Color-F4-SSD-138	StabBC 42///Kab 51/Legacy//Kab 51	Tibet Black 8/// Luca/Merlin//Luca
13	OR101/DZ100341	StabBC 42///Kab 51/Legacy//Kab 51	Astrid//BGRC5110/BGRC5126//Billa
17	Tetonia/Karma	Tetonia	Karma (PI 60205)
292	Tibet Black 8///Luca/Merlin//Luca	Tibet Black 8	Luca/Merlin//Luca
25	Tibet Violet 1///Luca/Merlin//Luca	Tibet Violet 1	Luca/Merlin//Luca
3	Tibet Violet 1///Luca/Waxbar//Luca	Tibet Violet 1	Luca/Waxbar//Luca
5	Transit/DH 10.1126	Transit	Fridericus/Maja/Legacy//Maja//Doyce

