UNITED STATES DEPARTMENT OF AGRICULTURE
AGRICULTURAL RESEARCH SERVICE

in cooperation with

STATE AGRICULTURAL EXPERIMENT STATIONS

Results from the

UNIFORM OAT WINTER HARDINESS NURSERY

2010-2011

Compiled by

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This is a joint progress report of an investigation underway in the State Agricultural Experiment Stations and the Agricultural Research Service of the U. S. Department of Agriculture. It contains preliminary data which have not been sufficiently confirmed to justify general release; interpretations may be modified with additional experimentation. Confirmed results will be published through established channels. The report is primarily a tool for cooperators, their staff and those with special interest in agricultural research program development.

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CONTENTS

COOPERATING AGRICULTURAL EXPERIMENT STATIONS AND PERSONNEL

Country	State	AES Location	Personnel
USA	AR	Fayetteville	E. Mason
USA	IL	Lawrenceville	L. Phillippe
USA	NC	Laurel Springs/Waynesville	D. Marshall/M. Fountain
USA	TN	Knoxville	D. West
USA	LA	Baton Rouge	S. Harrison
USA	ID	Alberdeen	D. Obert
Poland	Blonie	Plant Breeding and Acclimatization Ins.	B. Lapinski
Czech Republic	Kromeriz	Agricultural Research Institute	M. Kadlíková
Hungary	Martonvasar	Agric. Res. Inst. of Hungary Academy	O. Veisz
Austria	Edelhof	Saatzucht Edelhof	S. Berger & H. Hofbauer

DIGEST

NUMBER OF TESTS:	10 tests (6 US States, 4 foreign countries)	
NUMBER OF ENTRIES:	16	
EXPERIMENTAL DESIGN:	Single-row, 5-foot plot Two replications Randomized complete block	
DATA RECORDED:	Percent winter survival	
DATA NOT USED IN ANALYSIS:	Fayetteville, AR Martonvasar, Hungary Alberdeen, ID Lawrenceville, IL Knoxville, TN Baton Rouge, LA	No Data No Data Not Planted Not Planted 100% Survival 100% Survival

COMMENTS:

- Analysis of markers assocated with winter hardiness was implemented beginning with the 2008-2009 nursery.
- All new oat lines will be evaluated with Simple sequence repeats (SSRs) associated with winter hardiness traits and continue to be to added to the report.

US STATE/COUNTRY	LOCATION	COOPERATORS' COMMENTS
Poland	Radzików	The survival was almost complete till the end of February when snow cover disappeared and frost caused severe damages (minimal temperature on Feb. 24, -9.5C on the ground level and -14.8C on 5cm above the ground).
Czech Republic	Kromeriz	The plants were grown in autumn without problem, but due the low temperatures during January and February and absence of snow cover, plants were badly damaged. The number of survived plants was low
Austria	Edelhof	Sowed on October 19th, 2010. Bad germination no: 1,2,3,4. Some very warm days at the begenning of February. Very dry winter condition. Snow layer: 50 days but on unfrozen soil.

Table 1. Entries in the 2010-2011 Uniform Oat Winter Hardiness Nursery.

Entry No	Entry name	Pedigree	Yrs in Nursery	Contributors	
1	Fulgum (ck)	CI 708	73		
2	Norline (ck)	CI 6903	50		
3	Winter Turf (ck)	CI 3296	70		
4	Wintok (ck)	CI 3424	70		
5	NC07-3801lb	SS76-40/TAM397	2	Murphy	NC
6	NC07-3834y	RODGERS/SS76-40	1	Murphy	NC
7	NC07-3843y	RODGERS/SS76-40	2	Murphy	NC
8	NC07-3966yv	LA9339/SS76-40	1	Murphy	NC
9	NC08-2590y	SS76-40 / TX00D161	1	Murphy	NC
10	NC08-2723yv	SC961246 / TX00D161	1	Murphy	NC
11	NCBYDV 121	Brooks/A. Macrostachya//Brooks	1	Murphy	NC
12	PR-4H6	F1(Wintok x Avena macrostachya B6) x Leggett95-43Cn4	1	Lapinski	Poland
13	PR-5Q5	F1(Ebmeyer992212 x Avena macrostachya B6) x free poll	1	Lapinski	Poland
14	Win/Nor-1	Wintok x Norline	8	Livingston, Murphy	NC
15	Win/Nor-10	Wintok x Norline	9	Livingston, Murphy	NC
16	Win/Nor-10b	Selection from Win/Nor-10	7	Livingston, Murphy	NC

Top 10 ranked survival entries for 2010-2011

Rank	Ent No.	Entry	Pedigree	% Survival (across locations)
1	13	PR-5Q5	F1(Ebmeyer992212 x Avena macrostachya B6) x free pollination	54
2	6	NC07-3834y	RODGERS/SS76-40	53
3	5	NC07-3801lb	SS76-40/TAM397	49
4	14	Win/Nor-1	Wintok x Norline	49
5	4	Wintok (ck)	CI 3424	47
6	12	PR-4H6	F1(Wintok x Avena macrostachya B6) x Leggett95-43Cn4	46
7	15	Win/Nor-10	Wintok x Norline	45
8	16	Win/Nor-10b	Selection from Win/Nor-10	45
9	8	NC07-3966yv	LA9339/SS76-40	44
10	11	NCBYDV 121	Brooks/A. Macrostachya//Brooks	40
			LSD (0.05)	**14**

Table 2a. Winter Oat Survival (%) at Various Stations (sorted by entry number)

Ent. No.	Entry Name	Ranked Means	Means across loc	Radzikow Poland	Kromeriz Czech Republic	Edelhof Austria	Laurel Spring NC
1	Fulgum (ck)	16	11	0	4	0	40
2	Norline (ck)	13	32	14	16	56	43
3	Winter Turf (ck)	15	25	0	8	48	43
4	Wintok (ck)	5	47	27	21	75	65
5	NC07-3801lb	3	49	8	31	63	95
6	NC07-3834y	2	53	47	24	58	85
7	NC07-3843y	11	39	12	10	47	88
8	NC07-3966yv	9	44	1	18	70	88
9	NC08-2590y	12	38	18	16	37	83
10	NC08-2723yv	14	31	9	9	26	83
11	NCBYDV 121	10	40	0	21	48	93
12	PR-4H6	6	46	12	19	74	80
13	PR-5Q5	1	54	30	5	84	100
14	Win/Nor-1	4	49	57	14	44	83
15	Win/Nor-10	7	45	66	14	15	85
16	Win/Nor-10b	8	45	50	11	33	85
	Average		41	22	15	48	77
	LSD (0.05)		14	*ns	*ns	*ns	31
	CV(%)		16	*ns	*ns	*ns	19

*ns = not significant

Table 2b. Winter Oat Survival (%) at Various Stations (sorted by rank)

Ent. No.	Entry Name	Ranked Means	Means across loc	Radzikow Poland	Kromeriz Czech Republic	Edelhof Austria	Laurel Spring NC
13	PR-5Q5	1	54	30	5	84	100
6	NC07-3834y	2	53	47	24	58	85
5	NC07-3801lb	3	49	8	31	63	95
14	Win/Nor-1	4	49	57	14	44	83
4	Wintok (ck)	5	47	27	21	75	65
12	PR-4H6	6	46	12	19	74	80
15	Win/Nor-10	7	45	66	14	15	85
16	Win/Nor-10b	8	45	50	11	33	85
8	NC07-3966yv	9	44	1	18	70	88
11	NCBYDV 121	10	40	0	21	48	93
7	NC07-3843y	11	39	12	10	47	88
9	NC08-2590y	12	38	18	16	37	83
2	Norline (ck)	13	32	14	16	56	43
10	NC08-2723yv	14	31	9	9	26	83
3	Winter Turf (ck)	15	25	0	8	48	43
1	Fulgum (ck)	16	11	0	4	0	40
	Average		41	22	15	48	77
	LSD (0.05)		14	*ns	*ns	*ns	31
	CV(%)		16	*ns	*ns	*ns	19

*ns = not significant

Table 3. Uniform Oats Winter Hardiness Nursery Under Controlled Environment Freeze Test

Entry #	Entry Name	Survival Rating[1]	% Survival[2]
1	Fulgum (ck)	0.5	8
2	Norline (ck)	2.4	88
3	Winter Turf (ck)	1.7	71
4	Wintok (ck)	2.9	65
5	NC07-3801lb	0.8	55
6	NC07-3834y	1.3	65
7	NC07-3843y	1.9	78
8	NC07-3966yv	1.1	55
9	NC08-2590y	1.5	78
10	NC08-2723yv	2.1	84
11	NCBYDV 121	2.7	96
12	PR-4H6	1.6	68
13	PR-5Q5	1.9	88
14	Win/Nor-1	1.7	85
15	Win/Nor-10	2.4	93
16	Win/Nor-10b	2.3	85
	Average	1.8	72
	LSD (5%)	0.6	14
	CV	16	9

Parameters:

-2 reps/10 plants per rep planted in cone-tainers (Livingston et al. 2005, Crop Science, 45:1545-1558)
-5 weeks at 13°C; 12 hours light/dark period; 400µmole light intensity
-3 weeks at 3°C; 12 hours light/dark period; 350µmole light intensity
-3 days @ -3°C in the dark (subzero acclimation)
-Frozen @ 1°C/hour to -12°C for 3 hours
-Thawed @ 2°C/hour to 3°C
-Plants were watered once with 0.001% (v/v) Vitavax fungicide solution
-Plants were allowed to recover for 3 weeks in the greenhouse
-Plants were rated for regrow after 21 days by visually assessing leaves and roots.

[1]Rating:
0 = Completely dead
1 = 1 survived (green) shoot or 1 primary root
2 = 1 or 2 survived (green) shoots or 1 survived shoot and 1 or 2 primary roots
3 = 1 or 2 survived shoots with developed roots (primary and secondary roots)
4 = 95% survived shoots with well developed roots
5 = 100% survived with very little or no sign of freeze damage; same as unfrozen plants

[2]Survival (%):
-50% of plants with rating of 1plus all plants rated >2 divided by total number of plants frozen multipled by 100

Marker-Assisted Selection (MAS)

Winter hardiness is related to multiple quantitative traits, including winter field survival, crown freezing tolerance, vernalization response, and photoperiod. Crown freezing tolerance (CFT) is measured in controlled freeze tests and is an important component to winter hardiness. Photoperiod (PPD) and vernalization response (VRN) are frequently correlated with winter field survival and freezing tolerance because these traits contribute to delaying new growth until after the danger of freezing temperatures has passed. This avoidance response, affected by the combination of certain photoperiod and vernalization traits, can be useful for increasing winter field survival. The 7C-17 translocation is thought to contain a cluster of genes for increased tolerance to freezing temperatures and has been significantly correlated with winter field survival and crown freezing tolerance.

Genetic markers are fragments of DNA that are linked with known genes or traits. Associating markers with winter hardiness component traits (above) provides a valuable tool for oat research programs. Simple sequence repeats, known as SSRs or microsatellites, are a popular marker choice due to their relative low cost and ease of use. Oat SSR markers were previously evaluated for their association with winter hardiness component traits in a 'Fulghum' x 'Norline' population and in an oat association mapping population consisting of 25 spring sown, 36 fall sown, and two facultative oat lines. The markers associated with selected traits were chosen for testing with lines from the Uniform Oats Winter Hardiness Nursery, and the data are presented in Table 4.

If the DNA fragment associated with the phenotype of interest is present, this suggests that the line may have the winter hardiness trait. For example, the SSR marker HVM20 is associated with crown freeze tolerance and the 7C-17 translocation. The presence of the HVM20-142bp allele could translate to increased crown freeze tolerance and would be desirable in a marker-assisted selection program.

In the case of winter hardiness, a combination of traits is necessary, and marker selection at this stage is preliminary. Even though entries in the nursery may be winter hardy, they may not necessarily possess all the winter hardiness component markers. Further research will clarify which combination of traits, and therefore which SSR markers, are most informative for the development of a marker-assisted selection program.

Table 4. Markers Associated with Winter Hardiness Traits

Entry No.	Primer			AM2	AM102	AM270S-1	HVM20	JAO4042	JAO4234a	JAO4234b
	Traits[1]			RS, LS, CFT	RS, LS, CFT	FT, TR, LS, CFT	LS, RS, CFT, TR	TR	VRN, RS, LS, CFT	CFT
	Allele Size (bp)			164	220	206	142	262	260	283
	Entry name	# of Significant Alleles Associate with Winter Hardiness	Field % Surv.							
1	Fulgum (ck)	2	11	no	yes*	no	no	no	no	no
2	Norline (ck)	9	32	no	yes	yes	yes	yes	no	yes
3	Winter Turf (ck)	5	25	yes	yes	no	no	yes	no	no
4	Wintok (ck)	10	47	yes	yes	yes	yes	yes	no	yes
5	NC07-3801lb	7	49	no	yes	yes	yes	yes	no	no
6	NC07-3834y	7	53	yes	yes	yes	yes	yes	no	no
7	NC07-3843y	7	39	yes	yes	yes	yes	yes	no	no
8	NC07-3966yv	6	44	no	yes	yes	yes	yes	no	no
9	NC08-2590y	8	38	yes	yes	yes	yes	yes	no	no
10	NC08-2723yv	8	31	no	yes	yes	yes	yes	no	no
11	NCBYDV 121	8	40	no	yes	yes	yes	yes	no	yes
12	PR-4H6	9	46	no	yes	yes	yes	yes	no	yes
13	PR-5Q5	3	54	no	no	no	no	yes	no	no
14	Win/Nor-1	12	49	yes	yes	yes	yes	yes	no	yes
15	Win/Nor-10	11	45	yes	yes	yes	yes	yes	no	yes
16	Win/Nor-10b	10	45	yes	yes	yes	yes	yes	no	yes

* Results differ from previous testing with 'Fulghum' and 'Norline' and control DNA samples, and may represent variation within these lines using these markers.

Table 4 (cont.)

	Primer	JAO4636	VRN1	Xncs15-3	AME23	AME178	AME184a	AME184b
	Traits[1]	CFT	VRN	CFT, TR	MAT, LPPD, SPPD, VRN, NO-VRN, MAT-VLD, RS, LS, CFT	RS	CFT	WFS, MAT, LPPD, SPPD, VRN, NO-VRN, MAT-VLD
	Allele Size (bp)	286	390	232	263	182	190	193
Entry No.	Entry name	no	no	no	no	no	yes	yes
1	Fulgum (ck)	yes	yes	yes	yes	no	no	yes*
2	Norline (ck)	no	no	no	yes	no	no	yes
3	Winter Turf (ck)	yes	yes	yes	no	no	no	yes
4	Wintok (ck)	no	no	yes	no	yes	no	yes
5	NC07-3801lb	no	no	yes	no	no	no	yes
6	NC07-3834y	no	no	yes	no	no	no	yes
7	NC07-3843y	no	no	yes	no	no	no	yes
8	NC07-3966yv	no	yes	yes	no	no	no	yes
9	NC08-2590y	no	yes	yes	no	yes	yes	no
10	NC08-2723yv	no	no	yes	no	yes	no	yes
11	NCBYDV 121	yes	yes	yes	no	no	no	yes
12	PR-4H6	no	no	yes	no	no	no	yes
13	PR-5Q5	yes	yes	yes	yes	yes	no	yes
14	Win/Nor-1	yes	yes	yes	yes	no	no	yes
15	Win/Nor-10	yes	yes	yes	yes	no	no	no
16	Win/Nor-10b	no	no	no	no	no	yes	yes

* Results differ from previous testing with 'Fulghum' and 'Norline' and control DNA samples, and may represent variation within these lines using these markers.

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Footnotes for table 4.

Entries in the 2010-2011 Oat Winter Hardiness Nursery were evaluated with SSR and PCR markers associated with winter hardiness traits identified by Maloney et al. (submitted). Alleles shown are associated with positive effects on winter hardiness traits in the "Fulghum' x 'Norline' mapping population and the oat association mapping population.

1. **WFS** = Winter field survival. Winter field survival was estimated after spring re-growth as the percent survival for the plots corrected for plot variation in germination or fall growth.
   FT = Freeze Tolerance
   TR = Translocation 7C-17
   MAT = Maturity
   MAT-LVD = Maturity in vernalized long day treatment
   PPD = Photoperiod. Includes long and short photoperiod effects after 42 days of differing photoperiod treatments. Photoperiod, combined with vernalization and heading date, can mediate avoidance of freezing temperatures.
   LPPD = Long Photoperiod
   SPPD = Short Photoperiod
   VRN = Vernalization. Vernalization response to differing temperature treatments during seed germination, as described in Wooten et al. (Crop Sci (2009) in press). Vernalization time, along with photoperiod and heading date, may result in avoidance of freezing temperatures.
   NO-VRN = No vernalization
   CFT = Crown Freeze Tolerance. Crown freezing tolerance was evaluated by preparing and freezing crowns in a controlled environment chamber as described by Wooten et al. (2009). After three weeks of re-growth, recovery for each crown was visually measured on a scale of 0-10 (0=complete plant death, 10=no freezing damage).
   RS = Root Score
   LS = Leaf Score
2. JAO primer sequences may be requested from Joe Anderson at USDA-ARS, Purdue University.
3. VRN1 fragments are amplified by PooidVRN1 and Asintron-Rev in: Preston, J.C. and Kellogg, E.A. (2008), Plant Physiol. 146, no. 1: 265-276. VRN1 is involved in regulating response to vernalization in wheat and oats.
4. Marker is associated with presence or absence of the translocation 7C-17. Fulghum non-T7C-17, Norline T7C-17. Presence of the translocation is associated with increased crown freezing tolerance (Wooten et al. Crop Sci 47:1832-1840 (2007)).