Registration of 'Lustre' durum wheat

Andrew C. Hogg, Patrick Carr, Jed Eberly, Chengci Chen, Calla Kowatch-Carlson, Frankie Crutcher, Peggy Lamb, Kyla McNamara, Eleri Haney, Ken Kephart, Valerie Smith, Linda Dykes, Xianming Chen, Li Huang, Mike Giroux

Registration of 'Lustre' Durum Wheat

A.C. Hogg[1], P.M. Carr[2], J.O. Eberly[2], C. Chen[3], C. Kowatch[3], F.D. Crutcher[3], S. Hoesel, P.F. Lamb[4], K. McNamara[4], K.D. Kephart[5], V. Smith[5], L. Dykes[6], X. Chen[7], L. Huang[1], M.J. Giroux[1]

Affiliations:

[1]Dep. of Plant Sciences & Plant Pathology, Montana State Univ., Bozeman, MT 59717-3140, USA

[2]Central Agricultural Research Center, 52583 US Hwy 87, Moccasin, MT 59462, USA

[3]Eastern Agricultural Research Center, 1501 North Central, Sidney, MT 59270, USA

[4]Northern Agricultural Research Center, 3710 Assinniboine Rd., Havre, MT 59501-8412, USA

[5]Southern Agricultural Research Center, 748 Railroad Hwy., Huntley, MT 59037, USA

[6]USDA-ARS, Cereal Crops Research Unit, Fargo, ND 58102, USA

[7]USDA-ARS, Wheat Health, Genetics and Quality Research Unit, Pullman, WA 99164-6430, USA

Correspondence: M.J. Giroux, Dep. of Plant Sciences & Plant Pathology, Montana State Univ., Bozeman, MT 59717-3150, USA. Email: mgiroux@montana.edu

Abbreviations: CARC, Central Agricultural Research Center; Cd, cadmium; *cdu1,* gene controlling cadmium accumulation in grain; EARC, Eastern Agricultural Research Center; IT, infection type; MAES, Montana Agricultural Experiment Stations; SARC, Southern Agricultural Research Center; NARC, Northern Agricultural Research Center; *ssIIa*, starch synthase IIa; WTARC, Western Triangle Agricultural Research Center

ABSTRACT

‘Lustre’ is a spring durum wheat [*Triticum turgidum* L. ssp. *durum* (Desf.)] developed by the Montana Agricultural Experiment Station and released in 2020. Lustre was bred using the single seed descent method and was selected for its yield performance under dryland conditions across Montana, low grain cadmium accumulation, good pasta firmness, high grain protein, high yellow semolina color, and low semolina ash. Lustre performs well in both the North Central and North East regions of Montana, where most Montana durum is produced and intended for pasta production. Lustre has similar stripe rust resistance/susceptibility as top grown durum cultivars in the state with susceptibility at the seedling stage and high-temperature adult-plant resistance. Lustre is moderately susceptible to Fusarium head blight like other durum cultivars. Luster is resistant to the predominant races of stem and leaf rust and is moderately resistant to fungal leaf spot complex. Lustre is approximately 89.4 cm tall, with a yellow green color and a heading date one day later than the cultivar ‘Mountrail’. Lustre has an erect flag leaf and an erect tapering head having white glumes and awns.

INTRODUCTION

In the United States, durum wheat [*Triticum turgidum* L. ssp. *durum (*Desf.)] is grown primarily for pasta production. Starch-based foods in which the ratio of amylose to amylopectin has been increased have a lower glycemic index and improve digestive system health due to the increase in resistant starch (Regina et al. 2006, Sissons et al. 2020). Starch branching enzyme II (*sbeII*) and starch synthase IIa (*ssIIa*) are the predominant genes controlling the ratio of amylose to amylopectin in wheat seeds. As with bread wheat, the ratio of amylose to amylopectin in durum has major implications for both nutrition and product quality. Improved pasta quality may be achieved by increasing amylose content in durum. Pasta prepared from high amylose *ssIIa* and *sbeIIa* null durum has increased firmness, resistant starch, and protein content (Hogg et al. 2015; Hazard et al. 2015). While some quality traits are improved, seed size and milling yields are significantly reduced in full *ssIIa* null lines due to a reduction in overall starch content (Hogg et al., 2015). Selecting for *ssIIa* alleles that retain partial function or durum containing null mutations in only one of the two *ssIIa* genes is crucial to maintain seed size and milling yields while increasing product quality.

In addition to selecting for traits such as grain yield and quality, low accumulation of cadmium (Cd) in grain is an important trait in durum wheat. Compared to other cereals, durum wheats may accumulate high levels of grain Cd (Perrier et al., 2016), representing a risk to human health. Consumption of Cd may lead to several health concerns (Grant et al., 1998; Olsson et al., 2005; Menke et al., 2009; Nishijo et al., 2006). The major gene controlling Cd accumulation in durum is c*du1* and selection for the functional *cdu1* allele significantly reduces grain Cd levels (Clarke et al., 1997).

The objective of this research was to develop a high yielding durum wheat with improved semolina and pasta quality, low grain cadmium accumulation, and disease resistance. This was achieved by combining a single *ssIIa* null allele along with the low accumulation *cdu1* allele into a high yielding background. 'Lustre' was selected for high yield, disease resistance, pasta quality, low cadmium uptake, and resistance to stem and leaf rust.

METHODS

Pedigree and breeding history

'Lustre' (Reg. no. ____, PI ____) is a spring durum wheat having the pedigree 'Divide' (PI 642021, Elias and Manthey, 2007)//'Mountrail' (PI 607540, Elias and Miller, 2000)/M175. The experimental line M175 was selected from an ethyl methanesulfonate (EMS) mutant population that had parents 'Mountrail'/PI 330546 (Martin et al., 2012; Hogg et al., 2013). PI 330546 was obtained from the National Small Grains Collection and carries the allele for low Cd accumulation (*cdu1*) and a null *ssIIa-A* allele (deletion in first exon at position 145-174 of coding sequence) that confers firmer noodles (Martin et al. 2012). PI 330546 is resistant to stem rust and has high-temperature adult-plant resistance to stripe rust but is susceptible at the seedling stage. PI 330546 is susceptible to Hessian fly and is taller than modern varieties (approximately 140 cm). PI 330546 has long slender heads that have white awns and glumes with oblong brown/tan seeds. Offspring from the cross 'Mountrail'/PI 330546 were advanced to the F_5 generation via single seed descent with selection for agronomics and yellow/amber grain color. In the F_5 generation the best agronomic plants possessing the *ssIIa-A* null allele and the low accumulation *cdu1* allele were selected and bulked and F_6 seed from selected lines was treated with 0.5% EMS. Mutagenized plants were advanced two generations in the greenhouse to the M_2 generation. Plants were genotyped in the M_2 for novel mutations in *ssIIa-B*. Line M175 was homozygous for *cdu1*, the null *ssIIa-A* allele from PI330546 and for a newly created allele in *ssIIa-B* that contains a missense mutation resulting in amino acid change D327N (Hogg et al., 2013). Line M175 was first crossed to Mountrail and the resultant F_1 was then crossed to Divide. The resultant F_1's were genotyped for the two mutations in *ssIIa-A* and *ssIIa-B* and dual heterozygous plants were advanced. Approximately 600 Mountrail//Divide/M175 F_2's were advanced by single seed descent to the F_4 generation at which point a whole head was harvested. In 2014, 262 F_5's were planted in spaced head-rows in Bozeman, MT at the Post Agronomy Farm and the best agronomic plant (tillering, height, disease, head size) was selected from each row and threshed using a single plant thresher. Lines were genotyped for *ssIIa-A*, *ssIIa-B*, and *cdu1* by direct sequencing of PCR products.

Line selection and evaluation

In 2015, 262 $F_{5:6}$ lines along with several varietal checks were grown in replicated short rows (1.2 m) using a randomized complete-block design (RCBD) at the Montana State Post Agronomy Farm in Bozeman, MT under dryland and irrigated conditions. Lines were evaluated for agronomic traits (height, heading date, maturity date, yield) and quality traits (protein, seed size) and a single row from the irrigated environment was harvested with a binder and Vogel thresher for seed stock. The top 40 yielding lines across both environments were selected for further testing and $F_{5:7}$ plants were grown in replicated plots using a RCBD in 2016 at the Post Farm under dryland and irrigated conditions (1.2 x 3 m) and in Conrad, MT under dryland conditions (1.5 x 4.5 m). Lines were evaluated for agronomic traits (height, heading date, maturity date) and quality traits (protein, mixograph, semolina color). To assess end-product quality grain was milled and noodles prepared as in Martin et al., (2012). Milling to semolina was carried out following AACCI Approved Method 26-41.01 with modifications and noodle analysis was carried out according to AACCI Approved Methods 14-22.01 and 66-50.01. The top performing lines (MTD16001-11, $F_{5:8}$) were selected for testing in the Montana Intrastate Durum Variety Trials. From 2017-2019 the Montana Intrastate Variety Trials consisted of 24 entries with three replicates per entry, which were planted in either an alpha lattice design or RCBD. Plot size, row number and row spacing varied by location to accommodate local equipment requirements. Plots were seeded with approximately 2.2 million seeds per hectare. Lustre was tested for three years (2017-2019) as MTD16005 at 14 locations across Montana to evaluate agronomics and quality traits were assessed by the USDA-ARS quality lab in Fargo, ND. Lustre was evaluated in the Montana Intrastate Durum Variety Trials at eight Montana Agricultural Experiment Station (MAES) locations in 2017 ($F_{5:8}$). Lustre was grown at these eight MAES locations again in 2018 ($F_{5:9}$) and 2019 ($F_{5:10}$) along with six off-station locations evaluated in both 2018 and 2019. MAES locations tested included Bozeman Post Agronomy Farm (irrigated and dryland), Southern Agricultural Research Center (SARC; dryland), Eastern Agricultural Research Center (EARC; irrigated, dryland and 3 off-station), Western Triangle Agricultural Research Center (WTARC; dryland), Central Agricultural Research Center (CARC; dryland), and Northern Agricultural Research Center (NARC; dryland and 3 off-station). Lustre was selected for high yield, disease resistance, pasta quality, low cadmium uptake, and resistance to stem and leaf rust.

Purification and increase of seed stock

In 2018, $F_{5:9}$ plants were grown in a 1.2 x 21.3 m plot under dryland conditions for initial seed increase. The rows were rogued for off-types several times throughout the season before being bulk harvested. In 2019, a 0.12 ha field of $F_{5:10}$ plants was planted as breeder's seed. The field was rogued to eliminate off-types several times throughout the season prior to being bulk harvested and approximately 25 bushels of $F_{5:11}$ seed was delivered to the Montana State University Foundation Seed Program.

Statistics

Replicated agronomic and quality data from the Montana Intrastate Durum Variety Trials (2017-2019) were analyzed using analysis of variance (ANOVA) with PROC GLM (SAS v. 9.4, SAS Institute, Cary, NC). Each combination of location and year was considered a unique environment. Experiments grown at the same location in the same year, but with different irrigation regimes, were considered separate location-years, or environments. Fungal leaf spot data from 2019 was analyzed separately by location using ANOVA with PROC GLM. Fusarium head blight data was analyzed separately by year using ANOVA with PROC GLM. Where ANOVA was conducted significant differences between entries were determined using an unrestricted LSD test ($\alpha = 0.05$). Data for stripe rust, leaf rust, stem rust, cadmium concentration, and aluminum tolerance were single rep observations within a given year.

CHARACTERISTICS

Agronomic characteristics

Lustre is a spring durum wheat that carries the *cdu1* allele for low grain cadmium accumulation as well as a null *ssIIa-B* allele that increases pasta firmness. Lustre is a standard height durum of approximately 89.4 cm tall, similar to other full height durum varieties, has white glumes and awns, and has a heading date one day later than 'Mountrail', 'Tioga' and 'Divide', two days later than 'Joppa' and five days later than 'Alzada' (Table 1). Lustre has an erect flag leaf and an erect tapering head. It has 0.2% or 20 in 10,000 spikes that are taller.

Field Performance

Under all locations and conditions (2017-2019, 24 location-years, no off-station) Lustre (5,030 kg ha^{-1}) yielded significantly more than the top grown cultivars ‘Divide’ (4,889 kg ha^{-1}), ‘Joppa’ (4,788 kg ha^{-1}), ‘Tioga’ (4,849 kg ha^{-1}), and ‘Alzada’ (4,439 kg ha^{-1}) (Table 1). Under only dryland conditions (2017-2019, 17 location-years, no off-station) Lustre (4,102 kg ha^{-1}) yielded significantly higher than ‘Divide’ (3,894 kg ha^{-1}), ‘Joppa’ (3,853 kg ha^{-1}), ‘Tioga’ (3,867 kg ha^{-1}), and ‘Alzada’ (3,712 kg ha^{-1}) (data not shown). Averaged over EARC, WTARC, CARC, NARC dryland environments (2017-2019, 12 location-years, no off-station) Lustre was the highest yielding line (3,457 kg ha^{-1}) which was significantly higher than ‘Divide’ (3,147 kg ha^{-1}), ‘Joppa’ (3,114 kg ha^{-1}), ‘Tioga’ (3,166 kg ha^{-1}), and ‘Alzada’ (3,168 kg ha^{-1}) (data not shown). Over three years at EARC dryland (2017-2019, no off-station) Lustre had an average yield (4,412 kg ha^{-1}) higher than ‘Divide’ (4,136 kg ha^{-1}), ‘Joppa’ (3,948 kg ha^{-1}), ‘Tioga’ (4,277 kg ha^{-1}), and ‘Alzada’ (3,867 kg ha^{-1}), however no significant differences were detected (data not shown). At CARC, NARC, and WTARC (2017-2019, 9 location-years), Lustre was the second highest yielding line (3,134 kg ha^{-1}) which was significantly higher than ‘Divide’ (2,825 kg ha^{-1}), ‘Joppa’ (2,831 kg ha^{-1}), ‘Tioga’ (2,804 kg ha^{-1}), and ‘Alzada’ (2,932 kg ha^{-1}) (data not shown). Lustre’s grain protein is similar to the top grown cultivars ‘Divide’, ‘Joppa’, and ‘Alzada’ (Table 1). The grain volume weight of Lustre is similar to ‘Mountrail’ and ‘Alzada’ but less than ‘Divide’ and ‘Joppa’ (Table 1), and Lustre’s individual kernel size and weight is similar to other durum cultivars with the exception of ‘Alzada’ which has larger kernels (Table 1.).

End-use quality characteristics

Lustre (5.0) has improved protein strength based on mixograph scores compared to ‘Mountrail’ (3.1), but comparable to ‘Divide’ (5.1) and lower than ‘Joppa’ (5.8) and ‘Alzada’ (6.8) (Table 2).

Gluten index was measured in 2019 from Bozeman dryland and NARC dryland samples. Lustre (57) had a statistically similar gluten index to ‘Divide’ (74) that was higher than ‘Mountrail’ (32) but lower than ‘Alzada’ (95) (data not shown). The average semolina yield of Lustre (61.7 %) falls within the range of currently grown cultivars as does its semolina brightness (L) and yellowness (b*) (L=84.7, b*=28.6) (Table 2). Semolina from Lustre had higher protein content (135 g kg^{-1}) than ‘Divide’, ‘Joppa’, and ‘Alzada’ and an ash content similar to ‘Divide’ and ‘Joppa’, but lower than ‘Alzada’ (Table 2). In 2017, whole grain samples from EARC dryland experiments were evaluated for cadmium accumulation. Compared to lines with the high Cd accumulation allele (avg=0.17 ppm) Lustre had half the amount of Cd present (0.08 ppm) as expected due to the presence of the low accumulation allele (Table 3).

Disease resistance and tolerance to acidic soils

Lustre was evaluated for resistance/susceptibility to stripe rust at Mt. Vernon, WA and Pullman, WA over two years. Pullman measurements were collected on July 17 of both years, and measurements at Mt. Vernon were collected on both June 7 and June 27. In both years, lines were evaluated for infection type (IT) on a scale from 0 to 9, where 0-3 are considered resistant, 4-6 intermediate, and 7-9 susceptible. Severity was recorded as a percentage. In both years, the susceptible check variety had an IT of 8 and an average severity of 95-100%. Lustre has a similar stripe rust reaction as the top grown check cultivars, with susceptibility at the seedling stage and high-temperature adult-plant resistance. At Pullman, Lustre had average IT of 3.5 with 10% severity. At Mt. Vernon, Lustre had an average IT of 6 with 55% severity early in growth and IT of 3 with 25% severity later in growth. Fusarium head blight susceptibility was evaluated in 2018 and 2019 at EARC. Lustre performed similarly as other susceptible top grown cultivars in terms of severity, fusarium damaged kernels and DON (data not presented). Lustre showed all-stage resistance to leaf rust isolate PBJJG-MSU09 and stem rust race TCMLK, the most prevalent race in Montana.

In 2019, Lustre was evaluated for leaf spot caused by naturally occurring *Stagonospora nodorum* and/or *Pyrenophora tritici-repentis* in Bozeman under irrigated and dryland conditions. Leaf spot disease was evaluated on a scale of 0-5 (0 = no visible leaf spot symptoms, 1 = 10%, 2 = 25%, 3 = 50%, 4 = 75%, 5 = 100% upper canopy leaves with symptoms). In both environments Lustre had moderate resistance to leaf spots (1.7 dryland, 2.1 irrigated) with more symptoms than ‘Divide’ or ‘Joppa’ (0 dryland and 0 irrigated), but significantly less than ‘Alzada’ which was very susceptible (4 dryland, 4.3 irrigated). Lustre was tested by Washington State University for tolerance to acidic soils and was determined to be as susceptible as currently grown varieties.

AVAILABILITY

Lustre contains a patented trait conferring improved resistant starch and pasta quality. Use of Lustre requires a Material Transfer Agreement (research use only) or a commercial license to the trait, as well as permission from the originator (Montana Agricultural Experiment Station). Seed requests may be sent to the corresponding author. Seed of Lustre has been deposited in the USDA-ARS National Plant Germplasm System, where it will be available for distribution after the expiration of applicable patents and Plant Variety Protection. Seed of Lustre will be maintained by the Montana State University Foundation Seed Program who will also collect fees from any seed sales. Application has been made for U.S. Plant Variety Protection with the certification option (____).

ACKNOWLEDGEMENTS

SUPPLEMENTAL MATERIAL

CONFLICT OF INTEREST

The authors declare no conflict of interest.

FUNDING INFORMATION

Funding for the development of Lustre was provided by the Montana Wheat and Barley Committee, the Montana Research and Economic Development Initiative, the Montana Agricultural Experiment Station and by Dow AgroSciences.

REFERENCES

American Association of Cereal Chemists. (2010) Approved Methods of Analysis. 11th ed. AACC International, St. Paul, MN.

Clarke, J.M., Leisle, D., Kopytko, G.L. (1997) Inheritance of cadmium concentration in five durum wheat crosses. Crop Sci. 37:1722–1725.

Codex Alimentarius General Standard for Contaminants and Toxins in Food and Feed (accessed November 3, 2020) CXS 193-1995 (Amended 2019). Retrieved from http://www.fao.org/fao-who-codexalimentarius/sh-proxy/en/?lnk=1&url=https%253A%252F%252Fworkspace.fao.org%252Fsites%252Fcodex%252FStandards%252FCXS%2B193-1995%252FCXS_193e.pdf.

Elias, E.M. and Miller, J.D. Registration of 'Mountrail' Durum Wheat (2000) Crop Sci. 40:1499-1500.

Elias E.M. and Manthey F., (2007) Registration of 'Divide' durum wheat. J. Plant Regist. 1:7–8.

Grant, C.A., Buckley, W.T., Bailey, L.D., Selles F., (1998) Cadmium accumulation in crops. Can. J. Plant Sci. 78:1–17.

Hazard, B., Zhang, X., Mahmoudreza, N., Hamilton, M.K., Rust, B., Raybould, H.E., … Dubcovsky, J. (2015) Mutations in durum wheat *SBEII* genes affect grain yield components, quality, and fermentation response in rats. Crop Sci. 55:2813-2825.

Hogg, A.C., Gause, K., Hofer, P., Martin, J.M., Graybosch, R.A., Hansen, L.E., Giroux, M.J. (2013) Creation of a high-amylose durum wheat through mutagenesis of starch synthase II (*SSIIa*). J. Cereal Sci. 57:377-383.

Hogg, A.C., Martin, J.M., Manthey, F.A., Giroux, M.J. (2015) Nutritional and quality traits of pasta made from *SSIIa* null high-amylose durum wheat. Cereal Chem. 92:395-400.

Martin, J.M., Hogg, A.C., Hofer, P., Manthey, F.A., Giroux, M.J. (2012) Impacts of *SSIIa-A* null allele on durum wheat noodle quality. Cereal Chem. 91:176-182.

Menke, A., Muntner, P., Silbergeld, E.K., Platz, E.A., Guallar, E., (2009) Cadmium levels in urine and mortality among U.S. adults. Environ. Health Perspect. 117: 190–196.

Nishijo, M., Morikawa, Y., Nakagawa, H., Tawara, K., Miura, K., et al… (2006) Causes of death and renal tubular dysfunction in residents exposed to cadmium in the environment. Occup. Environ. Med. 63:545–550.

Olsson, I.M., Eriksson, J., Oborn, I., Skerfving, S., Oskarsson, A., 2005. Cadmium in food production systems: a health risk for sensitive population groups. Ambio 34:344–351.

Perrier, F., Yan, B., Candaudap, F., Pokrovsky, O.S., Gourdain, E., Meleard, B., Bussière, S., Coriou, C., Robert, T., Nguyen, C., Cornu, J.Y. (2016) Variability in grain cadmium concentration among durum wheat cultivars: impact of above ground biomass partitioning. Plant Soil 404:307−320.

Regina, A., Bird, A., Topping, D., Bowden, S., Freeman, J., Barsby, T., Kosar-Hashemi, B., Li, Z., Rahman, S., Morell, M. (2006) High-amylose wheat generated by RNA interference improves indices of large-bowel health in rats. Proc. Natl. Acad. Sci. U.S.A. 103(10):3546-3551.

Sissions, M., Sestili, F., Botticella, E., Masci, S., Lafiandra, D. (2020) Can manipulation of durum wheat amylose content reduce the glycaemic index of spaghetti? Foods 9(6):693.

FIGURES AND TABLES

Table 1. Agronomic performance of Lustre and check cultivars from Montana Intrastate Durum Variety Trials (2017-2019).

Cultivar	Grain yield[a]	Grain volume weight	Heading date[b]	Plant height[b]	Grain protein[c]	Individual seed weight	Individual seed diameter
	$kg\ ha^{-1}$	$kg\ m^{-3}$	d from 1 Jan.	cm	$g\ kg^{-1}$	mg	mm
Mountrail	4,930	775	178	86.4	145	40.3	2.8
Divide	4,889	783	178	89.9	146	41.6	2.8
Tioga	4,849	780	178	92.7	149	42.8	2.9
Joppa	4,788	785	177	89.7	143	41.4	2.8
Alzada	4,439	776	174	72.1	146	44.8	3.0
Lustre	5,030	772	179	89.4	146	41.0	2.8
CV (%)	6.3	1.1	0.5	3.7	2.9	3.9	2.0
LSD (0.05) [d]	181	5	1	2.3	2.0	0.9	0.03
Prob > F[d]	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001
Location-years	24	24	17	17	24	24	24

[a]Grain yield reported on a 13% moisture basis.
[b]Data from dryland locations only
[c]Grain protein reported on a 12% moisture basis.
[d]Fisher's unrestricted LSD at the 0.05 probability level. Significance determination is based on the inclusion of the following entries: Alkabo, Grenora, Carpio, CDC-Dynamic, CDC-Fortitude, CDC-Precision, CDC-Vivid, MTD16001-11.

Table 2. Quality performance of Lustre and check cultivars over 24 location-years from Montana Intrastate Durum Variety Trials (2017-2019).

Cultivar	Large seeds	Small seeds	Hardness index	Grain ash[a]	Falling number	Bran	Shorts	Semolina	Semolina protein[a]	Semolina ash[a]	L[b]	*b**[b]	*a**[b]	Mixograph pattern
	%	%		%	sec	%	%	%	g kg^{-1}	%				
Mountrail	44.9	15.5	74.5	1.46	430.5	27.7	10.1	62.2	135	0.64	85.2	25.7	-2.7	3.1
Divide	58.1	11.4	73.7	1.41	420.9	27.7	10.1	62.2	132	0.60	85.1	27.3	-2.9	5.1
Tioga	62.3	10.1	73.0	1.49	412.7	27.7	10.1	62.2	138	0.62	84.9	29.3	-3.1	6.0
Joppa	56.9	11.6	76.2	1.46	420.1	27.8	10.4	61.8	132	0.61	85	29.4	-3.1	6.0
Alzada	75.6	5.5	74.5	1.52	445.3	26.0	11.8	62.1	133	0.67	84.2	30.8	-3.0	6.8
Lustre	50.0	12.4	74.6	1.41	433.4	27.6	10.7	61.7	135	0.60	84.7	28.6	-2.9	5.0
CV (%)	12.6	28.0	3.0	3.8	5.3	2.9	3.7	1.5	3.4	5.0	0.4	2.6	4.2	12.7
LSD (0.05)[c]	4.0	1.8	1.3	0.03	12.8	0.5	0.2	0.5	3.0	0.02	0.2	0.4	0.1	0.4
Prob > F [c]	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001

[a] Reported on a 14% moisture basis.

[b] Color measurements L=whiteness, *b**=yellowness, *a** =redness

[c]Fisher's unrestricted LSD at the 0.05 probability level. Significance determination is based on the inclusion of the following entries: Alkabo, Grenora, Carpio, CDC-Dynamic, CDC-Fortitude, CDC-Precision, CDC-Vivid, MTD16001-11.

Table 3. Elemental cadmium analysis of whole grain for EARC dryland 2017

ID	*Cdu1* allele[a]	Cd[b] (mg kg^{-1})
Divide	High	0.17
MTD16001	High	0.12
MTD16002	Low	0.05
MTD16004	High	0.16
Lustre	**Low**	**0.08**
MTD16006	Low	0.07
MTD16007	Low	0.09
MTD16008	Low	0.06
MTD16009	High	0.13
MTD16010	High	0.14
MTD16011	Low	0.07

[a]*Cdu1*- gene controlling cadmium accumulation either the high or low accumulation allele.
[b]Single observation of bulked replications. European import standards are set to 0.2 mg kg^{-1} Cd (Codex Alimentarius CDX 193-1995, Amended 2019)