



Winter survival response of canola to meteorological variables and adaptative areas for current canola germplasm in the United States

Mario A. Secchi^{a,*}, Leonardo M. Bastos^a, Michael J. Stamm^a, Yancy Wright^b, Chris Foster^b, Carlos D. Messina^c, Ignacio A. Ciampitti^{a,*}

^a Department of Agronomy, Kansas State University, 2004 Throckmorton Plant Sciences Center, Manhattan, Kansas 66506, US

^b John Deere, Johnston, IA 50131, US

^c Corteva Agriscience, Research and Development, Johnston, IA 50131, US

ARTICLE INFO

Keywords:

Brassica napus
Rapeseed
Environmental factors
Cold temperature
Oilseed rape
Winter kill

ABSTRACT

The introduction of winter canola (*Brassica napus* L.) into rotations with winter wheat (*Triticum aestivum* L.) in the United States (US) revealed economic and agronomic benefits as well as improved weed and pest control. Canola stand establishment during the fall and plant survival over the winter are critical to the success of this crop. The environment plays a key role influencing survival, but in-depth evaluation of meteorological factors has not been conducted. This research study aimed to: i) identify meteorological factors underpinning winter canola survival, ii) build probabilistic response models based on historical meteorological data for different severities of winter kill across the US, and iii) define areas of adaptation of current germplasm. A winter survival dataset was compiled from the National Winter Canola Variety Trials from 2003 until 2018 (190 site-years) and auxiliary meteorological data over the last 40 years. A regression tree analysis indicated that meteorological variables related to minimum temperature, fluctuating temperatures above and below 0°C, and windchill during the cold period were the main factors accounting for winter kill. Cold periods across all site-years were classified into three clusters: cold periods with high (96%), medium (70%), and low (28%) average plant winter survival. For 94 US sites, the probabilities of these conditions were calculated and summarized in a map that defined areas of adaptation: a large area south of 35° N latitude for the US was identified with greater potential for overwintering success. Based on the response under multiple meteorological conditions, four distinct genotype survival groups were identified (tolerant, semi-tolerant, semi-susceptible, and susceptible). Groups with a greater number of genotypes differ in the impact of meteorological conditions on survival for the medium cluster. In regions with more favorable conditions for overwintering success, farmers may be open to introducing this crop to diversify their farming system.

Introduction

The United States produced 3.6 million tons of canola (*Brassica napus* L.) in 2019, representing 5.2% of global production (USDA-NASS, 2019; FAO, 2020). Although the state of North Dakota harvested 83% of US canola as spring canola (USDA-NASS, 2019), other southern US states have the potential to introduce winter canola, also known as winter oilseed rape or double-low rapeseed, where winter wheat (*Triticum aestivum* L.) is the only option as a winter crop or as a service crop to diversify the current maize (*Zea mays* L.)-soybean (*Glycine max* [L.] Merr) farming system. Since the 1970s, a loss of genetic diversity of crops has been reported as a result of the Green Revolution (Jacques and

Jacques, 2012). Inclusion of winter canola into farming systems with winter wheat, as opposed to mono-cropping, has proven to break weed and pest cycles (Bushong et al., 2012). Increasing crop diversity in current agricultural systems can be the only path to meeting sustainability goals (Renard and Tilman, 2019).

Fall stand establishment and winter survival have been suggested as key limiting factors to the success of winter canola production (Assefa et al., 2014). Broadleaf winter survival is a complex trait, and several stresses influence this process such as prolonged exposure to subzero temperatures, ice encasement, diseases, wind desiccation, and soil heaving (Levitt, 1956). Plant tolerance to environmental stresses decreases as resources necessary for plant survival become depleted

* Corresponding author at: Department of Agronomy, Kansas State University, 2004 Throckmorton Plant Sciences Center, Manhattan, Kansas 66506, United States.
E-mail addresses: secchi@ksu.edu (M.A. Secchi), ciampitti@ksu.edu (I.A. Ciampitti).

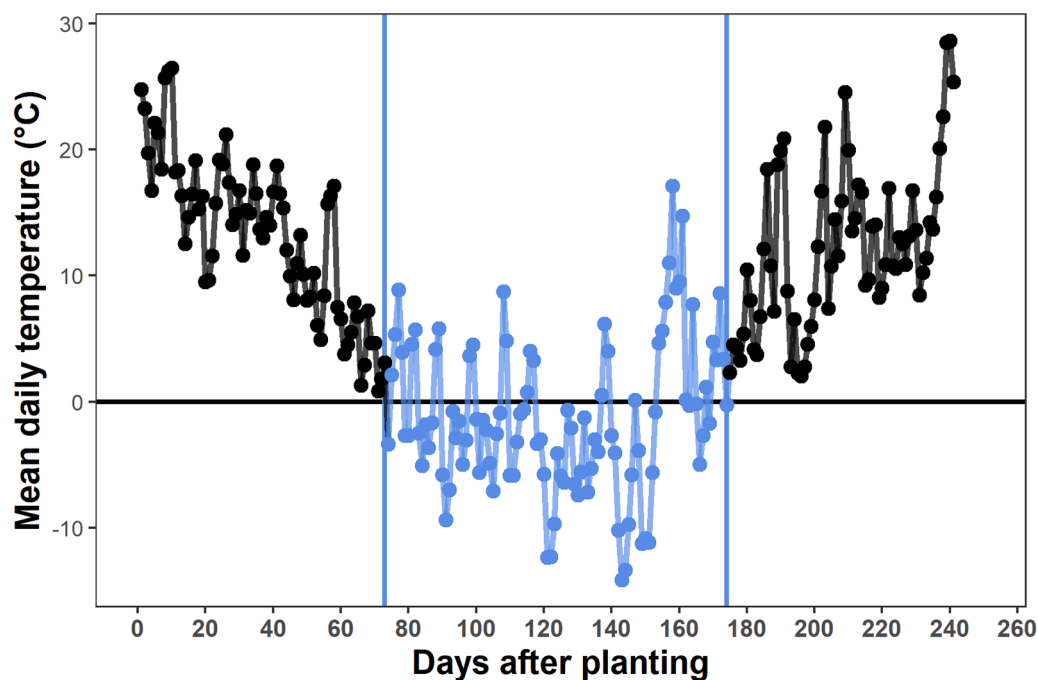


Fig. 1. Example of the cold period. Mean daily temperature (°C) since planting to harvest at Manhattan, KS (2010-2011 season). Blue points represent the cold period. Vertical blue bars represent the beginning and end of cold period as the first and last time that mean daily temperature was below 0°C, respectively.

throughout the winter (Gusta et al., 1982). Temperature and precipitation during the winter period are critical for effective winter survival (Fowler et al., 1981; Waalen et al., 2013). More in-depth studies for this trait in winter canola were carried out in Europe. Salmon (1918) identified temperature and soil moisture as critical factors for winter kill of grain crops. Rainfall and temperature during the winter season, which can vary dramatically among years and sites, are key environmental variables in current farming systems (Assefa et al., 2014). Although there is no clear understanding of genotype by environment interaction effects for winter survival in the US, producers have concerns regarding low plant establishment and winter survival (George et al., 2012; Stamm and Watson, 2013).

A significant breeding effort has increased the winter hardiness of canola genotypes (Rife et al., 2001; Stamm et al., 2015). Because of the complexity of the trait, a comprehensive understanding of how meteorological factors impact winter survival and yield should be pursued to facilitate winter canola production in new areas. This understanding will facilitate breeding to expand the area of adaptation and production and could be integrated with whole-genome prediction methodology (Messina et al., 2018). While a few site-specific winter survival analyses were conducted (Holman et al., 2011; Waalen et al., 2013), a comprehensive synthesis of meteorological variation in the US and its influence on this critical plant trait is lacking. Building foundational knowledge of crops such as canola and other grain, oil, or service crops will be required to transform current (undiversified) farming systems.

The overall objective of this work is to determine the area of adaptation of current winter canola germplasm based on winter survival.

This knowledge will facilitate crop diversification, breeding efforts, and genetic evaluation that may feed the expansion of the area of adaptation. The specific aims of this research study were to: i) model winter canola survival based on meteorological factors, ii) build probabilistic response models based on historical meteorological data for different severities of winter kill, and iii) define areas of adaptation of current germplasm across the US. A large dataset compiled from the National Winter Canola Variety Trials (NWCVT), conducted from 2003 to 2018 across 94 unique sites with auxiliary weather data for each site-year were assembled and analyzed.

Materials and Methods

Data and Predictors

Field trials

The field dataset was curated from the National Winter Canola Variety Trial (NWCVT) (Stamm et al., 2019). The purpose of this national network of trials is to evaluate canola winter survival, yield performance, and other important agronomic traits on multiple varieties across various US states. The trials also aim to find suitable areas of adaptation of new genotypes and increase the visibility of winter canola across the country. The field dataset comprises 94 sites covering 23 US states, spanning 2003 through 2018, for a total of 333 site-years. Only site-years with information on winter survival were included, decreasing the number of sites and site-years to 54 and 190, respectively. The experimental design for each trial defined by site and year was a

Table 1
Description of secondary daily weather variables.

Variable name	Unit	Abbreviation	Definition
Mean temperature	°C	Tmean	(Tmax + Tmin)/2,
Delta temperature	°C	DeltaT	Tmax - Tmin
Growing degree days	°C.d.	GDD	(Tmax + Tmin)/2; if Tmax > 30°C, then Tmax=30; if Tmin < 0°C, then Tmin=0.

Tmax and Tmin are the maximum and minimum air temperature, in°C, respectively.

Table 2

Description of meteorological variables during the CP (mean, cumulative, minimum, maximum, and counts).

Variable name	Unit	Abbreviation	Definition
Cold period duration	days	N	Number of days between beginning and end of cold period
Number of temperature cycles	count	ncycle	Number of times Tmean shifts from negative to positive or vice-versa
Slope	°C	-	Slope between cumulative GDD vs. days after planting
Tmean descriptors*			
Warmest, Warmest_pct	count, %		Number of times Tmean >= 5°C, Warmest/N
Warm, Warm_pct	count, %		Number of times 0°C <= Tmean < 5°C, Warm/N
Mild, Mild_pct	count, %		Number of times -5°C <= Tmean < 0°C, Mild/N
Cold, Cold_pct	count, %		Number of times -10°C <= Tmean < -5°C, Cold/N
Colder, Colder_pct	count, %		Number of times -15°C <= Tmean < -10°C, Colder/N
Coldest, Coldest_pct	count, %		Number of times Tmean < -15°C, Coldest/N
DeltaT descriptors*			
Extreme, Extreme_pct	count, %		Number of times DeltaT >= 16.7°C, Extreme/N
High, High_pct	count, %		Number of times 13.4°C <= DeltaT < 16.7°C, High/N
Medium, Medium_pct	count, %		Number of times 10°C <= DeltaT < 13.4°C, Medium/N
Low, Low_pct	count, %		Number of times DeltaT < 10 °, Low/N
Minimum daily mean temperature	°C	minTmean	-
Mean daily mean temperature	°C	meanTmean	-
Maximum daily mean temperature	°C	maxTmean	-
Minimum daily minimum temperature	°C	minTmin	-
Maximum daily maximum temperature	°C	maxTmax	-
Minimum delta temperature	°C	minDeltaT	-
Mean delta temperature	°C	meanDeltaT	-
Maximum delta temperature	°C	maxDeltaT	-
Mean wind velocity at 10 m	m s ⁻¹	meanWind	-
Mean wind chill [#]	°C	meanWindchill	$13.12 + 0.6215 \times Tmean - 11.37 \times W^{0.16} + 0.3965 \times Tmean \times W^{0.16}$
Cumulative precipitation	mm	cPrecip	-
Cumulative reference (alfalfa) evapotranspiration	mm	cET	-
Cumulative solar radiation	W m ⁻²	cSolar	-
Cumulative vapor pressure deficit	kPa	cVPD	-
Cumulative canola growing degree days	°C.d.	cGDD	SUM (Tmax +Tmin)/2

Tmean is the average between maximum and minimum daily temperature, in °C. DeltaT is the difference between maximum and minimum daily temperature, in °C.

* Tmean and DeltaT descriptors were calculated both as the number of days during winter within a given conditional statement, and as a percentage of this count in relation to total days of winter duration.

[#] W is the daily averaged wind velocity at 10 m, in km h⁻¹

randomized complete block with three or four replications. The number of genotypes included in any given site and the number of replications varied depending on the site and year. Genotypes included in each site and year changed based on maturity and year of commercialization. Winter survival ratings were measured after dormancy was completed, or approximately when the average daily temperature exceeded 4°C. This coincides with the time period of rapid, new leaf development, just before the plant enters the reproductive phase at bolting, and when the threat of further losses is low. Winter survival is a visual estimate of the percentage of plants alive in spring relative to those present before the winter period (Stamm et al., 2012). Based on field conditions up to the point of rating winter survival, the plants that do not initiate new leaf growth are observed as lost to winter kill.

Weather dataset

Daily weather data were extracted from Google Climate Engine (Huntington et al., 2017) for the period 1979-2019 (40 years) for each site. The variable set included daily minimum and maximum temperatures (°C), precipitation (mm), wind velocity (m s⁻¹), solar radiation (W m⁻²), vapor pressure deficit (kPa), and evapotranspiration (mm) calculated using alfalfa (*Medicago sativa* L.) as reference. The daily weather data was utilized to: i) create secondary weather variables to characterize winter harshness; ii) identify the beginning and end of the cold period (CP); iii) build a model to predict meteorological survival clusters with meteorological variables during the CP, and iv) predict meteorological survival clusters and calculate the probability of occurrence over 40 years. For each site-year, secondary daily weather variables were created by calculating means and difference between maximum and minimum for temperature (°C) and growing degree days (°C.d.) (Table 1). Mean temperature was then utilized to identify the beginning and end of the CP as the period when temperatures were

below 0°C. An example of the mean daily temperatures preceding, during, and after the CP for the Manhattan, KS, 2010-2011 growing season is shown in Fig. 1.

Predictors

After the CP was determined for each site-year, the meteorological data was filtered by date to contain only information within this period. Secondary summarized meteorological variables were calculated including CP duration days (N), number of times when mean temperature shifts from negative to positive or vice-versa (ncycle), slope between cumulative GDD (with minimum base temperature of 0°C and maximum temperature of 30°C) and days after planting (Slope), and class descriptors for mean temperature (Table 2). All daily variables were summarized by averaging, summing, extracting minimum and maximum values, or counting within the CP (Table 2). Windchill calculations were performed following the equation presented by Oszczewski and Bluestein (2005).

Analyses

A series of analyses divided into seven steps were conducted to estimate the contribution of genotype (G), environment (E), and interaction (GxE) to the overall variance for canola winter survival (S). Because S was not normally distributed, we used a log transformation. Once variance components were estimated (step 1), a mixed linear model was utilized to estimate the E and GxE effects (step 2). Predictions from this model, Best Linear Unbiased Estimators (BLUEs) for E effects, and Best Linear Unbiased Predictors (BLUPs) for the GxE effects were produced by solving for model-based E marginal means and GxE variance components, respectively. Environment BLUEs for S were transformed into

survival classes using cluster analyses (step 3). These categorized data were used in modeling meteorological survival class as a function of meteorological variables (step 4) using conditional inference. Step 5 used the model developed in step 4 to simulate meteorological survival class as a function of meteorological predictors and classify each site-year combination for the risk level of *S*. Step 6 repeats G×E analyses using meteorological survival class as predictors rather than site-year as factor. BLUEs from step 6 were transformed into 4 genotype survival classes. A final mixed model was used to model *S* as a function of *E* and *G* survival classes and their interaction (step 7).

In step 1 we modeled the data using a random-effect model (equation 1) ($n=23,225$),

$$\log(S) = \bar{G}_i + \bar{E}_j + \overline{GE}_{ij} + \varepsilon_{ijk} \quad (1)$$

where all terms were considered random effects with error $N \sim (0, \sigma)$ and a general symmetric positive-definite variance-covariance matrix structure. *E* here is defined by the site-year identifier.

Step 2 used a mixed model (equation 2),

$$\log(S) = \bar{G}_i + E_j + \overline{GE}_{ij} + \varepsilon_{ijk} \quad (2)$$

where *E* was considered a fixed effect, and *G* and G×E considered random effects. *G* was considered a random effect as the inference is over a sample of genotypes and not any genotype in particular. *E* here is defined by the site-year identifier.

In step 3, environment BLUEs for survival, on the response scale, ($n=190$) were clustered into three groups: poor, medium, and high; using *k* means algorithm. The optimal number of clusters (i.e., $k=3$) was

selected by testing *k* values from 1 to 10 and choosing the one with the most votes from 30 different indices. The categorical meteorological survival class (MS_c) was integrated with meteorological predictors as described above.

In step 4 we used the dataset containing 39 summarized meteorological predictors (Table 2), to model meteorological survival class MS_c ($n=190$) as a function of meteorological factors using conditional inference tree methodology. The modeling process included model parameterization and model fitting. During model parameterization, the model with the best values for the hyperparameters of maximum depth and alpha was found by performing leave-one-out cross-validation. Maximum depth controls the number of horizontal node layers of the tree, and alpha controls the significance level for a variable to be selected to enter the tree. Maximum depth values of 2, 3, 4, and 5 were evaluated along with alpha values of 0.01, 0.02, 0.03, 0.04, 0.05, and 0.1. Hyperparameter values of maximum depth=4 and alpha=0.1 were then chosen based on overall classification accuracy. Thereafter, model fitting was conducted using all the data and the hyperparameters were calculated in model parameterization. Leave-one-out cross-validation was conducted to estimate the model performance metrics of overall and category-specific accuracy, sensitivity, and specificity. Using the classification tree was possible to predict meteorological survival class based on meteorological covariates (MS_{ce}).

Step 5 utilized long-term daily weather records (1979-2019 period) for each of the 94 sites included in the multi-environment trials to estimate the area of adaptation of current canola germplasm based on the risk of survival. The long-term meteorological data was processed in the

Table 3
Summary statistics for meteorological variables during the cold period.

Variable	Unit	Minimum	Mean	Median	Maximum	CV (%)
Survival	%	0	83.9	96.2	100	31.4
N	days	11	111.4	109	257	31.8
ncycle	count	5	23.3	23	61	39.2
Slope	°C	0.2	3	2.6	10.5	64.4
Warmest	count	0	28.9	28	106	57
Warmest_pct	%	0	0.3	0.3	0.7	59.1
Warm	count	2	32.9	30	100	41.7
Warm_pct	%	0.1	0.3	0.3	0.5	27.6
Mild	count	3	31.5	32	75	46.2
Mild_pct	%	0.1	0.3	0.3	0.5	36
Cold	count	0	12.9	11	47	73.7
Cold_pct	%	0	0.1	0.1	0.3	70.7
Colder	count	0	4.1	3	33	125
Colder_pct	%	0	0	0	0.2	172.8
Coldest	count	0	1.2	0	25	266
Coldest_pct	%	0	0	0	0.2	563.8
Extreme	count	0	21.2	12	86	103
Extreme_pct	%	0	0.2	0.1	0.7	101.1
High	count	2	25.5	22	75	62.1
High_pct	%	0	0.2	0.2	0.5	47.8
Medium	count	1	29.8	28	76	46.6
Medium_pct	%	0	0.3	0.3	0.5	36.5
Low	count	0	34.9	27	130	79.7
Low_pct	%	0	0.3	0.3	0.8	66.4
minTmean	°C	-24.9	-12.6	-12.6	-2.3	-37.7
meanTmean	°C	-7.2	1.3	1	9.1	220.7
maxTmean	°C	4.1	14.5	14.5	21.9	22.5
minTmin	°C	-31.1	-18.4	-18.2	-6.9	-27.2
maxTmax	°C	6.6	22.7	22.5	34.5	17.9
minDeltaT	°C	0.5	4.4	4.3	12.4	46.3
meanDeltaT	°C	7.1	12.5	12.5	19.1	21.1
maxDeltaT	°C	15.1	22.3	21.5	33.9	17.7
meanWind	m s ⁻¹	2.5	4.2	4.3	5.9	13.9
meanWindchill	°C	-12.9	-2.5	-2.7	7.5	-138.6
cPrecip	mm	1.8	149	107.6	608.7	82.3
cET	mm	29.9	270.6	245.7	701.3	44
cSolar	W m ⁻²	1423.7	12478.9	11736.4	27764.7	36.5
cVPD	kPa	4.1	40.6	35.8	96	47
cGDD	°C.d.	13.1	333.4	307.2	1161.7	53.8

CV=coefficient of variation.

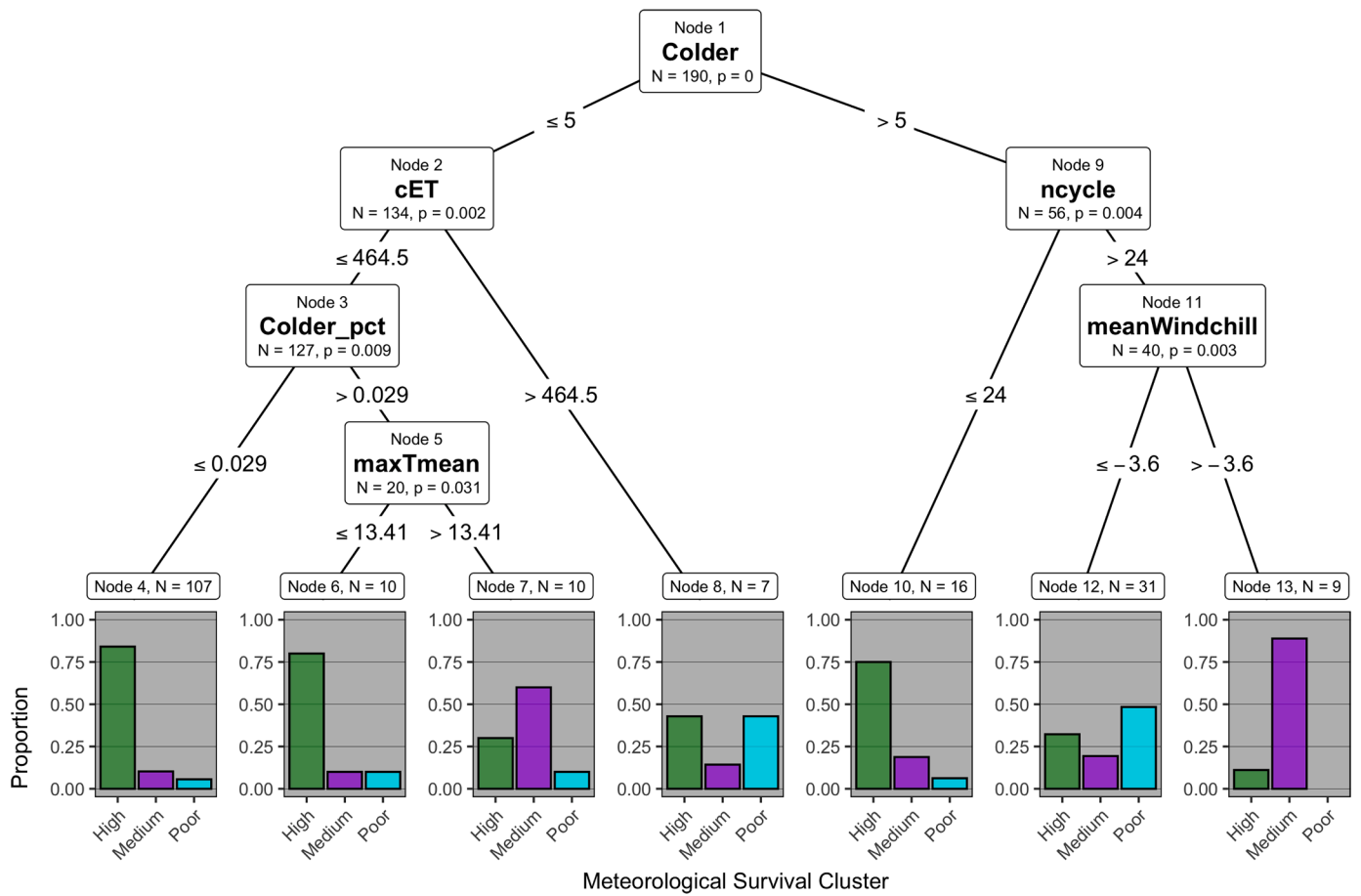


Fig. 2. Conditional inference tree of canola meteorological survival clusters as explained by cold period-summarized meteorological variables from 190 sites-years classified into seven terminal nodes. Terminal node bars represent the proportion (left y-axis) of site-years within each meteorological survival cluster (from left to right at each node: high, medium, and poor) at that node.

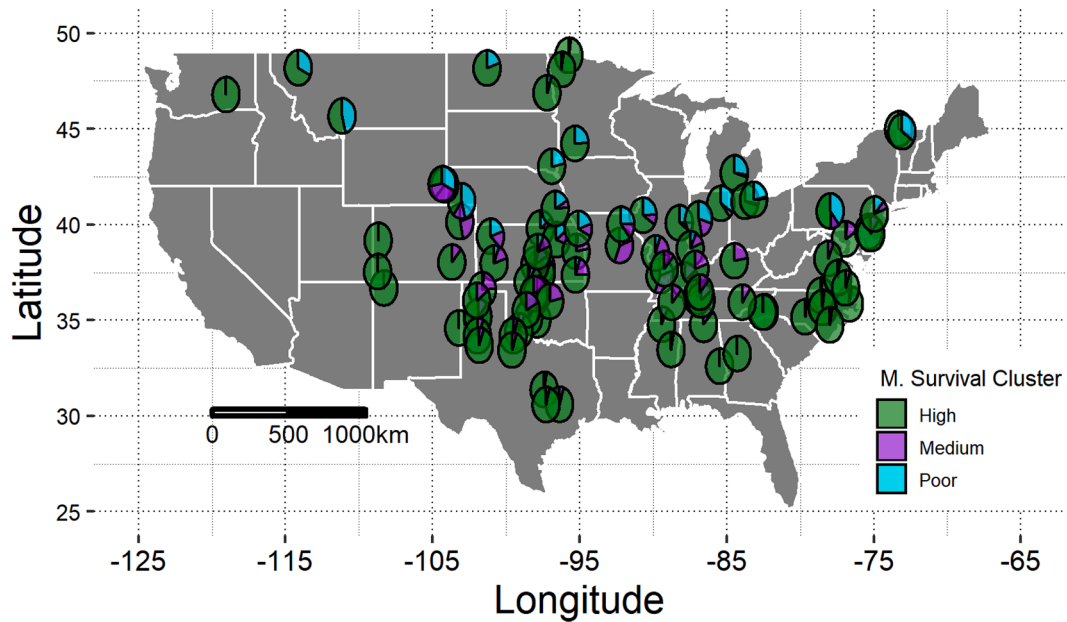


Fig. 3. US map with the sites included in the National Winter Canola Variety Trial dataset. Each pie chart represents a site with the slices representing the proportion of cold periods classified as high, medium, and poor meteorological survival clusters over a period of 40 years (1979-2019).

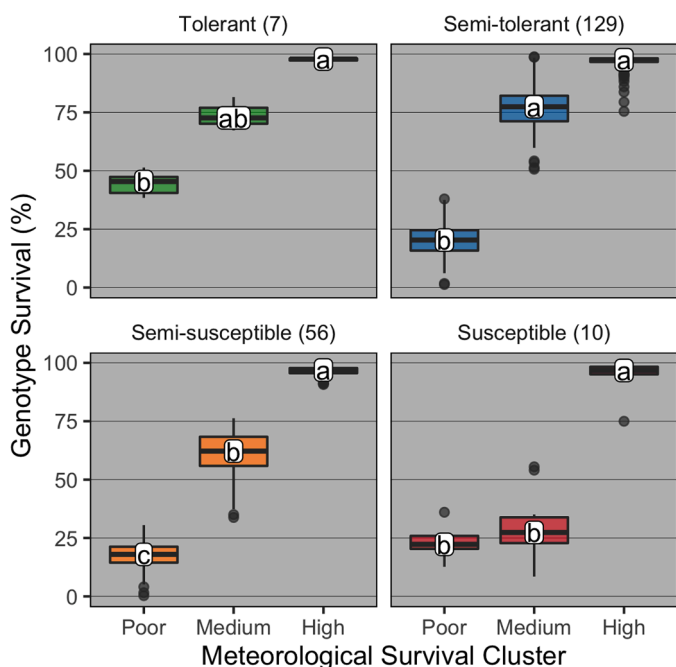


Fig. 4. Boxplots of winter survival of 202 canola genotypes across three different meteorological survival clusters (poor, medium, and high) separated into four distinct genotype survival groups (tolerant, semi-tolerant, semi-susceptible, and susceptible). Boxplots portray the 25th (lower hinge), 50th (solid black line), and 75th (upper hinge) percentiles, largest value no further than 1.5 inter-quartile range (lower whisker), smallest value at most 1.5 inter-quartile range (upper whisker), and outlying observations (points). Boxplots within a panel followed by the same letter are not statistically different at alpha=0.05. Total number of genotypes within each panel is shown in parenthesis.

same manner as described under *Predictors*. Only years with a CP (mean temperature below 0°C) were kept, causing the number of years per site to vary between 19 and 40. In this simulation step, the trained tree model was used to predict the CP of each site-year (MS_c) into a risk class (S_r), poor, medium, and high, based on the simulation for all years for each site. The original data set (n=23,225) was integrated with predictions for MS_c . Only genotypes present at least once in each of poor,

medium, and high meteorological survival clusters were kept (n=19,919).

In step 6, a mixed-effect model was fitted to the data (equation 3),

$$\log(S) = G_i + MSC_j + GMS_{c_{ij}} + \bar{E}_k + \bar{E}b_{kl} + \epsilon_{ijklm} \quad (3)$$

with G and MS_c defined as survival class and their interaction $G \times MS_c$ considered a fixed effect, and block (b) nested within the site-year random effect. Pairwise comparisons on BLUEs for genotype by MS_c interaction were performed across meteorological survival clusters using alpha=0.05. Based on the pairwise comparison letter separation result, four distinct genotype survival behaviors across meteorological survival clusters were identified, hereafter referred to as genotype survival group GS_g .

Step 7 used a mixed-effect model (equation 4),

$$\log(S) = GSg_i + MSC_j + GSgMS_{c_{ij}} + \bar{E}_k + \bar{E}b_{kl} + \bar{E}G_{km} + \epsilon_{ijklm} \quad (4)$$

where GSg_i , MS_c_j and their interaction were fixed effects, and block (b) and genotype nested within the site-year random effect. Pairwise comparisons for $GSg_i \times MS_c_j$ were performed within MS_c_j using alpha=0.05.

Software

All analyses were conducted within the R framework (R Core Team, 2019). Mixed model analyses were solved using the function lmer from package lme4 (Bates et al., 2015). Tree models were developed using the function ctree (Hothorn et al., 2006) included in the package partykit (Hothorn, 2015). Cluster analyses were conducted using the functions kmeans included in the R package stats, with Euclidean distance, and the final number of clusters was determined with the function NbClust included in the package NbClust (Charrad et al., 2014).

Results

This study sought to understand the contribution of G, E and $G \times E$ on the determination of total variation on winter survival of canola. Results from the analyses of variance components showed that G, E and $G \times E$ explained 3%, 71%, and 7% of the variation in winter canola survival, respectively. The rest of the variation was pooled into model residuals (19%). The effect of $G \times E$ is twice as large as G indicating that $G \times E$ is an important determinant of survival. However, the sum of both terms (10%) is just a small proportion of the variation explained by E. Later

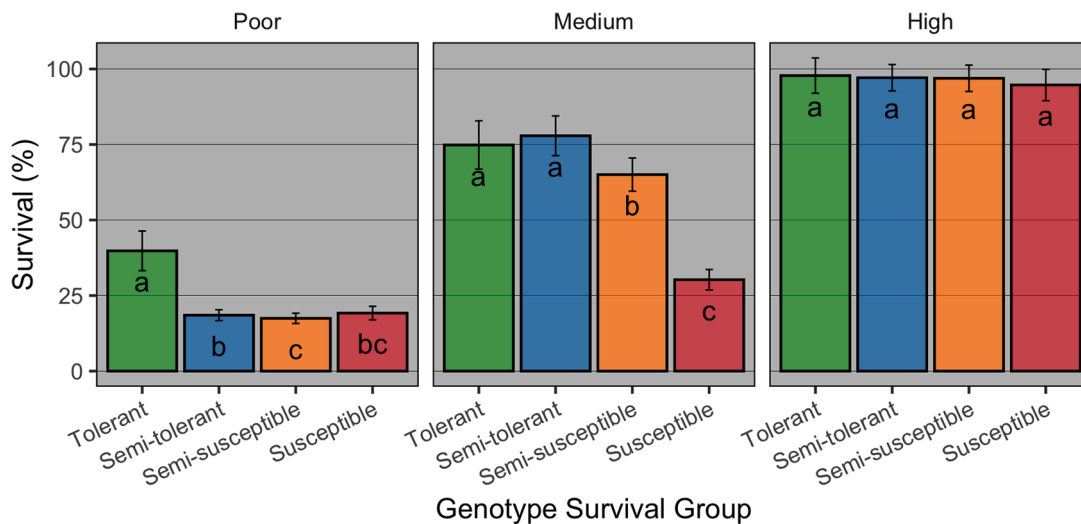


Fig. 5. Canola winter survival means across four different genotype survival groups (tolerant, semi-tolerant, semi-susceptible, and susceptible) for each meteorological survival cluster (poor, medium, high). Means within a panel followed by the same letter are not statistically different at alpha=0.05. Bars represent model-derived standard error.

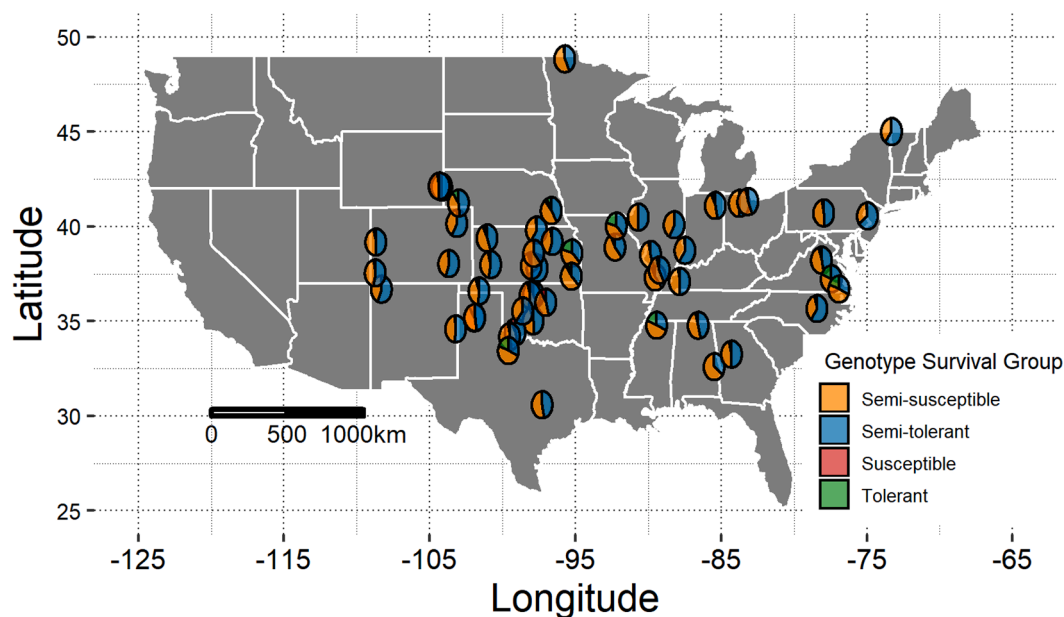


Fig. 6. US map with the sites included in the National Winter Canola Variety Trial, displaying the distribution of each genotype survival group.

sections of this paper will focus on modeling the environmental determinants of survival, and to a lesser degree on the model to explain GxE.

Overall, winter survival averaged 84%, but ranged from 0 to 100%, indicating a broad range of variation (Table 3). Site-year modeled survival was grouped into poor, medium, and high survival clusters. The minimum, mean, and maximum survival and number of site-years per survival cluster were 0, 28, 48, and 27 for the poor; 50, 70, 82, and 36 for the medium; and 83, 96, 100, and 127 for the high, respectively. Summary statistics for meteorological variables during the CP had large variability, with coefficients of variation ranging from 14% (meanWind) to 564% (Coldest_pct). The wide range in coefficients of variation for all meteorological variables was expected due to the geographical extent of the dataset, ranging from Texas to northern Minnesota and from eastern North Carolina to Washington.

The conditional inference tree classified site-year CPs into seven terminal nodes based on the evaluation of meteorological variables (Fig. 2). The most relevant variables classifying the CP of 190 site-years into meteorological survival clusters and its specific binary splits of the final tree model were in the order of importance from high to low: i) Colder (5 days), ii) ncycle (24 cycles), iii) cET (465 mm cumulative ET), iv) meanWindchill (-3.6°C), v) Colder_pct (3%), and vi) maxTmean (13.4°C); (see Table 2 for full description of the predictors). The leave-one-out cross-validation procedure resulted in a model fit with overall accuracy of 58%. Accuracy for the poor, medium, and high meteorological survival clusters were 48%, 40%, and 60%, respectively. Category-specific sensitivity for the same classes were 10%, 0%, and 71%, and for specificity were 85%, 79%, and 49%, respectively. Site-year CP was classified as poor, medium, and high survival clusters in terminal nodes 12 (n=31); 7 and 13 (n=19), and 4, 6, 8, and 10 (n=140), respectively.

The classification model was used to classify each CP for all 94 sites during the 1979-2019 period, into one of the meteorological survival clusters. Thereafter, the proportion of each meteorological survival cluster overall years (from 19 to 41 years depending on the site), was

calculated for each site (Fig. 3). Overall, sites with a greater proportion of poor, medium, and high meteorological survival were found at latitudes >39° N, between 35° N and 39° N, and <35° N, respectively.

Canola winter survival was determined by meteorological survival cluster alone and as part of the interaction with genotype ($p < 0.0001$). Survival means were extracted across meteorological survival cluster as this was the most relevant type of comparison. BLUEs for genotype plus genotype by S_r interaction led to the identification of four different genotype-specific behaviors: tolerant (7 genotypes), semi-tolerant (129 genotypes), semi-susceptible (56 genotypes), and susceptible (10 genotypes) (Fig. 4).

BLUEs for G plus GxE by meteorological cluster show that all four genotype survival groups had similar mean survival in the high meteorological survival cluster (from 95% to 98%, Fig. 5). In the medium meteorological survival cluster, survival was greatest in tolerant and semi-tolerant genotype survival groups (78% and 75%, respectively), and lowest in the susceptible group (30%). In the poor meteorological survival cluster, only the tolerant genotype survival group had a significant greater survival (40%), while the other genotype survival groups ranged from 17% to 19% survival.

A breakdown of the genotype survival groups provides insight into the current state of winter canola genotype development and testing in the US. The susceptible canola genotypes are a mix of commercial and experimental genotypes bred outside the US that were grown in the country briefly before more adapted materials could be accessed. The semi-susceptible genotype survival group contains many experimental and commercial genotypes developed outside the US, but also some of the first genotypes developed specifically for US environments. This group contains the first genotypes to be grown on a widespread basis and the very first hybrid to be introduced to the market. The semi-tolerant genotype survival group contains many experimental and commercial genotypes, but the majority are from past and current US breeding programs. This includes a large number of experimental and new commercial genotypes from the Kansas State University breeding program, which has the goal of improving winter hardiness since its

conception in the mid-1990s. Newer hybrids from Europe with enhanced levels of winter hardiness and those containing the semi-dwarfing trait, a trait of significant benefit to winter survival in harsher European environments, are also a large proportion of this group. Other group members include widely grown US open-pollinated genotypes and popular European commercial hybrids. The tolerant survival group contains a few foundational breeding lines from the Kansas State University breeding program and the most winter hardy, commercially available genotypes on the market today. More specific information on the genotypes such as name, type and decade of release is provided in [appendix A](#). The most evenly distributed groups over all the evaluated states were the semi-tolerant and semi-susceptible genotyped groups ([Fig. 6](#)).

Discussion

Understanding the impact of meteorological factors on survival of winter canola will help to define breeding and agronomic objectives to close yield gaps. At the same time, mapping meteorological risk for winter survival will not only facilitate the introduction of winter canola within current cropping systems but improve overall diversity and sustainability. Although managing a new crop can be a difficult task, winter canola offers an alternative for sites where winter crops are limited to one species. However, some site-specific factors can limit its production including: available agronomic and varietal information, producers willing to grow a new crop, delivery points within a reasonable transportation distance, obtaining a good stand at planting, heat stress at flowering, and challenges at harvest caused by shattering of pods ([Stamm and Watson, 2013](#)). Identifying winter canola genotypes that will overwinter and the optimum planting date for a given region are two critical steps that must be resolved before the crop can be introduced into new areas ([Holman et al., 2011](#)). The two larger winter survival groups indicate the broad adaptation of semi-susceptible and semi-tolerant genotypes among geographical regions in the US. This indicates the potential for the development of more tolerant genotypes for new areas. Even though this study was one of the first to provide a comprehensive analysis of winter canola survival, a few factors limited our quantitative evaluation: i) lack of quantification of plants before and after winter, which increased the subjectivity of the evaluation; and ii) quality of weather data. Even though gridded data seemed to be robust for temperature ([Mourtzinis et al., 2017](#)), other weather variables may have been less accurately estimated.

The outcomes presented here on winter canola survival provide foundational knowledge for canola breeding processes to select genotypes better adapted to cold environments. These results may give breeders more ways to quantify the “type” of winter kill they observe. [Waalén et al. \(2013\)](#) provided one of the first in-depth characterizations of meteorological factors affecting winter canola survival in Norway, emphasizing not only the effect of the stress but also the importance of timing. Temperature fluctuations in the US Great Plains during the winter seem to trigger phases of dormancy and re-growth, creating significant stress on the plant ([Rife, 2003](#)). Cold acclimation (exposure to low temperature for temperate plants to achieve maximum freezing tolerance), de-acclimation (fully cold-acclimated plants are exposed to warm temperatures), and re-acclimation (re-exposure to cold acclimating temperatures) are complex processes studied in-depth in Canada ([Trischuk et al., 2014](#)). The “perfect” sequence of events to reduce winter kill might be to enter into growth cessation with adequate cold acclimation processes, followed by a winter period without extreme events of freezing temperatures, and finishing with a slow and gradual growth elongation and de-acclimation period. This agrees with [Rapacz \(1998\)](#), who showed that oilseed rape almost doubled its frost resistance through growth cessation during cold acclimation. In addition, [Rapacz \(2002\)](#) demonstrated in central Europe (Poland), that further re-acclimation is limited if bolting has begun during de-acclimation.

Based on our findings, winter survival was negatively affected by more than 24 cycles of mean daily temperature shifting from negative to positive or vice-versa, followed by cold temperatures with wind chill temperatures below -3.6°C . According to [Levitt \(1972\)](#), elongation growth may interfere with cold acclimation as a result of competition for photoassimilates between growth and acclimation, thus, the plant may be more susceptible to frost due to greater water content in the seedling. [Rife and Zenali \(2003\)](#) reported that de-acclimated seedlings could be re-acclimated, with the accumulation of dehydrins in canola linked to the development of frost tolerance ([Schilling, 2004](#)). Likewise, carbohydrate concentration increased during cold acclimation in winter canola ([Trischuk et al., 2014](#)) correlating to the photosynthetic capacity of the plants ([Hurry et al., 1995](#)).

In summary, plant, meteorology, and management factors such as days without snow cover, root collar diameter, the height of the crown (rosette) at the beginning of the winter, ice encasement, topography, conditions at planting, stand establishment ([Trischuk et al., 2014](#); [Waalén et al., 2013](#)), plant density, crop residue on the soil, leaf development ([Lääniste et al., 2007](#)), dehydration during sunny and/or windy days while the soil is frozen ([Sovero, 1993](#)), prolonged exposure to subzero temperatures, diseases, and soil heaving ([Levitt, 1956](#)) may be involved and interact to influence this important plant trait. This evidence, mostly from northern regions such as Europe and Canada, along with our results, suggests that winter survival is a complex trait.

Future research should focus on improving winter survival measurements, integrating new technologies to improve rapid phenotyping with the goal of increasing standardization and precision, and reducing the subjectivity, labor, and time to collect data for this relevant trait of canola. Lastly, investing resources to understand the physiological processes underpinning this trait and its interaction with other factors such as meteorology, management, and genotype will be relevant for increasing productivity and stability of yield over time.

Conclusions

Our analysis of National Winter Canola Variety Trial data indicated that during the winter period, the most relevant meteorological variables affecting winter survival were related to minimum temperature and its fluctuations. There are three important outcomes of this study. First, we found that the number of days with temperatures between -10°C and -15°C , the number of cycles when the temperature fluctuates above or below 0°C , and wind chill temperature during the cold period were the main meteorological variables that explained mean winter survival across 190 site-years. Second, we documented the potential to have near 100% winter survival below 35°N latitude in places where vernalization requirements could be satisfied, as well as in Minnesota, Washington, western Colorado, and near the Pacific coast. Third, we found that most broadly-adapted genotypes were classified as semi-tolerant (129) and semi-susceptible (56) to winter kill in the US, indicating there is potential to develop more tolerant genotypes for new areas and widespread use of semi-tolerant genotypes exists. Caution should be taken in specific states between 35°N and 40°N latitude where continental conditions are highly diverse and winter survival can be problematic.

Declaration of Competing Interest

The authors declare that there is no conflict of interest.

Acknowledgments

This work was supported by John Deere, Corteva Agriscience, USDA-NIFA Supplemental and Alternative Crops award number 2019-38624-30289, and the USDA-NIFA multistate project KS00-0042-NC7. Contribution no. 21-127-J from the Kansas Agricultural Experiment Station.

Appendix A. Genotype groups information

Genotype group	Name	Decade of release	Type
<i>Tolerant</i>	Celsius	2000s	Open-pollinated
	Explus	2010s	Hybrid
	KS2098	2000s	Open-pollinated
	KS2169	2000s	Open-pollinated
	KS9012	2000s	Open-pollinated
	Torrington	2010s	Open-pollinated
	USI2002	2000s	Open-pollinated
<i>Semi-tolerant</i>	15.WC.05633	2010s	Open-pollinated
	15.WD.1	2010s	Open-pollinated
	45D03	2000s	Hybrid
	46W14	2000s	Hybrid
	46W99	2010s	Hybrid
	AAMU-33-07	2010s	Open-pollinated
	Alabaster	2010s	Hybrid
	ARC00004-2	2000s	Open-pollinated
	ARC00005-2	2000s	Open-pollinated
	ARC00024-2	2000s	Open-pollinated
	ARC2189-1	1990s	Open-pollinated
	ARC2189-2	1990s	Open-pollinated
	ARC91019-50-e2	2000s	Open-pollinated
	ARC97019	2000s	Open-pollinated
	ARC98007	2000s	Open-pollinated
	Argos	2010s	Hybrid
	Artoga	2010s	Hybrid
	Atora	2010s	Hybrid
	Banjo	2000s	Hybrid
	Casino	1990s	Open-pollinated
	CHH2311	2010s	Hybrid
	CWH042	2010s	Hybrid
	CWH081	2000s	Hybrid
	CWH095	2000s	Hybrid
	CWH111	2000s	Hybrid
	CWH116	2000s	Hybrid
	CWH633	2000s	Open-pollinated
	DK Exstorm	2010s	Hybrid
	DK Imiron CL	2010s	Hybrid
	DK Imistar CL	2010s	Hybrid
	DK Imiron CL	2010s	Hybrid
	DK Sensei	2010s	Hybrid
	DKW13-69	2000s	Open-pollinated
	DKW44-10	2000s	Open-pollinated
	DKW45-25	2010s	Open-pollinated
	DKW47-15	2000s	Open-pollinated
	DSV05103	2000s	Hybrid
	DSV05104	2000s	Hybrid
	DSV07100	2000s	Hybrid
	Dynastie	2010s	Hybrid
	Extra	2010s	Hybrid
	Falstaff	2000s	Open-pollinated
	Forza	2000s	Open-pollinated
	Garou	2010s	Hybrid
	Gospel	2000s	Hybrid
	Griffin	2010s	Open-pollinated
	Hamour	2010s	Hybrid
	Hornet	2000s	Hybrid
	HPX-6271	2010s	Open-pollinated
	HPX-6406	2010s	Open-pollinated
HPX-7228	2010s	Open-pollinated	
HyCLASS107W	2000s	Open-pollinated	
HyCLASS110W	2000s	Open-pollinated	
HyCLASS125W	2010s	Open-pollinated	
HyCLASS225W	2010s	Open-pollinated	
Kadore	2010s	Open-pollinated	
KS2004	2000s	Open-pollinated	
KS2064	2000s	Open-pollinated	
KS2185	2000s	Open-pollinated	
KS2427	2000s	Open-pollinated	
KS3018	2000s	Open-pollinated	
KS3067	2000s	Open-pollinated	
KS3068	2000s	Open-pollinated	
KS3074	2000s	Open-pollinated	

(continued on next page)

(continued)

Genotype group	Name	Decade of release	Type
	KS3077	2000s	Open-pollinated
	KS3132	2000s	Open-pollinated
	KS3254	2000s	Open-pollinated
	KS3350	2000s	Open-pollinated
	KS4085	2010s	Open-pollinated
	KS4428	2010s	Open-pollinated
	KS8285	2000s	Open-pollinated
	KS8367	2000s	Open-pollinated
	KS9124	2000s	Open-pollinated
	KS9183	2000s	Open-pollinated
	KSR07352S	2010s	Open-pollinated
	KSR07363	2010s	Open-pollinated
	KSR4653S	2010s	Open-pollinated
	KSUR1211	2010s	Open-pollinated
	KSUR21	2010s	Open-pollinated
	Kuga	2010s	Hybrid
	Maestro	2000s	Hybrid
	MH 09DJ058	2010s	Hybrid
	MH 07J14	2010s	Hybrid
	MH 10G11	2010s	Hybrid
	MH 10L23	2010s	Hybrid
	NK Petrol	2010s	Hybrid
	NK Technic	2010s	Hybrid
	NPZ0326	2000s	Hybrid
	NZ0404	2000s	Hybrid
	NPZ0591RR	2000s	Hybrid
	NPZ0791RR	2000s	Hybrid
	Ovation	2000S	Open-pollinated
	Phoenix CL	2010s	Hybrid
	Plurax CL	2010s	Hybrid
	Popular	2010s	Hybrid
	Quartz	2010s	Open-pollinated
	Raffiness	2010s	Hybrid
	Rally	2000s	Hybrid
	Riley	2010s	Open-pollinated
	Rumba	2010s	Open-pollinated
	Safran	2010s	Open-pollinated
	Satori	2000s	Hybrid
	Sitro	2000s	Hybrid
	SLM0402	2000s	Hybrid
	Star 915w	2010s	Open-pollinated
	SW 013022	2000s	Open-pollinated
	SW 013121	2000s	Open-pollinated
	SW 013173	2000s	Open-pollinated
	SW 013211	2000s	Open-pollinated
	SW 013253	2000s	Open-pollinated
	SY Saveo	2010s	Hybrid
	Talent	2000s	Hybrid
	TCI Exp 983	2010s	Open-pollinated
	TCL.06.M1	2000s	Open-pollinated
	TCL.06.M3	2000s	Open-pollinated
	TCL.06.M4	2000s	Open-pollinated
	Visby	2010s	Hybrid
	VSX-3	2010s	Open-pollinated
	WC.15.7.5	2010s	Open-pollinated
	WC.9.7.5.7	2010s	Open-pollinated
	X01W692C	2010s	Hybrid
	X02W534C	2010s	Hybrid
	X10W443C	2010s	Hybrid
	X10W665C	2010s	Hybrid
	X12W377C	2010s	Hybrid
	X12W447C	2010s	Hybrid
	X13W029C	2010s	Hybrid
<i>Semi-susceptible</i>	46W94	2000s	Hybrid
	AAMU-18-07	2000s	Open-pollinated
	Abilene	1990s	Open-pollinated
	ARC2180-1	2000s	Open-pollinated
	ARC90016-pr377	2000s	Open-pollinated
	ARC92004-1	2000s	Open-pollinated
	ARC92007-2	2000s	Open-pollinated
	ARC97018	2000s	Open-pollinated
	Baldur	2000s	Hybrid
	Baros	2000s	Open-pollinated
	Ceres	1990s	Open-pollinated
	Chrome	2010s	Open-pollinated

(continued on next page)

(continued)

Genotype group	Name	Decade of release	Type
	Claremore	2000s	Open-pollinated
	Dimension	2000s	Hybrid
	DK Sensei	2010s	Hybrid
	DK Severnyi	2010s	Hybrid
	DKW13-62	2000s	Open-pollinated
	DKW41-10	2000s	Open-pollinated
	DKW46-15	2000s	Open-pollinated
	Edimax CL	2010s	Hybrid
	Einstein	2010s	Hybrid
	Flash	2000s	Hybrid
	Hekip	2010s	Hybrid
	Hidyille	2010s	Hybrid
	Hybrigold	2010s	Hybrid
	Hybristar	2010s	Hybrid
	Hybrisurf	2010s	Hybrid
	HyCLASS115W	2000s	Open-pollinated
	HyCLASS154W	2000s	Open-pollinated
	Inspiration	2010s	Hybrid
	Jetton	1990s	Open-pollinated
	Kalif	2000s	Open-pollinated
	Kiowa	2000s	Open-pollinated
	Kronos	1990s	Hybrid
	KS3302	2000s	Open-pollinated
	KS7436	2000s	Open-pollinated
	Mercedes	2010s	Hybrid
	MH 12AY04	2010s	Hybrid
	MH 12AY27	2010s	Hybrid
	MH 12AY36	2010s	Hybrid
	MH 06E10	2010s	Hybrid
	MH 09E3	2010s	Hybrid
	Plainsman	1990s	Open-pollinated
	PT211	2010s	Hybrid
	Rasmus	2000s	Open-pollinated
	Sumner	2000s	Open-pollinated
	SY Marten	2010s	Hybrid
	Taurus	2000s	Hybrid
	Titan	2000s	Hybrid
	Trabant	2000s	Hybrid
	Viking	2000s	Open-pollinated
	Virginia	2000s	Open-pollinated
	VSX-2	2000s	Open-pollinated
	Wichita	1990s	Open-pollinated
	Wotan	2000s	Open-pollinated
	X01W522C	2000s	Hybrid
<i>Susceptible</i>	Albatros	2010s	Hybrid
	ARC98015	2000s	Open-pollinated
	DKW13-54	2000s	Open-pollinated
	DKW13-86	2000s	Open-pollinated
	DSV06201	2000s	Hybrid
	DSV06202	2000s	Hybrid
	HPX-567	2000s	Open-pollinated
	KS7436-055	2000s	Open-pollinated
	MH 09H19	2010s	Hybrid
	TCL06.M2	2000s	Open-pollinated

References

- Assefa, Y., Roozeboom, K., Stamm, M., 2014. Winter Canola Yield and Survival as a Function of Environment, Genetics, and Management. *Crop Sci* 54, 2303–2313. <https://doi.org/10.2135/cropsci2013.10.0678>.
- Bushong, J.A., Griffith, A.P., Peeper, T.F., Epplin, F.M., 2012. Continuous Winter Wheat Versus a Winter Canola-Winter Wheat Rotation. *Agron. J.* 104, 324–330. <https://doi.org/10.2134/agronj2011.0244>.
- Bates, Douglas, Maechler, Martin, Ben Bolker, S.W., 2015. Fitting Linear Mixed-Effects Models Using lme4. *Journal of Statistical Software* 67 (1), 1–48. <https://doi.org/10.18637/jss.v067.i01>.
- FAO, 2020. Food Outlook - Biannual Report on Global Food Markets: June 2020. Food Outlook, 1. Rome. 32. <https://doi.org/10.4060/ca9509e>.
- Fowler, D.B., Gusta, L.V., Tyler, N.J., 1981. Selection for Winterhardiness in Wheat. III. Screening Methods. *Crop Sci.* 21, 896–901. <https://doi.org/10.2135/cropsci1981.0011183X002100060023x>.
- George, N., Tungate, K., Beeck, C., Stamm, M., 2012. Exploring Genotype by Environment Interaction in Winter Canola in North Carolina. *J. Agric. Sci.* 4. <https://doi.org/10.5539/jas.v4n2p237>.
- Gusta, L.V., Fowler, D.B., Tyler, N.J., 1982. Factors influencing hardening and survival in winter wheat. In: Sakai, A., Li, P.H. (Eds.), *Plant cold hardiness Freez. Stress Mech. Crop Implic. Acad. Press, New York*, pp. 23–40.
- Holman, J., Maxwell, S., Stamm, M., Martin, K., 2011. Effects of Planting Date and Tillage on Winter Canola. *cm* 10. <https://doi.org/10.1094/CM-2011-0324-01-RS>.
- Hothorn, T., Hornik, K., Zeileis, A., 2006. “Unbiased Recursive Partitioning: A Conditional Inference Framework.”. *Journal of Computational and Graphical Statistics* 15 (3), 651–674. <https://doi.org/10.1198/106186006X133933>.
- Hothorn, T., Z.A., 2015. “partykit: A Modular Toolkit for Recursive Partytioning in R.”. *Journal of Machine Learning Research* 16, 3905–3909.
- Huntington, J.L., Hegewisch, K.C., Daudert, B., Morton, C.G., Abatzoglou, J.T., McEvoy, D.J., Erickson, T., 2017. Climate engine: Cloud computing and visualization of climate and remote sensing data for advanced natural resource monitoring and process understanding. *Bull. Am. Meteorol. Soc.* 98, 2397–2409. <https://doi.org/10.1175/BAMS-D-15-00324.1>.
- Jacques, P.J., Jacques, J.R., 2012. Monocropping Cultures into ruin: The loss of food varieties and cultural diversity. *Sustainability* 4, 2970–2997. <https://doi.org/10.3390/su4112970>.

- Lääniste, P., Jõudu, J., Eremeev, V., Mäeorg, E., 2007. Sowing date influence on winter oilseed rape overwintering in Estonia. *Acta Agric. Scand. Sect. B Soil Plant Sci.* 57, 342–348. <https://doi.org/10.1080/09064710601029554>.
- Levitt, J., 1972. Responses of plants to environmental stresses. Academic Press, New York.
- Levitt, J., 1956. The hardiness of plants. Acad. Press, New York, NY.
- Charrad, Malika, Ghazzali, Nadia, Veronique Boiteau, A.N., 2014. NbClust: An R Package for Determining the Relevant Number of Clusters in a Data Set. *Journal of Statistical Software* 61 (6), 1–36.
- Messina, C.D., Technow, F., Tang, T., Totir, R., Gho, C., Cooper, M., 2018. Leveraging biological insight and environmental variation to improve phenotypic prediction: Integrating crop growth models (CGM) with whole genome prediction (WGP). *Eur. J. Agron.* 100, 151–162. <https://doi.org/10.1016/j.eja.2018.01.007>.
- Mourtzinis, S., Rattalino Edreira, J.I., Conley, S.P., Grassini, P., 2017. From grid to field: Assessing quality of gridded weather data for agricultural applications. *Eur. J. Agron.* 82, 163–172. <https://doi.org/10.1016/j.eja.2016.10.013>.
- Osczevski, R., Bluestein, M., 2005. The new wind chill equivalent temperature chart. *Bull. Am. Meteorol. Soc.* 86, 1453–1458. <https://doi.org/10.1175/BAMS-86-10-1453>.
- R Core Team, 2019. R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria.
- Rapacz, M., 2002. Cold-deacclimation of oilseed rape (*Brassica napus* var. *oleifera*) in response to fluctuating temperatures and photoperiod. *Ann. Bot.* 89, 543–549. <https://doi.org/10.1093/aob/mcf090>.
- Rapacz, M., 1998. The after-effects of temperature and irradiance during early growth of winter oilseed rape (*Brassica napus* L. var. *oleifera*, cv. *Gorczański*) seedlings on the progress of their cold acclimation. *Acta Physiol. Plant* 20, 73–78. <https://doi.org/10.1007/s11738-998-0046-9>.
- Renard, D., Tilman, D., 2019. National food production stabilized by crop diversity. *Nature* 571, 257–260. <https://doi.org/10.1038/s41586-019-1316-y>.
- Rife, C.L., Auld, D.L., Sunderman, H.D., Heer, W.F., Baltensperger, D.D., Nelson, L.A., Johnson, D.L., Bordovsky, D., M., H.C., 2001. Registration of ‘Wichita’ rapeseed. *Crop Sci* 41, 263–264.
- Rife, C.L., Zenali, H., 2003. Cold tolerance in oilseed rape over varying acclimation durations. *Crop Sci* 43, 96–100.
- Salmon, S.C., 1918. A PRELIMINARY NOTE ON SOIL MOISTURE AND TEMPERATURE FACTORS IN THE WINTER-KILLING OF GRAIN CROPS. *Science* (80-) 47 (1207). <https://doi.org/10.1126/science.47.1207.173>.
- Schilling, B.S., 2004. PhD thesis. University of Saskatchewan, Saskatoon, SK Canada, p. 163.
- Sovero, M., 1993. Rapeseed, a new oilseed crop for the United States. J. Janick J.E. Simon (eds.), *New Crop*. Wiley, New York. p. 302-307.
- Stamm, M., Berrada, A., Buck, J., Cabot, P., Claassen, M., Cramer, G., Dooley, S.J., Godsey, C., Heer, W., Holman, J., Johnson, J., Kochenower, R., Krall, J., Ladd, D., Moore, J., O’Neill, M.K., Pearson, C., Phillips, D.V., Rife, C.L., Santra, D., Sidwell, R., Sij, J., Sta, D., W., W., 2012. Registration of ‘Riley’ winter canola. *J. Plant Reg* 6, 243–245.
- Stamm, M., Cramer, G., Dooley, S.J., Holman, J., Phillips, D., Rife, C., Santra, D.K., 2015. Registration of ‘Griffin’ winter canola. *J. Plant Reg* 9, 144–148.
- Stamm, M., Dooley, S., Aiken, R., Angadi, S., Asfeld, E., Baldwin, B., Begna, S., Bell, J., Berg, S., Blair, M., Braden, I., Brown, J., Cabot, P., Cebert, E., Damicone, J., Darby, H., Davis, J., Dinkla, G., Esser, A., Holman, J., Job, A., Johnson, J., Jones-Diamond, S., Hol, J., Z., S., 2019. National Winter Canola Variety Trial.SRP1150. *Kans. Ag. Exp. St. and Coop. Ext. Ser.*.
- Stamm, M.J., and S.L. Watson, 2013. Winter Canola Production in Kansas: Producer Experiences. Kansas State University Agricultural Experiment Station and Cooperative Extension Service. MF3093. <https://bookstore.ksre.ksu.edu/pubs/MF3093.pdf>.
- Trischuk, R.G., Schilling, B.S., Low, N.H., Gray, G.R., Gusta, L.V., 2014. Cold acclimation, de-acclimation and re-acclimation of spring canola, winter canola and winter wheat : The role of carbohydrates, cold-induced stress proteins and vernalization. *Environ. Exp. Bot.* 106, 156–163. <https://doi.org/10.1016/j.envexpbot.2014.02.013>.
- USDA-NASS, 2019. Acreage ISSN: 1949-1522.
- Hurry, V.M., Strand, Å., Tobiæson, M., Gardeström, P., Öquist, G., 1995. Cold hardening of spring and winter wheat and rape results in differential effects on growth, carbon metabolism and carbohydrate content. *Plant Physiol.* 109, 697–706.
- Waelen, W., Øvergaard, S.I., Åssveen, M., Eltun, R., Gusta, L.V., 2013. Winter survival of winter rapeseed and winter turnip rapeseed in field trials, as explained by PPLS regression. *Eur. J. Agron.* 51, 81–90. <https://doi.org/10.1016/j.eja.2013.06.004>.