BioEnergy Research (2019) 12:509–523
https://doi.org/10.1007/s12155-019-09998-1

Agronomic Performance of *Brassicaceae* Oilseeds in Multiple Environments Across the Western USA

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Published online: 2 July 2019

Abstract
Brassicaceae oilseed crops can provide rotation benefits to dryland wheat (*Triticum aestivum* L.) and supply feedstock for biofuel production. However, growers face decisions about what oilseed crop is best suited for an environment. The objective of this study was to determine how varying production environments affect the agronomic performance of modern cultivars of six *Brassicaceae* crop species and identify ideal genotypes for seven growing environments spanning four ecoregions. A field experiment was replicated in Colorado, Idaho, Iowa, Minnesota, Montana, North Dakota, and Oregon, USA, between 2013 and 2016 to measure seed and oil yields of seed for four cultivars of *Brassica napus*, two of *B. carinata*, two of *B. juncea*, two of *Sinapis alba*, one of *B. rapa*, and one of *Camelina sativa*. Also, $\delta^{13}C$ signature of seed was used as an indicator of water limitation. Generally, across all genotypes, seed and oil yields increased with increased growing season precipitation. Modern commercial cultivars of *B. napus* and *B. juncea* had the highest seed oil contents and generally produced the greatest oil yields across most environments, although they were not always the highest seed yielders. For instance, *B. carinata* over six site years in North Dakota and Minnesota yielded greater than *B. napus* producing as much as 2471 kg ha^{-1} in Minnesota. Camelina produced competitive seed yields in some of the drier environments and its $\delta^{13}C$ signature indicated that it had the greatest drought resistance. However, seed oil content of some of these high yielding genotypes may need improvement before they are viable as biofuel feedstock.

Keywords Yield · Seed oil concentration · Renewable jet fuel · Carbon isotope discrimination · Drought stress

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Abbreviations

C Carbon

Introduction

Oilseed crops in the family *Brassicaceae* (henceforth *Brassicas*) have been targeted for integration into dryland wheat production systems in the western United States (USA). The agronomic benefits of *Brassica* rotation crops are reported to be many and include reduced wheat diseases [1–3], improvement in soil quality [4] and structure [5], utilization of deep soil nitrate that might otherwise cause ground water contamination [6], and elimination of certain weed problems associated with continuous cereal grain production [7, 8]. Yield increases in wheat after being planted in a rotation following *Brassica* species such as canola (*Brassica napus*) and Indian mustard (*Brassica juncea*) compared with continuous wheat have been reported [9, 10]. Moreover, in some western US areas where a traditional fallow-winter cereal rotation is used, farmers are looking for drought resistant, low water use crop alternatives for the fallow period, and certain *Brassica* species could fill this void [11].

Oilseed crops are also seen as a potential feedstock for production of renewable jet fuel [12]. Certified renewable jet fuel from high-quality seed oils have been commercially demonstrated and certified for aviation use, but full-scale production has been limited partly by a lack of oilseed crops purposely grown for production of renewable jet fuel (personal communication, Steve Csonka, 15 Nov. 2018). One reason for this lack of availability is the relatively high price of oilseeds needed for farmers to shift from production of other crops to oilseed feedstocks. This high price of oilseed feedstocks makes it difficult to achieve price-point equivalency with petroleum jet fuel [13, 14]. In the near term, renewable jet fuel provides a significant short-term technology pathway for helping meet air transportation and military needs for alternative fuel. The commercial aviation industry has established a goal of carbon neutral growth by 2020 and cutting petroleum jet fuel use 50% by 2050 [15].

However, there are constraints that must be considered before growers make decisions about what oilseed crop to raise in a particular environment. No one oilseed crop or cultivar can optimally meet all needs for oilseed feedstock across the environmentally diverse Midwest and western US states. Crops and cultivars differ in needs for nutrients, light, water, and temperature. In addition, rainfall, cumulative heat units, and frost influence oilseed quality and yields [16]. Therefore, climate variability is a major factor affecting not only yield and oil concentration of seed, but also the consistency of oilseed feedstock supplies to crushers and biorefineries.

Among the issues to be addressed is the need to characterize the crop suitability of plant species best adapted to different environments and uses. Genotype by environment (G × E) studies on *Brassica* oilseed plants have been conducted in other parts of the world and have helped to target the best species for a certain region, as well as highlight plant traits for further breeding improvements [17, 18]. However, currently, little information on *Brassica* crops that are most productive in different agroecological conditions in the western USA is available at the regional and national levels. The agronomic performance of a crop species is the result of genotype, environment, and their interaction. Multi-environment trials are useful for assessing the G × E interaction prior to growing a crop since most yield traits are influenced by the environment. The objective of this study was to determine how varying production environments affect the agronomic performance and seed quality attributes of modern cultivars of six *Brassicaceae* species and identify ideal genotypes for seven growing environments. Under rainfed conditions, we hypothesized that adaptability of genotypes will vary among environments differing in growing season climate and soil types.

Materials and Methods

Brassicaceae Species and Experimental Locations

Environmental effects on plant growth, seed yield, and seed oil yield of 12 spring annual *Brassica* genotypes representing six species (*Brassica napus* L., *B. rapa* L., *B. juncea* (L.) Czern., *B. carinata* A. Braun, *Sinapis alba* L., and *Camelina sativa* (L.) Crantz) were tested in seven states of the USA (Table 1; Akron, CO; Moscow, ID; Ames, IA; Morris, MN; Sidney, MT; Mandan, ND; and Echo, OR) over 4 years (2013–2016). This multi-environment trial spanned four different ecoregions (Table 1; Fig. 1) and was designed to assess and compare a wide range of commercial oilseed cultivars developed either for food or industrial use (Table 2). Cultivars of *B. napus*, *B. rapa*, *B. juncea*, and *S. alba* were chosen based on their popularity for production in *Brassica* producing areas of the northern USA and Canada; *B. carinata* were provided by Agrisoma (Agrisoma Biosciences Inc., Gatineau, Quebec, Canada) and are current cultivars developed for production in North America; and *C. sativa* (CO46) was chosen for its early maturity and proven production in the Northern Great Plains. Characterization of the geography and climate of each experimental site (i.e., environment) is given in Table 1.

Data from certain site years were not available for analysis due to crop failure including Colorado in 2013 due to extreme drought and Montana in 2015 due to hail prior to crop maturity. Partial crop failure was experienced at the Oregon site in 2013 due to poor emergence and stand establishment. An eighth test site had been established at Temple, TX, but was eliminated due to multiple crop failures caused primarily by flooding and weed pressure.

Table 1 Geographical location, soil type, and USDA Ecoregion characterization, including 30-year (or long-term) mean annual precipitation and temperature

Location (USA)	Site ID	Coordinates/elevation	Soil type	USDA Ecoregion†	Mean annual precipitation (mm)	Mean annual temperature (°C)
Akron, CO	CO	40° 09′ N, 103° 09′ W; 1383 m	Weld Silt Loam	Prairie Gateway	418	9.8
Ames, IA	IA	42° 00′ N, 93° 47′ W; 331 m	Canisteo silty clay loam	Heartland	910	9.7
Moscow, ID	ID	46° 73′ N, 117° W; 785 m	Palouse silt loam	Fruitful Rim	635	8.8
Morris, MN	MN	45° 35′ N, 95° 54′ W; 345 m	Barnes loam	Heartland	665	5.9
Sidney, MT	MT	47° 46′ N, 104° 14′ W; 692 m	Williams loam	Northern Great Plains	364	7.5
Mandan, ND	ND	46° 46′ N, 100° 54′ W; 591 m	Temvik-Wilton silt loam	Northern Great Plains	456	4
Echo, OR	OR	45° 44′ N, 119° 3′ W; 319 m	Ritzville silt loam	Fruitful Rim	271	11.8

† Based on farm resources regions described by the US Department of Agriculture-Economic Research Service

Experimental Design and Cultural Practices

In each field test, cultivars were planted in the same randomized complete block experimental design with four replicates. Generally, plot size was 3.6 m × 7.6 m but varied with the seeding equipment available at a location. Best management practices for seed bed preparation, tillage, seeding rate, row spacing, fertility, and pest control were based on grower guides established for each of the *Brassica* crops. Rates for fertilizer nitrogen (N) were based on expected yield goals with allowance for residual soil test N. Primary cultural practices are summarized in Table 3.

Chemical weed control options for broadleaf weeds are limited for non-GMO *Brassica* crops. Control of grassy and broadleaf weeds during spring and summer included applications of glyphosate (*N*-(phosphonomethyl)glycine) and other herbicides as needed (Table 4). Herbicides were applied with self-propelled sprayers with spray booms delivering up to 100 L ha^{-1}. Insecticides were applied for control of cabbage aphid (*Brevicoryne brassicae*) in July (OR and ID), flea beetles (*Phyllotreta cruciferae* Goeze) in late April to early May (ID and MN), and false chinch bugs (*Nysius raphanus*) in July 2014 (CO). No pesticides were applied at the IA site.

Meteorological Data

Meteorological data including air temperature and precipitation were collected at automated standard weather stations located on-site or nearby each study site. At a location, all cultivars were sown in all plots within a 1-day period. In

Fig. 1 US map showing the location of environments used in the study

Table 2 Species, cultivar, use, and source of seed evaluated in multi-environment trial

Species	Cultivar	Use	Source
B. napus	DK3042RR	Food	Dekalb
B. napus	Invigor L130	Food	Bayer
B. napus	Gem	Industrial	Univ. of Idaho
B. napus	Empire	Food	Univ. of Idaho
B. carinata	ACC A110	Industrial	Agrisoma
B. carinata	080814EM	Industrial	Agrisoma
B. juncea	Oasis	Food	Viterra
B. juncea	Pacific Gold	Food	Univ. of Idaho
C. sativa	CO46	Industrial	Montana State Univ.
B. rapa	Eclipse	Industrial	Univ. of Alberta
S. alba	Idagold	Food	Univ. of Idaho
S. alba	Tilney	Food	Coleman's Mustard

general, early and late maturing cultivars were harvestable within 1 week of each other. Average air temperature and accumulated precipitation during the growing season (i.e., from sowing to final harvest) at each site during the study period are reported in Table 5, while the long-term average annual temperature and precipitation are given in Table 1. There was no instance in which a site received precipitation after cultivars had reached physiological maturity such that the accumulated precipitation could be used to compare productivity among sites.

Plant Measurements

Plots at all sites were harvested for seed using plot-scale combines. Harvested seed was cleaned by means of an air-screen cleaner. Cleaned seed was oven dried to constant weight for determination of yield. All seed and oil yield values are presented on a dry matter weight basis. Seed oil concentration was determined using the method of Oblath et al. [19] and Gesch et al. [20] on cleaned samples using pulsed nuclear magnetic resonance (pNMR, Bruker mq-CU 20-series, Billerica, MA). In brief, seed samples of 2.5–3.0 g were weighed into 18 mm × 150 mm test tubes (Pyrex, Corning, NY) and heated to 40 °C for at least 30 min before analysis with the pNMR. Oil content was

Table 3 Cultural practices used for *Brassicaceae* crops grown at experimental sites in Akron, Colorado (CO), Ames, Iowa (IA), Moscow, Idaho (ID), Morris, Minnesota (MN), Sidney, Montana (MT), Mandan, North Dakota (ND), and Echo, Oregon (OR)

Cultural practice	Nutrient	Crop‡	CO	IA	ID	MN	MT	ND	OR
Row spacing (cm)		All	20	25	18	31	20	19	19
Seed depth (cm)		All	2	2	2.5	2	1.6	1.3	2.5
Tillage†		All	NT	CONV	CONV	CONV	NT	NT	NT
Seeding density (seed ha^{-1} × 1,000,000)		Bn, Br, Sa	2.94	1.50	1.24	1.48	1.48	1.48	1.24
		Bc	2.94	1.50	0.86	1.48	0.86	1.48	1.24
		Bj mustard	2.36	1.50	1.24	1.48	1.48	1.48–1.83	1.83
		Bj canola	2.36	1.50	1.83	1.48	1.48	1.48–1.83	1.83
		Cs	6.08	3.50	3.95	3.95	1.48	3.95	3.95
Application rate (kg ha^{-1})	N	Bn, Br, Bc, Bj	44	106	112	135	116	119–121	61–86
	P_2O_5		0	78	36	45	29	29	0
	K_2O		0	136	0	45	27	0	0
	S		0	17	25	22	22	0–22	12–17
	N	Sa	44	106	112	114	116	121	61–86
	P_2O_5		0	78	36	38	29	29	0
	K_2O		0	136	0	38	27	0	0
	S		0	17	25	19	22	0–22	12–17
	N	Cs	44	106	112	90	116	121	61–86
	P_2O_5		0	78	36	34	29	29	0
	K_2O		0	136	0	34	27	0	0
	S		0	17	25	0	22	0–22	12–17
Nutrient placement method§		All	BRD	INC	INC	INC	INC	INC	INC

† NT no-tillage and CONV conventional tillage

‡ Bn, Br, Bc, Bj, Sa, and Cs *Brassica napus*, *B. rapa*, *B. carinata*, *B. juncea*, *Sinapis alba*, and *Camelina sativa*

§ BRD broadcast and INC incorporated

Table 4 Pest management in *Brassicaceae* oilseed crops grown at Akron, Colorado (CO); Moscow, Idaho (ID); Morris, Minnesota (MN); Sidney, Montana (MT); Mandan, North Dakota (ND); and Echo, Oregon (OR)

Year	Date	CO	ID	MN	MT	ND	OR
2013	Pre-plant	Glyphosate at 1.1 kg a.i. ha^{-1}	Trifluralin at 1.1 kg a.i. ha^{-1}	Trifluralin at 1.1 kg a.i. ha^{-1}	Glyphosate at 0.7 kg a.i. ha^{-1}		
	May		Thiamethoxam at 0.02 kg a.i. ha^{-1}			Clethodim at 0.14 kg a.i. ha^{-1}	
	June		Clopyralid at 0.13 kg a.i. ha^{-1}; quizalofop at 0.06 kg a.i. ha^{-1}				
	July		Lambda-cyhalothrin at 0.04 kg a.i. ha^{-1}				
2014	Pre-plant	Glyphosate at 1.1 kg a.i. ha^{-1}	Trifluralin at 1.1 kg a.i. ha^{-1}	Trifluralin at 1.1 kg a.i. ha^{-1}	Glyphosate at 0.7 kg a.i. ha^{-1}	Glyphosate at 1.54 kg a.i. ha^{-1} 2 weeks before planting, glyphosate at 0.96 a.i. ha^{-1} 2 days before planting, trifluralin at 1.1 kg a.i. ha^{-1} 1 day before planting	Glyphosate at 1.3 kg a.i. ha^{-1}
	May		Thiamethoxam at 0.02 kg a.i. ha^{-1}	Malathione at 0.37 L a.i. ha^{-1} to control insects		Clethodim at 0.14 kg a.i. ha^{-1}	
	July	Lambda-cyhalothrin at 0.02 kg a.i. ha^{-1}	Lambda-cyhalothrin at 0.04 kg a.i. ha^{-1}				Lambda-cyhalothrin at 0.02 kg a.i. ha^{-1}
2015	Pre-plant	Glyphosate at 1.1 kg a.i. ha^{-1}	Trifluralin at 1.1 kg a.i. ha^{-1}	Trifluralin at 1.1 kg a.i. ha^{-1}	Glyphosate at 0.7 kg a.i. ha^{-1}	Glyphosate at 1.54 kg a.i. ha^{-1} 2 weeks before planting, glyphosate at 0.96 kg a.i. ha^{-1} 2 days before planting, trifluralin at 1.1 kg a.i. ha^{-1} 1 day before planting	Glyphosate at 1.8 kg a.i. ha^{-1}
	Early May		Thiamethoxam at 0.02 kg a.i. ha^{-1}	Malathione at 0.37 L a.i. ha^{-1} to control insects		Clethodim at 0.14 kg a.i. ha^{-1}	
	Late May		Clopyralid at 0.13 kg a.i. ha^{-1} to all except B. juncea, imazamox at 0.4 kg a.i. ha^{-1} to B. juncea, glyphosate at 0.63 kg a.i. ha^{-1} to DK3042, glufosinate-ammonium at 1.0 kg a.i. ha^{-1} to Invigor L130				
	July		Lambda-cyhalothrin at 0.04 kg a.i. ha^{-1}				Lambda-cyhalothrin at 0.02 kg a.i. ha^{-1}
2016	Pre-plant	Glyphosate at 1.1 kg a.i. ha^{-1}		Trifluralin at 1.1 kg a.i. ha^{-1}	Glyphosate at 0.7 kg a.i. ha^{-1}	Glyphosate at 0.7 kg a.i. ha^{-1}	Glyphosate at 2.2 kg a.i. ha^{-1}
	May		Thiamethoxam at 0.02 kg a.i. ha^{-1}				
	July		Lambda-cyhalothrin at 0.04 kg a.i. ha^{-1}				Lambda-cyhalothrin at 0.02 kg a.i. ha^{-1}

Table 5 Growing season mean temperature (Mean Temp) and accumulated precipitation (Acc. Prec.) from date of sowing (SD) to harvest (HD) for experimental sites at Akron, Colorado (CO); Ames, Iowa (IA); Moscow, Idaho (ID); Morris, Minnesota (MN); Sidney, Montana (MT); Mandan, North Dakota (ND); and Echo, Oregon (OR)

Site ID	2013				2014				2015				2016			
	SD	HD[†]	Mean Temp	Acc. Prec.	SD	HD	Mean Temp	Acc. Prec.	SD	HD	Mean Temp	Acc. Prec.	SD	HD	Mean Temp	Acc. Prec.
	–Month/day–		(°C)	(mm)	–Month/day–		(°C)	(mm)	–Month/day–		(°C)	(mm)	–Month/day–		(°C)	(mm)
CO	4/5[‡]	7/18	15.3	131	5/5	8/5	19.4	249	3/31	7/20	15.4	277	–	–	–	–
IA	4/6	7/26	16.8	390	4/18	8/12	18.3	512	3/1	8/5	14.7	529	–	–	–	–
ID	5/2	9/4	17.0	108	5/1	9/2	17.0	112	4/27	8/4	17.5	145	–	–	–	–
MN	5/7	8/28	19.6	325	5/7	8/26	19.3	363	4/29	8/17	19.4	321	–	–	–	–
MT	5/4	8/5	17.1	293	4/25	8/1	15.8	158	4/15§	8/12	16.5	188	4/13	8/2	16.2	213
ND	5/15	9/6	19.3	343	5/22	9/15	18.0	293	5/5	8/21	18.3	301	–	–	–	–
OR	4/8¶	7/31	17.0	74.6	4/1	7/22	18.0	87	3/13	7/16	17.6	125	3/7	7/22	16.1	97

[†] Harvest date (HD) corresponds to the date for the latest harvested oilseed species which tended to be *B. carinata*

[‡] Complete crop failure occurred in 2013 at Akron, CO due to drought

[§] Complete crop failure occurred in 2015 at Sidney, MT due to hail

[¶] Partial to complete crop failure occurred at 2013 at Echo, OR due to poor emergence and stand establishment

determined on a dry weight basis, calculated using the moisture content of the seed. Seed oil yield was calculated by multiplying total dry seed yield (kg ha^{-1}) by percent seed oil content.

Seed samples were prepared for ^{13}C isotope analysis by grinding approximately 4 g of dried seed to a fine powder in an Udy mill. Concentrations of $\delta^{13}C$ were determined on subsamples of milled seed using continuous-flow isotope ratio mass spectrometry (20–20 Europa, Europa Scientific, Cheshire, England) using the method described by Gesch et al. [21]. Variation in discrimination against ^{13}C isotope during photosynthesis is useful for characterizing drought induced stomatal limitations and enzymatic processes [22]. Values for δ (‰) were calculated as follows:

$$\delta = \frac{R_{\text{sample}}}{R_{\text{standard}}} \times 1000 \qquad (1)$$

where R_{sample} is the $^{13}C/^{12}C$ ratio of the sample and R_{standard} is the $^{13}C/^{12}C$ ratio of a standard calibrated to the PDB standard, which is a limestone fossil of *Belemnitella americana* from the Cretaceous Pee Dee formation in South Carolina [23].

Statistical Analyses

Two-factor analysis of variance (ANOVA) was used to analyze the G × E (12 cultivars by seven environments) data for the dependent variables. The means of four plot replicates at each environment for each cultivar at each year were used as replicates since not all cultivars by environment data were collected in the same years. Data were analyzed using PROC GLIMMIX in SAS (SAS Inst., Cary, NC) using cultivar (i.e., genotype) and environment as fixed effects. A Gamma distribution for analyzing seed oil content and seed $\delta^{13}C$ and a Poisson distribution for analyzing seed yield and seed oil yield were used because the data were non-uniformly distributed. Differences in least squares means were used to make pairwise comparisons if a significant F-test was obtained from an ANOVA at $P \leq 0.05$ with a Bonferroni adjustment for cultivar and environment main effects. If a significant cultivar × environment interaction was obtained from ANOVA at $P \leq 0.05$, the SLICE option in SAS was used to examine cultivar differences at each environment using a Bonferroni adjustment (Tables 6, 7, 8, and 9). The means reported are after back transforming the data.

To better characterize genotype adaptability and performance over the range of environments used in the study, a stability analysis was performed on seed and seed oil yield data using the regression technique of Finlay and Wilkinson [24]. For each genotype, its mean at each environment was plotted against the environment population mean (i.e., mean of all genotypes at that environment) and a linear regression performed (Figs. 2 and 4). The regression lines were compared to the 1:1 mean population line (slope = 1), and the regression slope for each genotype indicates level of stability (b). According to Finlay and Wilkinson [24], a b approximating 1.0 indicates average stability, $b > 1.0$ indicates below average stability, and $b < 1.0$ indicates above average stability. Additionally, within each location, seed yield and oil yield of cultivars were ranked into three categories: upper third, middle third, and lower third. The percent that each cultivar ranked in the upper, middle, or lower third categories was then calculated over all locations (Figs. 3 and 5).

Table 6 Seed yield as effected by *Brassica* genotype at seven locations across the western USA. Means within columns followed by the same letter are not significantly different at the $P \leq 0.05$ level

Cultivar	Species	CO Seed yield (kg ha^{-1})	IA	ID	MN	MT	ND	OR	Cultivar mean
Invigor L130	*B. napus*	1549 a	1708 a	1316 b	2395 b	1849 a	1386 de	706 d	1474
DK3042RR	*B. napus*	1283 c	1158de	1684 a	2188 d	1625 b	1750 a	642 e	1389
Pacific Gold	*B. juncea*	1176 d	1110 f	1170 d	2146 d	1055 d	1350 e	898 a	1227
Empire	*B. napus*	1395 b	1201 d	1301 bc	1417 g	1418 c	866 g	699 d	1150
080814 EM	*B. carinata*	999 f	1174 d	926 g	2471 a	787 f	1635 b	636 e	1119
CO46	*C. sativa*	991 f	723 i	1332 b	1101 hi	1626 b	1686 b	720 cd	1110
Oasis	*B. juncea*	1006 f	1683 a	1265 c	1714 e	668 g	960 f	862 a	1106
Tilney	*S. alba*	786 h	1125 ef	1017 f	1497 f	1039 d	1524 c	752 bc	1070
AAC A110	*B. carinata*	1069 e	1287 c	885 g	2326 c	679 g	1809 a	433 g	1060
Idagold	*S. alba*	676 i	763 h	1043 f	1124 h	906 e	1437 d	858 a	946
Gem	*B. napus*	854 g	969 g	1060 f	1461 fg	1053 d	781 h	586 f	933
Eclipse	*B. rapa*	889 g	1527 b	1109 e	1064 i	489 h	722 i	775 b	889
	Site mean	1028	1164	1158	1664	1020	1265	702	

Results

Weather Conditions

The latest sowing and harvest dates (Table 5) were typically for the ND site in the Northern Great Plains and the MN site in the Heartland region while the CO and IA sites farthest south and the OR site farthest west (Table 1) were generally the earliest to be sown and harvested. The wettest environments during the growing season were IA, averaging 477 mm, followed next by MN, averaging 336 mm, over three seasons. The driest environment was OR, which averaged 103 mm across three growing seasons followed by ID (122 mm) and MT (221 mm; across 2013/2014/2016). The locations where *Brassica* genotypes were the earliest to be sown generally had the lowest mean temperature over the growing season. The MN and ND sites which tended to have the latest sowing and harvest dates also had the highest average growing season temperatures (Table 5).

Seed Yield

Seed yields greatly varied among experimental sites as well as among oilseed genotypes within a site (Table 6). The

Table 7 Seed oil content effected by *Brassica* genotype at seven locations across the western USA. Means within columns followed by the same letter are not significantly different at the $P \leq 0.05$ level

Cultivar	Species	CO (g kg^{-1})	IA	ID	MN	MT	ND	OR	Cultivar mean
DK3042RR	*B. napus*	434 a	452 a	418 a	486 a	454 a	450 a	360 a	435
Invigor L130	*B. napus*	427 ab	429 a	417 a	473 ab	447 a	425 ab	373 a	426
Gem	*B. napus*	422 ab	400 ab	424 a	460 abc	435 a	405 abc	393 a	419
Empire	*B. napus*	389 abc	393 ab	395 abc	426 abcd	419 ab	406 abc	366 a	398
Oasis	*B. juncea*	381 abc	399 ab	406 ab	453 abc	397 ab	372 bcd	351 ab	393
Eclipse	*B. rapa*	342 cd	395 ab	413 a	435 abcd	421 ab	367 bcd	372 a	391
Pacific Gold	*B. juncea*	369 abcd	365 bc	376 abc	410 cd	398 ab	381 bc	344 ab	377
AAC A110	*B. carinata*	362 bcd	356 bc	382 abc	421 bcd	326 cd	380 bc	305 bc	360
CO46	*C. sativa*	362 bcd	338 c	347 c	387 d	372 bc	359 cd	355 a	360
080814 EM	*B. carinata*	318 de	336 c	350 bc	382 d	320 d	328 d	297 c	332
Tilney	*S. alba*	270 ef	239 d	273 d	284 e	276 e	273 e	253 d	266
Idagold	*S. alba*	235 f	237 d	278 d	285 e	269 e	271 e	251 d	260
	Site mean	354	355	369	403	372	364	332	

Table 8 Seed oil yield as effected by *Brassica* genotype at seven locations across the western USA. Means within columns followed by the same letter are not significantly different at the $P \leq 0.05$ level

Cultivar	Species	CO (Oil yield kg ha^{-1})	IA	ID	MN	MT	ND	OR	Cultivar mean
Invigor L130	*B. napus*	684 a	733 a	543 b	1133 a	819 a	601 c	258 b	630
DK3042RR	*B. napus*	579 b	535 c	706 a	1065 b	721 b	796 a	231 cd	608
Pacific Gold	*B. juncea*	447 c	447 e	433 e	873 d	419 d	539 d	309 a	473
Empire	*B. napus*	583 b	481 d	510 bc	610 g	586 c	354 f	250 bc	462
Oasis	*B. juncea*	394 de	729 a	506 c	777 e	266 ef	390 e	305 a	447
CO46	*C. sativa*	370 ef	269 h	465 d	432 hi	598 c	626 c	251 bc	408
AAC A110	*B. carinata*	426 cd	481 d	338 f	980 c	250 fg	699 b	138 f	400
Gem	*B. napus*	339 fg	392 f	446 de	681 f	444 d	327 g	228 d	388
080814 EM	*B. carinata*	365 ef	415 f	316 fg	950 c	270 ef	554 d	194 e	388
Eclipse	*B. rapa*	324 g	631 b	458 de	460 h	203 h	278 h	288 a	354
Tilney	*S. alba*	229 h	303 g	277 h	426 i	283 e	418 e	199 e	295
Idagold	*S. alba*	162 i	209 i	293 gh	321 j	239 g	393 e	221 d	253
	Site mean	381	439	426	673	381	474	234	

commercial lines of *B. napus* (Invigor L130 and DK3042RR) were generally the highest yielding genotypes across all environments (Table 6; far right column and Fig. 2) and produced the greatest yield at five out of the seven trial sites. Across environments, both Invigor L130 and DK3042RR yielded higher than the population means (Fig. 2; compare with 1:1 line), but their *b* values, 1.48 and 1.52, respectively, indicate environmental sensitivity (i.e., below average stability). This is clearly shown by their high yields in a favorable environment like MN, but lower relative yield in a less favorable environment like OR (Fig. 2).

Pacific Gold (*B. juncea*), which was one of the higher yielding genotypes in OR (Table 6), tended to have greater than average yields across environments with a *b* value of 1.30 (Fig. 2). The other *B. juncea* cultivar (Oasis) had average stability ($b = 0.95$) and yields compared with the population means (Fig. 2). In OR, which was the driest environment during the study (Table 5), *B. juncea* (Pacific Gold and Oasis) and *S. alba* (Idagold) tended to yield the greatest, although yields over all genotypes were generally low at OR.

The *B. carinata* cultivars, 080814EM and AAC A110, showed the greatest environmental sensitivity (below average stability), with *b* values of 2.03 and 2.11, respectively (Fig. 2). In MN, 080814EM produced the greatest seed yield averaging 2471 kg ha^{-1} over 3 years and both cultivars were relatively high yielding in ND (Table 6). However, in less favorable

Table 9 Delta^{13}C discrimination of seed affected by *Brassica* genotype at seven locations across the western USA. Means within columns followed by the same letter are not significantly different at the $P \leq 0.05$ level

Cultivar	Species	CO (δ^{13}C ‰)	IA	ID	MN	MT	ND	OR	Cultivar means
CO46	*C. sativa*	− 27.2 b	− 29.1 abc	− 27.7 c	− 28.8	− 27.7 c	− 28.9 c	− 26.2 b	− 27.9
Tilney	*S. alba*	− 26.4 ab	− 28.8 abc	− 27.4 bc	− 29.2	− 26.6 bc	− 27.9 abc	− 24.9 ab	− 27.3
Invigor L130	*B. napus*	− 26.6 ab	− 29.4 bc	− 26.3 ab	− 29.5	− 26.3 abc	− 28.0 abc	− 24.6 a	− 27.2
Idagold	*S. alba*	− 26.4 ab	− 28.9 abc	− 27.1 abc	− 28.9	− 26.5 bc	− 27.9 abc	− 24.8 ab	− 27.2
Oasis	*B. juncea*	− 26.5 ab	− 28.7 abc	− 26.9 abc	− 29.2	− 25.2 ab	− 28.6 bc	− 24.8 ab	− 27.1
Eclipse	*B. rapa*	− 26.6 ab	− 29.6 c	− 27.0 abc	− 29.1	− 24.4 a	− 28.0 abc	− 25.2 ab	− 27.1
DK3042RR	*B. napus*	− 26.1 ab	− 29.4 bc	− 26.5 abc	− 29.3	− 26.2 abc	− 27.4 ab	− 24.4 a	− 27.0
AAC A110	*B. carinata*	− 26.4 ab	− 28.4 abc	− 26.5 ab	− 29.5	− 25.6 ab	− 27.5 ab	− 24.3 a	− 26.8
Pacific Gold	*B. juncea*	− 26.1 ab	− 28.1 a	− 26.6 abc	− 28.9	− 25.6 ab	− 27.2 ab	− 24.5 a	− 26.7
Empire	*B. napus*	− 25.9 ab	− 28.7 abc	− 26.4 ab	− 28.7	− 25.3 ab	− 27.3 ab	− 24.3 a	− 26.6
080814 EM	*B. carinata*	− 25.2 a	− 28.2 ab	− 26.0 a	− 29.4	− 25.7 ab	− 27.5 ab	− 24.3 a	− 26.5
Gem	*B. napus*	− 25.9 ab	− 28.4 abc	− 26.4 ab	− 28.9	− 24.8 ab	− 26.8 a	− 24.4 a	− 26.5
	Site means	− 26.3	− 28.8	− 26.7	− 29.1	− 25.8	− 27.7	− 24.7	

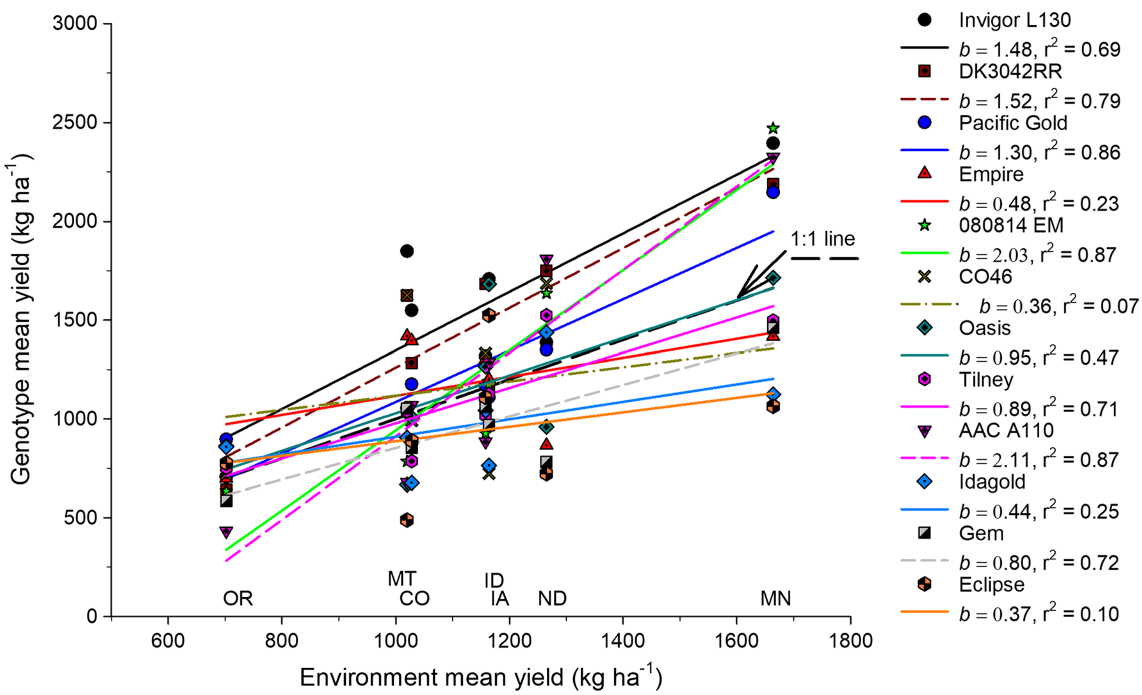
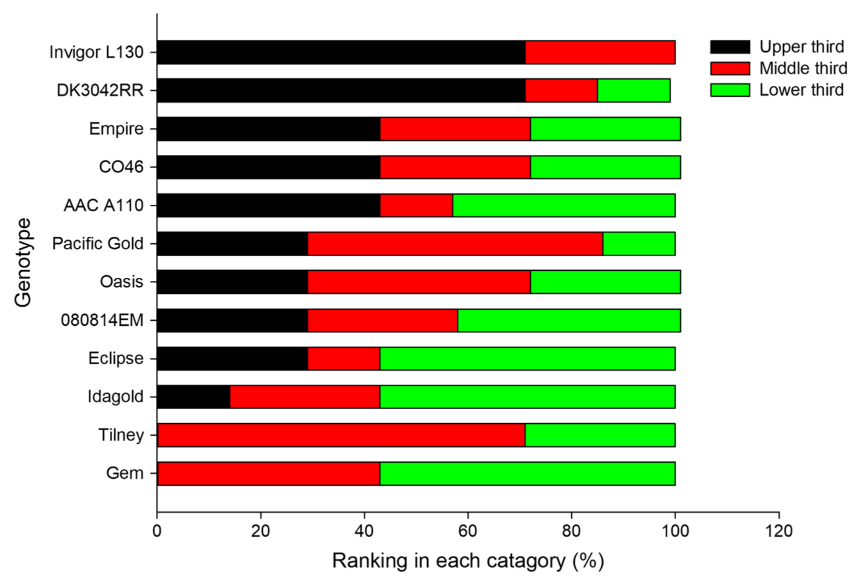


Fig. 2 Seed yield response for 12 *Brassicaceae* genotypes across the seven study environments using a Finlay-Wilkinson regression. The black dashed line represents the 1:1 population means (i.e., mean of all genotypes at each environment). State abbreviations mark the environment means which are also found in Table 6

environments like OR, MT, ID, and CO, the *B. carinata* cultivars were relatively low yielding (Table 6; Fig. 2). In ID, MT, and ND, *C. sativa* (CO46) was comparatively high yielding, while it was one of the lowest yielding genotypes at IA and MN (Table 6). The stability analysis indicated that CO46 was one of the most stable genotypes across environments ($b =$ 0.36) tending to yield high in less favorable environments but low in favorable ones (Fig. 2). Gem (*B. napus*) and Eclipse (*B. rapa*) also had low *b* values, 0.44 and 0.37, respectively, but consistently (stable) were low yielding across environments (Fig. 2; Table 6).

The cultivar-related data by environment lent itself well to grouping the cultivars into the upper, middle, and lower thirds based on values of the given factor measured (e.g., yield). By doing this, it was possible to estimate across site years the percent of time that a given cultivar was in each yield category and gain a better understanding of its performance across locations. For seed yield, Invigor L130 and DK3042RR, both *B. napus* cultivars, were in the upper third 71% of the time (Fig. 3) and Invigor L130 was never in the lower third. Empire (*B. napus*) and CO46 (*C. sativa*) were categorized in the upper third 43% of the time and both shared equal percentages

Fig. 3 Ranking of *Brassica* genotypes for seed yield in the upper, middle, and lower third across all site years of the study



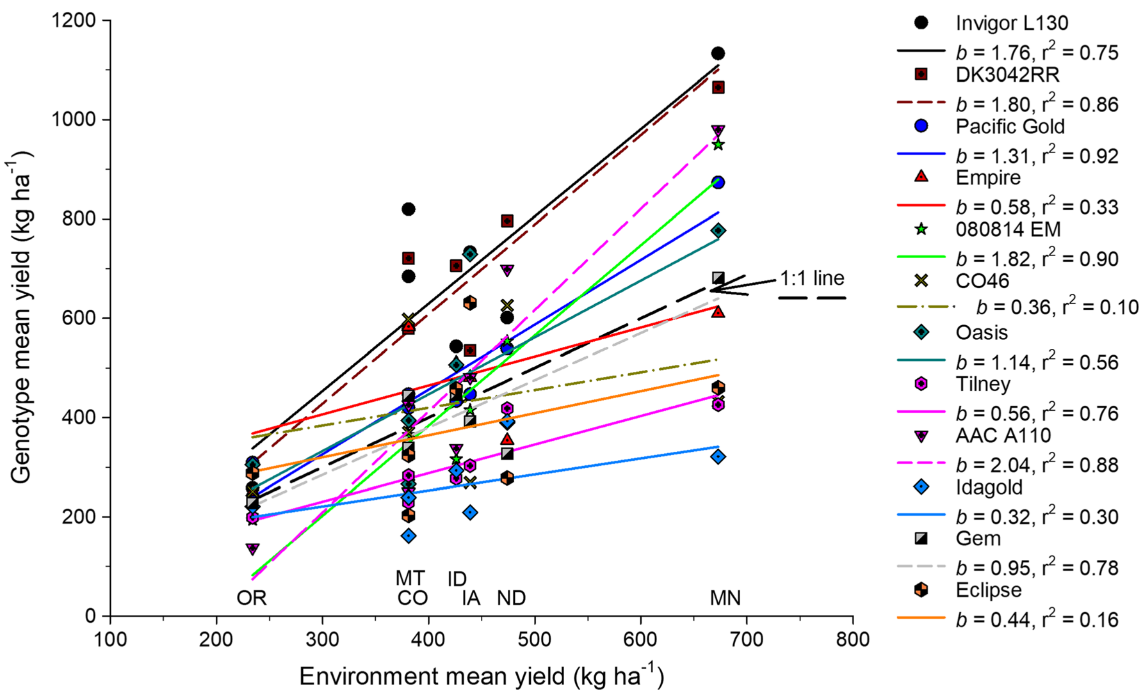


Fig. 4 Seed oil yield response for 12 *Brassicaceae* genotypes across the seven study environments using a Finlay-Wilkinson regression. The black dashed line represents the 1:1 population means (i.e., mean of all genotypes at each environment). State abbreviations mark the environment means which are also found in Table 8

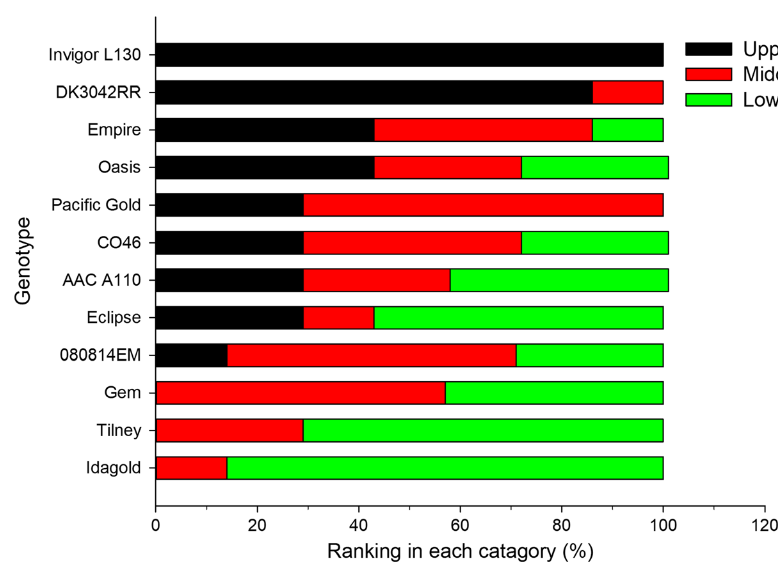
(29%) in the middle and lower third, while AAC A110 (*B. carinata*) also was in the upper third 43% but also occupied the lower third 43% of the time. Seed yields of Eclipse (*B. rapa*), Idagold (*S. alba*), and Gem (*B. napus*) occupied the lower third (57%) more than the other cultivars, and Gem and Tilney (*S. alba*) were never in the upper third.

Seed Oil Content and Yield

Seed oil content was clearly greatest for the *B. napus* cultivars followed by *B. juncea*, *C. sativa*, *B. carinata*, and *S. alba* (Table 7). When averaged across all cultivars, oil content varied across the different environments ranging from as high as 403 g kg^{-1} at MN to as low as 332 g kg^{-1} at OR. However, the pattern of cultivars with the highest, intermediate, and lowest oil content was similar across environments (Table 7). When averaged across site years, the range in oil content for *B. napus* was 398–435 g kg^{-1}, followed by *B. juncea* at 377–393 g kg^{-1}, *C. sativa* 360 g kg^{-1}, *B. carinata* 332–360 g kg^{-1}, and *S. alba* at 260–266 g kg^{-1}.

Seed yield is an important determinant of oil yield. Therefore, in most instances, across the different

Fig. 5 Ranking of *Brassica* genotypes for oil yield in the upper, middle, and lower third across all site years of the study



environments, the same genotypes that produced the greatest yields for a given site also tended to have the highest oil yield (Table 8; Fig. 4). Nevertheless, there were exceptions. For instance, at MN, although 080814EM (*B. carinata*) had the greatest seed yield (Table 6), its oil yield was significantly lower than *B. napus* lines Invigor L130 and DK3042RR by 183 and 115 kg ha^{-1}, respectively (Table 8), because the seed oil content of *B. carinata* was considerably lower than *B. napus* (Table 7). Likewise, 080814EM oil yield was lower, due to low oil content, than six other genotypes grown at the same site, although the seed yield of Idagold (*S. alba*) was one of the highest at OR.

The stability analysis of oil yield was similar to seed yield but with slightly better separation of the cultivars (Fig. 4). Invigor L130 and DK3042RR produced the greatest oil yield and were higher than the population mean (1:1 line) across environments but showed a high degree of environmental sensitivity with slopes (*b* values) of about 1.8. The *B. juncea* cultivars, Pacific Gold ($b = 1.31$) and Oasis ($b = 1.14$), showed near average adaptability across environments with yields above the population means, whereas Gem also had average stability ($b = 0.95$) but was low yielding. Tilney and Idagold (*S. alba*) and Eclipse (*B. rapa*) had above average stability (i.e., low environmental sensitivity) but were consistently low yielding. *Camelina sativa* also had relatively high stability ($b = 0.36$) and tended to yield higher than the population mean in less favorable environments but lower in more favorable environments (Fig. 4). Like seed yield, the *B. carinata* cultivars were the most unstable for oil yield, producing very high yields in favorable environments like MN but very low yields in less favorable environments like OR.

In terms of oil yield, Invigor L130 (*B. napus*) and DK3042RR were ranked in the upper third of all genotypes 100% and 86% of the time, respectively (Fig. 5). Empire (*B. napus*) and Oasis (*B. juncea*) at 43% were next highest to be found in the upper third but also both ranked in the lower third in some instances. Oasis and CO46 (*C. sativa*) were nearly evenly distributed among the upper, middle, and lower third. Cultivars Tilney and Idagold of *S. alba* occupied the lower third 71 and 86% of the time, respectively (Fig. 5).

Delta ^{13}C

At the MN site, there was no significant difference in δ^{13}C among any of the cultivars (Table 9). Although differences did occur at other sites, the range in δ^{13}C was generally narrow with little difference among most cultivars. The largest range was at the MT site where CO46 had the lowest δ^{13}C (− 27.7) and Eclipse had the highest (− 24.4). At the ND, ID, CO, MT, and OR sites, *C. sativa* (CO46) seed consistently had the lowest δ^{13}C signature while either Gem or 080814EM typically had the greatest. A comparatively low δ^{13}C signature generally indicates greater tolerance to water limiting conditions due to the influence of water stress on the photosynthetic mechanism [25]. In Table 9, the cultivars from top to bottom are in order of lowest to highest δ^{13}C. Averaged across all genotypes, the MN site recorded to lowest value (− 29.1) while OR had the highest (− 24.7).

Discussion

In the present study where 12 different modern oilseed cultivars representing six *Brassicaceae* species were evaluated, their performance greatly varied across the seven environments where they were grown. Ultimately, oil yield is the most important productivity factor impacting either biofuel or food use manufacturing, apart from production costs. Across experimental environments, the MN site resulted in the overall greatest oil yield averaged across all genotypes, producing about 200 kg ha^{-1} more oil than the next highest site which was ND (Table 8). The lowest oil producing environment was OR, which was also the driest site. Yield performance generally was closely associated with accumulated growing season precipitation, which was expected based on site characteristics (Table 1) and tended to decline along an east to west gradient. Other studies exploring G × E interactions of *Brassicaceae* species across environments in other parts of the world such as China [18] and Australia [17] have also highlighted seasonal precipitation as a major limiting factor of seed yield. Drought and heat stress are also known to decrease seed oil content [26, 27].

Using seed-derived δ^{13}C as an indirect indicator of water limitation severity, there was good association between average accumulated growing season precipitation and δ^{13}C averaged across all genotypes. The average seasonal accumulated precipitation for the IA, MN, ND, ID, CO, MT, and OR sites was 477, 336, 312, 122, 263, 158, and 103 mm, respectively (Table 5) in order of an increase in δ^{13}C with decreased precipitation (Table 9), indicating increased water limitation. Other factors that affect stomate functioning and CO_2 fixation such as light, salinity, and air pollutants (e.g., ozone) can also in-turn affect δ^{13}C discrimination [28]. Nitrogen availability, which impacts photosynthetic machinery, also influences δ^{13}C [25]. However, in the present study, N fertility was applied at rates such that it would not limit growth, and partitioning other factors mentioned that may have affected δ^{13}C was not in the scope of the study. Rather, based on vast differences in annual precipitation and temperature for the study environments (Table 1), it was reasoned that differences in δ^{13}C among genotypes across environments would be mostly due to water availability. An important factor to consider that may have contributed to water limitation or differences in plant water use is the interaction of evaporative demand with photosynthesis. Under semi-arid environments found at Akron, CO and Echo, OR, evaporative demand during the summer growing season is typically greater than that in humid mid-continental climates like found at Ames, IA and Morris, MN. Greater evaporative demand coupled with low precipitation in these semi-arid regions likely impacted photosynthesis leading to lower seed and oil yields and the higher degree of water limited stress as inferred from $\delta^{13}C$ measurements.

When averaged across all locations, the difference in $\delta^{13}C$ among most genotypes was small. However, especially at the driest environments (e.g., ID, CO, MT, and OR), CO46 (*C. sativa*) almost always had the lowest $\delta^{13}C$ signature, indicating a high degree of tolerance to water limiting conditions. Others have also demonstrated that camelina has a relatively high degree of drought tolerance compared to other *Brassicas* [29, 30]. This may in part be why camelina tended to be an above average yielder in drier, less favorable environments (Fig. 1). Alternatively, it could be related to its water use [31] and other resource use efficiency, as it did perform relatively poorly compared with other genotypes at high yielding environments. Also, Invigor L130, one of the top performing *napus* cultivars, tended to have a low $\delta^{13}C$ signature across all environments. Conversely, the genotypes Gem (*B. napus*), 080814EM (*B. carinata*), and Empire (*B. napus*) typically had the highest $\delta^{13}C$ values indicating lower tolerance to water limiting conditions, especially in the drier environments. Camelina (CO46) and the *S. alba* cultivars (Tilney and Idagold) which consistently had low $\delta^{13}C$ at most of the locations were also the earliest to mature while the *B. carinata* cultivars (080814EM and AAC A110) were typically the latest to mature and consistently had high $\delta^{13}C$ values. This relationship indicates potential drought avoidance by the earlier maturing *Brassica* genotypes. Similarly, Ngugi et al. [32] found that among 20 cowpea (*Vigna unguiculata* L. Walp.) cultivars tested across 17 dryland environments, ones with grain $\delta^{13}C$ signatures indicating low drought stress were usually the earliest to mature, which they attributed to drought avoidance. Also, for several canola cultivars evaluated in a G × E study by Zang et al. [33], it was shown that earlier flowering cultivars tended to yield better than medium flowering ones in low rainfall areas potentially through avoiding drought. Nevertheless, drought tolerance may still have played a significant role in the present study. For instance, camelina has been demonstrated to be a relatively low water user with good drought tolerance when grown in the arid southwestern USA [31].

In the present study, sowing dates were adjusted across environments in accordance with climate and soil conditions, which likely impacted seed and oil yield and $\delta^{13}C$ results. Dates varied from as early as March 7, 2016 at Echo, OR to as late as May 22, 2014 at Mandan, ND (Table 5). Nevertheless, agronomic practices were not necessarily optimized for a given environment and species, as optimum sowing date can vary with both factors. For instance, Guy et al. [34] reported that sowing date significantly not only impacted camelina yields across six experimental sites in the US Inland Pacific Northwest but also cited that growing season precipitation was perhaps the largest determinant of yield. Likewise, in the present study, growing season precipitation was a major determinant of seed and oil yields. Distribution of precipitation also influences optimum sowing date of spring crops, which in turn affects yield and seed attributes. For instance, the Pacific Northwest has mild winters and dry, hot summers with maximum temperatures reaching over 38 °C. Therefore, early spring sowing is favored to escape the onset of warm air temperatures exceeding 30 °C at which time flowering generally ceases [35]. In contrast, the continental climate of the Northern Great Plains has severely cold winters with temperature extremes as low as − 57 °C. Soils are slow to warm, and growers are often forced to seed in late spring [36].

As hypothesized, the species and cultivar that performed best at a given location varied with environment. However, based on the stability analysis, the popular commercial lines of *B. napus* (Invigor L130 and DK3042RR) generally produced the greatest seed and oil yields across all environments. This finding is not surprising given that more breeding and selection effort has been made for *B. napus* than other species used in this study. Moreover, these cultivars have been selected for production over a broad range of the USA, Canada, and Europe and all the *napus* lines used in the present study had higher seed oil contents than the other genotypes. The *napus* lines, Invigor L130 and DK3042RR, mimic what Tollenaar and Lee [37] referred to as "race horses" when studying maize (*Zea mays* L.) hybrids across a wide range of environments in the USA. Although they were generally high yielding across all environments, they also were environmentally sensitive (i.e., high *b* value), such that they were considerably more productive and better adapted to high yielding environments than low yielding ones (Figs. 2 and 4). Nevertheless, in the relatively high yielding environments of MN and ND, *B. carinata* produced seed yields greater than or equal to the two highest yielding *napus* lines, although in terms of oil yield *carinata* tended to be less. This indicates room for improvement of oil content in this species. Grown across a diversity of Canadian environments, Pan et al. [38] showed that newer selections of *B. carinata* performed as well or better than a commercial line of *B. juncea* indicating its high potential as an oil crop as breeding efforts increase. The *B. carinata* lines in the present study also had the greatest degree of environmental sensitivity (narrowly adapted) and, although high yielding in favorable environments like MN, did relatively poorly in less favorable environments like OR, perhaps due to breeding selection under high yielding or favorable environments [24, 37].

In the drier environments, *C. sativa* and to some extent *S. alba* were comparatively average to high seed yielders, but because of lower oil content, they generally had lower oil yields than most *napus* and *juncea* cultivars. For instance, camelina in ID, MT, and ND was the second highest seed yielding genotype, comparing well with the high yielding canola lines. Over the past decade, camelina has received attention in the USA and Europe as a low agricultural input alternative feedstock for biofuels, bioproducts, and healthy food use applications, and breeding efforts are underway to improve such traits as seed size, oil content, and yield [39]. In contrast, *S. alba* has potential as feedstock for biofuels [40] but has been primarily bred for condiment and culinary purposes without an emphasis on increasing oil content.

The US commercial aviation industry has a long-range goal of reducing its petroleum jet fuel use 50% by 2050 [15]. In 2017, the USA used 613,994,000 barrels (approximately 127 billion L) of jet fuel [41]. Although oilseed crops are envisioned as a significant near-term jet fuel feedstock, other sources (e.g., waste oil and increased fuel use efficiency) will also be necessary to achieve long-term goals. In this study, using the highest oil yielding genotypes by environment, average yield was 740 kg ha^{-1} or 804 L ha^{-1}, using an oil density of 0.92 kg ha^{-1} [42]. Therefore, the amount of arable land required for oilseeds to significantly impact reducing petroleum jet fuel use would be substantial. Land use competition for production of food or biofuel was beyond the scope of this paper. Nevertheless, the need to add new rotational crops to diversify and sustain agricultural systems in the USA is great. Farmers in the northern Great Plains are seeking crops to rotate with wheat and occupy fallow periods where certain oilseeds may fit [11]. In the inland Pacific Northwest, growers are interested in spring oilseeds to intensify the traditional winter wheat-fallow rotation in years of above average rainfall. Three-year rotations with a spring oilseed contain one less fallow year per 6-year period than winter wheat-fallow and can produce more bioenergy than fossil energy invested [43]. In the Midwest, the winter oilseeds pennycress (*Thlaspi arvense* L.) and winter camelina are being developed as dual-use crops to fit between corn-soybean rotations and, thus, could be produced on millions of hectares [44] in the Corn Belt region. One final note is that the seed meal produced from oilseed processing will contribute as animal feed and can provide a high-value protein source for human food uses [45].

Conclusions

This study demonstrated that although the particular genotype that performed the best at any given location differed, generally, when considered across all sites, modern commercial cultivars of *B. napus* and *B. juncea* produced the greatest oil yields. Seed and oil yields tended to decline with decreasing growing season precipitation from east to west across the Midwest and western USA. Based on seed δ^{13}C signature, results indicated that *C. sativa* exhibited the greatest tolerance to water limiting conditions of the genotypes studied and may be an ideal biofuel feedstock in drier environments of the USA. Seed yield of Ethiopian mustard (*B. carinata*) was equal or greater than high yielding *B. napus* cultivars in high-yield environments like MN but did not produce as great an oil yield due to lower seed oil content. For camelina that showed a relatively high degree of adaptability (i.e., stability), it is suggested that breeding should focus on greater oil content and resource use efficiency to take full advantage of high yielding environments, whereas selection for greater adaptability and higher oil content would benefit *B. carinata*, which demonstrated high-yield potential in certain environments. For *B. juncea*, which had average adaptability and slightly above average yields across environments and relatively high oil content, breeding for generally higher seed yields would be beneficial. Nevertheless, without government program support, the choice of *Brassica* oilseed species/cultivar for production in a given geographical region will likely depend on the profitability of feedstock production, which is not equal for the genotypes evaluated in this study. Feedstock cost has been identified as one of the biggest challenges for making renewable aviation and other biofuels economically competitive with petroleum-based fuels. Lastly, the choice of suitable oilseed feedstock will also depend on its fit with the prevalent cropping system(s) for a given region, which, especially in the case of newer oilseed alternatives such as camelina and Ethiopian mustard, requires further research.

Acknowledgments Numerous individuals provided technical assistance that was necessary to undertake a large multi-environment study. The authors thank John McCallum, Steve Umbarger, Aron Boettcher, Mandy Wuest, Hayley Peter-Contesse, Joe Boots, Chuck Hennen, Scott Larson, Chris Wente, Rick Greeson, Kim Hunter, Amber Williams, Michael Johnson, Mark Gaffri, Michelle Lair, Dale Spracklin, Rene France, Michelle Cryder, Craig Ford, Ashley Bateman, Delmer Schlenker, Cody Hardy, and David Poss for their valuable assistance in conducting these experiments.

Funding Information This work was supported by a Research Grant Award (2012-10008-19727) from the US Department of Agriculture, National Institute of Food and Agriculture.

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