Field Crops Research 164 (2014) 189–198

Contents lists available at ScienceDirect

Field Crops Research

journal homepage: www.elsevier.com/locate/fcr

# Improved wheat performance with seed treatments under dry sowing on permanent raised beds

M.J. Mulvaney, N. Verhulst, J.M. Herrera [1], M. Mezzalama, B. Govaerts *

International Maize and Wheat Improvement Center (CIMMYT), Apdo. Postal 6-641, 06600 Mexico, DF, Mexico

ARTICLE INFO

*Article history:*
Received 12 February 2014
Received in revised form 28 April 2014
Accepted 30 April 2014
Available online 21 May 2014

*Keywords:*
Conservation agriculture
Crop enhancement
Thiamethoxam
Pre-sowing irrigation

ABSTRACT

Two strategies for seeding irrigation are used for irrigated wheat. Wet sowing utilizes pre-sowing irrigation to germinate weed seeds and thus control weeds, followed by sowing. Dry sowing plants into dry soil that is irrigated soon afterward, resulting in higher soil moisture during germination and emergence than wet sowing. Field observations have indicated reduced emergence, plant stands and yield in dry compared to wet sowing on a Vertisol in northwestern Mexico. This disadvantage is more acute when dry sowing is conducted in permanent beds with residue retention (conservation agriculture) compared to the conventional system involving tillage with residue incorporation. To identify the causes of reduced plant stand and yield and examine control options, chemical seed treatment effects on durum wheat (*Triticum durum* Desf.) and bread wheat (*Triticum aestivum* L.) performance under wet and dry sowing were investigated over three seasons in a permanent bed system. Four seed treatments were applied: Control (no seed treatment); Carboxin + thiram + chlorothalonil (Vit-Dac; fungicides); Difenoconazole + mefenoxam (Dif-Mef; fungicides); and Thiamethoxam + difenoconazole + mefenoxam (TMX-Dif-Mef; insecticide and fungicides). Plant stands, root rot scores, normalized difference vegetative index (NDVI), and grain yield were determined. Under dry sowing, Dif-Mef and TMX-Dif-Mef increased plant stands by 87% and 104%, respectively, compared to Vit-Dac, and by 152% and 172%, respectively, compared to the control. Under dry sowing, TMX-Dif-Mef increased yield by 9.76% and 17.7% compared to Vit-Dac and the control, respectively. Bread and durum wheat were significantly different for both emergence and yield every growing season. Seed treatments effects were not significant under wet sowing. Treatment differences were not linked with root rot incidence later in the season. Several mechanistic hypotheses to explain the results were explored. TMX has been reported to alter genetic expression to enhance response to early season abiotic stresses, but this has not been reported for Dif-Mef. The different physical conditions during stand establishment, i.e. increased moisture and reduced temperature, under dry sowing compared to wet sowing, could have affected microbial populations which induced biological suppression of germination and/or emergence. Although more research is required to explain the underlying mechanism, wheat producers transitioning to a dry sowing system under conservation agriculture with permanent raised beds may avoid yield loss by utilization of a Dif-Mef or TMX-Dif-Mef seed treatment.



## 1. Introduction

Conservation agriculture is a system based on minimum tillage, crop residue retention, and crop rotation which aims to achieve increased profit and sustainably high production levels by reducing energy and labor costs, and improving soil structure, nutrient status, and water infiltration (Hobbs, 2007; Hobbs et al., 2008). Because of its potential to increase agricultural sustainability, conservation agriculture is promoted in many regions of the world (Pretty et al., 2011; Oladeebo and Mkhonta, 2013). The Yaqui Valley, in NW Mexico, is an area characterized by rainfall that is insufficient to sustain wheat (*Triticum* spp.) production (Hobbs et al., 2008), so wheat is

*Abbreviations:* Ctrl, Control; Dif-Mef, Difenoconazole + mefenoxam; NDVI, Normalized difference vegetative index; TMX, Thiamethoxam; TMX-Dif-Mef, Thiamethoxam + difenoconazole + mefenoxam; Vit-Dac, Carboxin + thiram + chlorothalonil.

* Corresponding author. Tel.: +52 55 5804 2004; fax: +52 55 5804 7558/59.
*E-mail address:* b.govaerts@cgiar.org (B. Govaerts).

[1] Present address: Instituto de Investigaciones en Biociencias Agrícolas y Ambientales (INBA-CONICET), Facultad de Agronomía, Universidad de Buenos Aires, Buenos Aires, Argentina.

http://dx.doi.org/10.1016/j.fcr.2014.04.017
0378-4290/

cultivated utilizing gravity irrigation. The area is representative of several major wheat-producing regions of the world, such as the Indus Valley in Pakistan, the Gangetic Valley in India, and the Nile Valley in Egypt (Braun and Payne, 2012), and served as an entry point for the semi-dwarf wheat introductions that characterized the Green Revolution in South Asia in the 1960s and 1970s (Borlaug, 2007). In contrast to most wheat regions, durum wheat (*Triticum durum* Desf.) is more extensively cultivated than bread wheat (*Triticum aestivum* L.), with the durum wheat cv. Cirno currently the most widely grown variety in the Yaqui Valley. Most farmers in the Yaqui Valley currently use conventionally tilled raised beds, forming new beds each season, and utilize furrow irrigation.

Two strategies for seeding irrigation can be used for irrigated wheat. Wet sowing utilizes a pre-sowing irrigation to germinate weed seeds and facilitate control of the first generation of weeds (Govaerts et al., 2009). After the emergence of weeds and before planting, they are controlled either mechanically (in conventional tillage systems) or with a broad-spectrum herbicide like glyphosate (in zero tillage systems). After pre-seeding irrigation, it is necessary to allow the soil to dry sufficiently before sowing. However, when precipitation occurs during this period, planting is delayed.

An alternative sowing system is dry sowing, in which the crop is sown directly into dry soil and irrigated soon afterward. Under wheat production, the term "dry sowing" is often used in the context of rain-fed conditions, e.g. Western Australia, referring to the practice of sowing in dry soil before the onset of the rainy season (McBeath et al., 2012), but in the arid region of NW Mexico, germination and emergence depend on irrigation. Therefore, despite the name, soil moisture is higher during germination and emergence under dry sowing than wet sowing. Although there is some literature on dry sowing in rainfed wheat systems (Smaling and Bouma, 1992; McBeath et al., 2012), there is almost no research available on dry sowing wheat under irrigated conditions. In Haryana, India, dry sowing wheat in soil infested with the *Heterodera avenae* nematode resulted in less nematode penetration, more tillers, and greater grain yield than wet sowing (Kanwar et al., 2013). Higher wheat tiller density and grain yields were also observed with dry sowing compared to wet sowing in Pakistan (Cheema et al., 1985).

Currently, wet sowing is the system most widely adopted by the farmers of the Yaqui Valley because of improved early season weed control. However, disadvantages of wet sowing include less efficient use of irrigation water and reduced flexibility of sowing date. The irrigation district has reduced water availability to farmers because of reduced rainfall in recent years combined with increased competition for water among industrial and domestic uses. As a consequence, the area has seen an increase in area planted with dry sowing. Therefore, CIMMYT decided to investigate and adapt dry sowing to conservation agriculture with permanent beds (i.e. no-tilled beds).

Field observations indicate a potential disadvantage of dry sowing compared to wet sowing due to delayed and uneven emergence, and reduced plant stands. This disadvantage becomes more acute when dry sowing is utilized in permanent beds with significant residue retention compared to the conventional system involving tillage and residue incorporation. Dry and wet sowing of wheat was compared on conventionally tilled and permanent beds during five growing seasons (unpublished data). On average plant stand density was reduced from 148 plants $m^{-2}$ with wet sowing to 51 plants $m^{-2}$ with dry sowing on permanent beds, whereas the effect of sowing system was smaller on conventionally tilled beds (170 plants $m^{-2}$ with wet sowing vs. 111 plants $m^{-2}$ with dry sowing). These treatments resulted in similar average yields with dry and wet sowing under conventionally tilled beds (7.7 t $ha^{-1}$ and 7.9 t $ha^{-1}$, respectively) but lower yields under permanent beds with dry sowing compared to wet sowing (7.1 t $ha^{-1}$ and 8.0 t $ha^{-1}$, respectively). The causes of the differences in plant stand density remain unknown.

It was hypothesized that reduced plant stands and slower early season growth under dry sowing compared to wet sowing may be overcome with the use of chemical seed treatments, some of which are purported to enhance crop growth, such as thiamethoxam (TMX). Although the mechanisms are poorly understood, TMX seed treatment has been shown to improve the germination of wheat (Larsen and Falk, 2013), soybean (*Glycine max* (L.) Merr.) (Dan et al., 2011) and palisade grass (*Urochloa brizantha* (Hochst. ex A. Rich.) R. Webster) (Macedo et al., 2013), but the results are not consistent in all cases or crops (Horii et al., 2007). Macedo and Camargo e Castro (2011) found that wheat seed treated with TMX had increased root development and tillering, and reduced nitrate reductase activity, while Perelló and Bello (2011) reported that wheat root biomass increased up to 600% with TMX-treated seed compared to the control under field conditions.

The objective of this study was to investigate the effects of seed treatments on the establishment, growth, and grain yield of bread and durum wheat under wet and dry sowing using conservation agriculture.

## 2. Materials and methods

### 2.1. Experimental conditions

The experiment was conducted at CIMMYT's experiment station near Ciudad Obregón in Cajeme, Estado de Sonora, Mexico (lat. 27.33° N, long. 109.09° W, 38 masl). The station is located in an arid climate with a mean annual temperature of 24.7 °C and average rainfall of 384 mm (1971–2000), of which 23% falls during the November-May wheat growing season (Verhulst et al., 2012). The soil is a Hyposodic Vertisol (Calcaric, Chromic) in the World Refeerence Base Classification System (IUSS Working Group WRB, 2007) or a fine, smectitic Chromic Haplotorrert in the USDA Soil Taxonomy Classification System (USDA Soil Survey Staff, 2003). It is characterized by low soil organic matter (SOM < 12 g $kg^{-1}$ soil) and slight alkalinity (pH 8) (Verhulst et al., 2009). The top 1.2 m soil had a clay texture (on average 300 g $kg^{-1}$ sand, 200 g $kg^{-1}$ silt, and 500 g $kg^{-1}$ clay). Bulk density ranged from 1.3 Mg $m^{-3}$ in the plough layer to 1.5 Mg $m^{-3}$ in the Vertic horizon.

The experiment was initiated during the 2009–10 winter growing season. The experiment was a maize (*Zea mays* L.)-wheat rotation, with maize cultivated in the summer and wheat in the winter. The trial comprised two wheat genotypes: the durum wheat cv. Cirno and the bread wheat cv. Roelfs. Each genotype was sown at two seeding rates (80 and 120 kg $ha^{-1}$) into two contrasting sowing systems (dry and wet sowing). The sowing systems differed in sowing irrigation management. Wet sowing plots were irrigated approximately three weeks before sowing (19, 21, and 26 days before sowing during 2009–10, 2010–11, and 2011–12, respectively) and not at sowing. Dry sowing plots were not irrigated before sowing and were irrigated within one day after sowing.

The seed treatments used were:

(i) A control without product application, a common practice among producers who save seed for sowing the following season,
(ii) Carboxin (fungicide, 50 g a.i. 100 $kg^{-1}$ seed) + thiram (fungicide, 50 g a.i. 100 $kg^{-1}$ seed) + chlorothalonil (fungicide, 150 g a.i. 100 $kg^{-1}$ seed) (Vit-Dac), a common commercial practice in the area,
(iii) Difenoconazole (fungicide, 18.6 g a.i. 100 $kg^{-1}$ seed) + mefenoxam (fungicide, 1.5 g a.i. 100 $kg^{-1}$ seed) (Dif-Mef), and

(iv) Difenoconazole (fungicide, 18.6 g a.i. 100 $kg^{-1}$ seed) + mefenoxam (fungicide, 1.5 g a.i. 100 $kg^{-1}$ seed) + thiamethoxam (insecticide, 10.075 g a.i. 100 $kg^{-1}$ seed) (TMX-Dif-Mef).

The liquid-formulated products were spread evenly inside a plastic bag, the seed was then added and shaken for approximately two minutes. The uniformly treated seed was spread as a thin layer on clean paper for drying (overnight or until the time of planting, usually within 24–48 h after treatment).

The reduced seed rate of 80 kg $ha^{-1}$ (vs. 120 kg $ha^{-1}$ commonly used) was included due to the capacity of wheat to tiller and compensate for stand reductions. Since high seeding rates may compensate for reduced stands, the reduced rate was included so that seed treatment differences would not be hidden by high planting rates.

The genotype and sowing rate treatments were arranged in randomized complete blocks with split-split-plots (on sowing system and seed treatment) and three replications. Plot size was 15 $m^2$ (1.5 m × 10 m) and plots consisted of two 0.75 m wide beds, for a total of four row plots (twin-row wheat on two beds).

The experimental area had been under permanent beds for more than 10 years. Each bed was reshaped without tillage prior to sowing. That is, the beds tended to flatten out and required reshaping of the furrows each season, but this was done without disturbing the soil on top of the beds themselves. This system is referred to as "permanent raised beds." Two wheat rows were sown on top of the beds leaving approximately 0.24 m between the rows. Seed depth under dry sowing was shallower than under wet sowing. Under dry sowing, seed depth was approximately 3 cm, while under wet sowing seed depth was approximately 7 cm. The shallow seed depth for dry sowing, which is common in the area, was meant to allow for rapid emergence as well as to prevent anoxia around the seed. Before sowing, 103 kg N $ha^{-1}$ and 52 kg $P_2O_5$ $ha^{-1}$ was band applied each wheat season in the center of the beds. At the end of the tillering stage, 175 kg N $ha^{-1}$ as urea was banded in the furrow. Wheat was sown on the 2nd of December 2009 in the 2009–10 season, the 7th of December 2010 in the 2010–11 season, and the 12th of January 2012 in the 2011–12 season. The late planting in the 2011–12 season was due to unseasonal rainfalls in November and December 2011. Four auxiliary irrigations were applied per season, for a total of approximately 520 mm of irrigation water applied per season. Pests were controlled as necessary throughout the experiment. The only fungicide application during the course of the study was of Folicur (active ingredient tebuconazol) on the 3rd of March 2012. No other foliar fungicides were applied throughout the experiment because the varieties used were resistant to rust (*Puccinia* spp.) (Figueroa-López et al., 2010, 2011).

### 2.2. Data collection

Meteorological data were obtained from a weather station located approximately 2 km away from the experiment.

Plant stand density was determined in each plot by averaging plant counts in three areas of 0.50 × 0.75 m (0.375 $m^2$) 20 days after sowing. That is, they were counted in three representative areas across a twin-row bed in each plot.

To screen for the incidence of root rot, a representative sample was obtained by randomly selecting 15 plants approximately at first node (2009–10) and anthesis (2010–11 and 2011–12). Plants were dug from approximately 0.10 m of row at four places in each plot, shaken gently to remove the loose soil, and then bulked in a plastic bag for transport to the laboratory and storage at 4 °C before processing. Plant roots were placed in water for 12 h and then washed under tap water to free the roots of soil. The roots were visually inspected for root rot lesions and decay using a magnifying glass, and rated on a 0–7 scale, modified from Schillinger et al. (1999), who used a 0–8 scale. The assessments included scoring for root lesions on seminal and crown roots of five plants. Total root rot score was determined by averaging the scores from seminal and crown roots.

Normalized difference vegetative index (NDVI) measurements were collected during the growing season using an optical handheld NDVI sensor (GreenSeeker™, NTech Industries, Inc, USA). Measurements were recorded by passing the sensor approximately 0.8 m over the crop canopy. The sensor covered a strip approximately 0.6 m wide, which included the two wheat rows of the measured bed. Measurements were recorded twice a week except when recent irrigation did not permit field entry.

Each plot was harvested at the end of the growing season and grain yield determined (Pask et al., 2012). Grain yield was adjusted so that all treatments are reported at 12% moisture content.

### 2.3. Data analysis

Significant effects were identified by analyses of variance (ANOVA) as implemented in SAS 9.3 using PROC MIXED procedures (SAS, 2013). Analyses were conducted within the growing season. Data were blocked by replications and split by sowing system and seed treatment factors. Sowing system × sowing rate × genotype × seed treatment factors and their interactions were held as fixed effects. Replication × sowing system and replication × sowing rate × genotype × seed treatment interactions were held as random effects, since the experimental design was blocked by replication and split-split plot on sowing system and seed treatment. Variables were considered significant if $p < 0.05$ unless otherwise stated. Means and standard errors of significant effects of the reduced models were obtained using PROC MEANS. Multiple pairwise means separation tests were conducted using least significant differences at the 95% confidence level with the %PDMIX800 macro (Saxton, 1998) within SAS 9.3. When multiple pairwise comparisons are mentioned, the p-value reported reflects the least (or greatest) significant difference and is denoted by "$p\leq$" or "$p\geq$", respectively.

NDVI curves were divided into four phases for purposes of analysis. These phases were establishment (approx. 0–22 days after sowing), early growth with increasing NDVI values (approx. 22–55 days after sowing), plateau NDVI values (approx. 55–119 days after sowing), and senescence with decreasing NDVI values (approx. >119 days after sowing). Exact phase delineations varied slightly between seasons and were determined empirically. Analyses of NDVI data were performed using R v.3.0.0 Statistical Programming Language (R Core Team, 2013) for the output of means, standard errors, and pairwise comparisons of slopes at the 95% confidence level (unless otherwise indicated) as implemented in the Least-Squares Means (LSMEANS) (Lenth, 2013), Linear and Nonlinear Mixed Effects Models (NLME) (Pinheiro et al., 2013), and Spatial and Space-Time Point Pattern Analysis Functions (SPLANCS) (Bivand et al., 2013) packages. Analyses of the plateau phase NDVI data were conducted using SAS 9.3 using PROC MIXED with the REPEATED option and first-order autoregressive covariance structure, since data were expected to be autocorrelated within an experimental unit. Multiple pairwise separation tests of plateau phase NDVI data were conducted as described above using the %PDMIX800 macro (Saxton, 1998).

## 3. Results

### 3.1. Growing conditions

The average annual rainfall between 2009–12 was 275 mm, 109 mm less than the long term (1971–2000) average. Total precipitation during the November–May wheat growing season was 40.1 mm in 2009–10, 1.6 mm in 2010–11, and 83.3 mm in 2011–12 (most of which occurred before sowing). Despite being characterized by high inter-annual variability, annual rainfall was always under the evaporative demand which, according to the annual refeerence ET0 (Penman–Monteith), averaged 1830 mm per year from 2009–2012. Rainfall was summer dominated; 1–32% of total annual rainfall occurred during the wheat growing season (November–May). The mean annual temperature was 20.0 °C (4.7 °C cooler than the long term average) while the mean monthly temperature ranged from 8.6 °C in January to 29.7 °C in July and August. The mean temperatures for the November–May growing seasons were 14.2, 13.7, and 13.9 °C for the 2009–10, 2010–11, and 2011–12 seasons, respectively. The 2010–11 growing season was characterized by a severe frost event approximately 60 days after sowing (Fig. 1).

Since germination and emergence may be affected by temperatures, the climatic conditions during sowing were examined. During the 2010–11 season, sowing occurred under warmer conditions than during the 2009–10 and 2011–12 seasons (Fig. 2). Fifty percent emergence for wet sowing occurred nine days after planting for wet-sown plots during 2009–10; for dry sowing 50% emergence ranged 9–12 days. During 2010–11, 50% emergence occurred nine days after planting on wet-sown plots and 9–11 days after planting on dry-sown plots. During 2011–12, emergence depended on the species: wet-planted durum (cv. Cirno) emerged five days after sowing, bread wheat (cv. Roelfs) emerged after 11 days. Dry-planted durum wheat emerged eight days after planting, while bread wheat emerged after 16 days. Only during 2009–10 did TMX-Dif-Mef emerge significantly faster than the control and Vit-Dac treatments under dry sowing (data not shown), and the effect was significant for both species. In these treatments, the time to emergence with TMX-Dif-Mef under dry sowing was equal to that of wet sowing. In all cases, emergence was delayed and more varied under dry sowing compared to wet sowing, although not always significantly so.

### 3.2. Plant stand density

Under dry sowing, plant stand density was consistently and significantly higher with Dif-Mef and TMX-Dif-Mef seed treatments compared to the control and Vit-Dac treatments (Fig. 3). Under dry sowing, Dif-Mef and TMX-Dif-Mef increased plant stands by 87% and 104%, respectively, compared to Vit-Dac, averaged over all seasons, genotypes, and sowing rates. Compared to the control, Dif-Mef and TMX-Dif-Mef increased plant stands by 152% and 175%, respectively, under dry sowing. Plant stand densities were similar for all treatments under wet sowing, however. Only during the 2010–11 season there was a significant interaction between seed treatment × genotype × sowing system ($p \leq 0.025$). Seed treatment effects did not differ by genotype during any season (seed treatment × genotype interactions, $p \leq 0.28$). However, there were significant differences between seed treatment effects among sowing systems during all seasons ($p \leq 0.0001$). Under wet sowing, seed treatment with TMX-Dif-Mef significantly increased plant stands only during the 2010–11 season compared to the control.

### 3.3. Root rot incidence

There was no clear effect of the seed treatment or sowing system on root rot incidence. The total root rot scores (mean ± SE) were 2.72 ± 0.04 (2009–10), 2.03 ± 0.06 (2010–11), and 1.58 ± 0.057 (2011–12), scored on a 1–7 scale. These scores were lower than the threshold considered to be yield-limiting (Schillinger et al., 1999). There was generally slightly lower incidence of root rot under dry sowing than wet sowing, but these differences were generally not significant (data not shown). Similar data were obtained for root rot incidence on seminal and crown roots.

### 3.4. Canopy closure and growth

During the 2009–10 season, the effects of seed treatments on NDVI were significantly different on dry sowing systems only. Under dry sowing, TMX-Dif-Mef and Dif-Mef exhibited significantly greater NDVI slopes than the control regardless of genotype during the early growth phase ($p < 0.00001$) (Fig. 4). However, only durum wheat cv. Cirno had greater early growth slopes with TMX-Dif-Mef and Dif-Mef compared to Vit-Dac ($p \leq 0.00003$); bread wheat cv. Roelfs did not ($p \geq 0.43$, data not shown). There was no significant difference between TMX-Dif-Mef and Dif-Mef for either genotype ($p > 0.99$). Under wet sowing, there were no significant differences among seed treatments regardless of genotype.

During 2010–11, Dif-Mef and TMX-Dif-Mef generally had slightly greater NDVI slopes during early growth compared to the control and Vit-Dac treatments under both genotypes and both sowing systems, but these differences were not significant ($p > 0.13$ for Cirno, $p > 0.22$ for Roelfs) (Fig. 4). There was no significant difference between TMX-Dif-Mef and Dif-Mef for Cirno ($p = 0.14$) or Roelfs ($p = 0.87$) during early growth. There were no significant

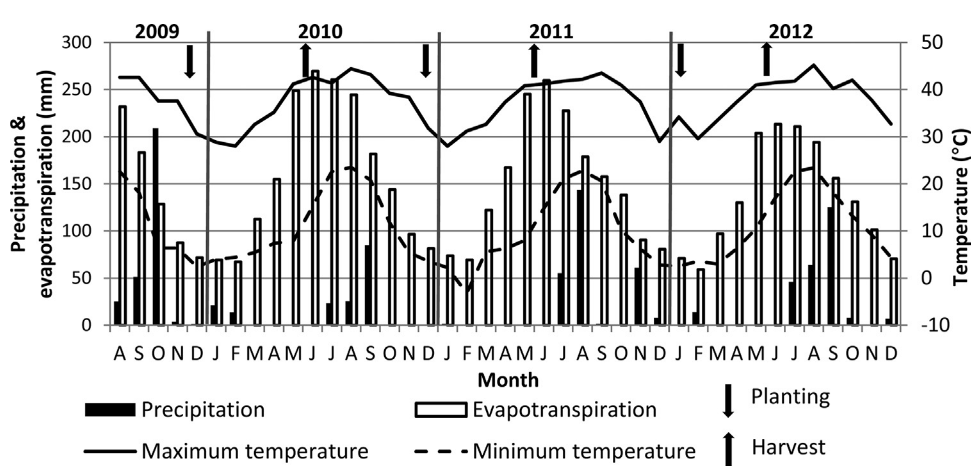


**Fig. 1.** Climate data during the study period. Maximum and minimum temperatures are absolute temperatures for each month, not average maximum and minimum temperatures.

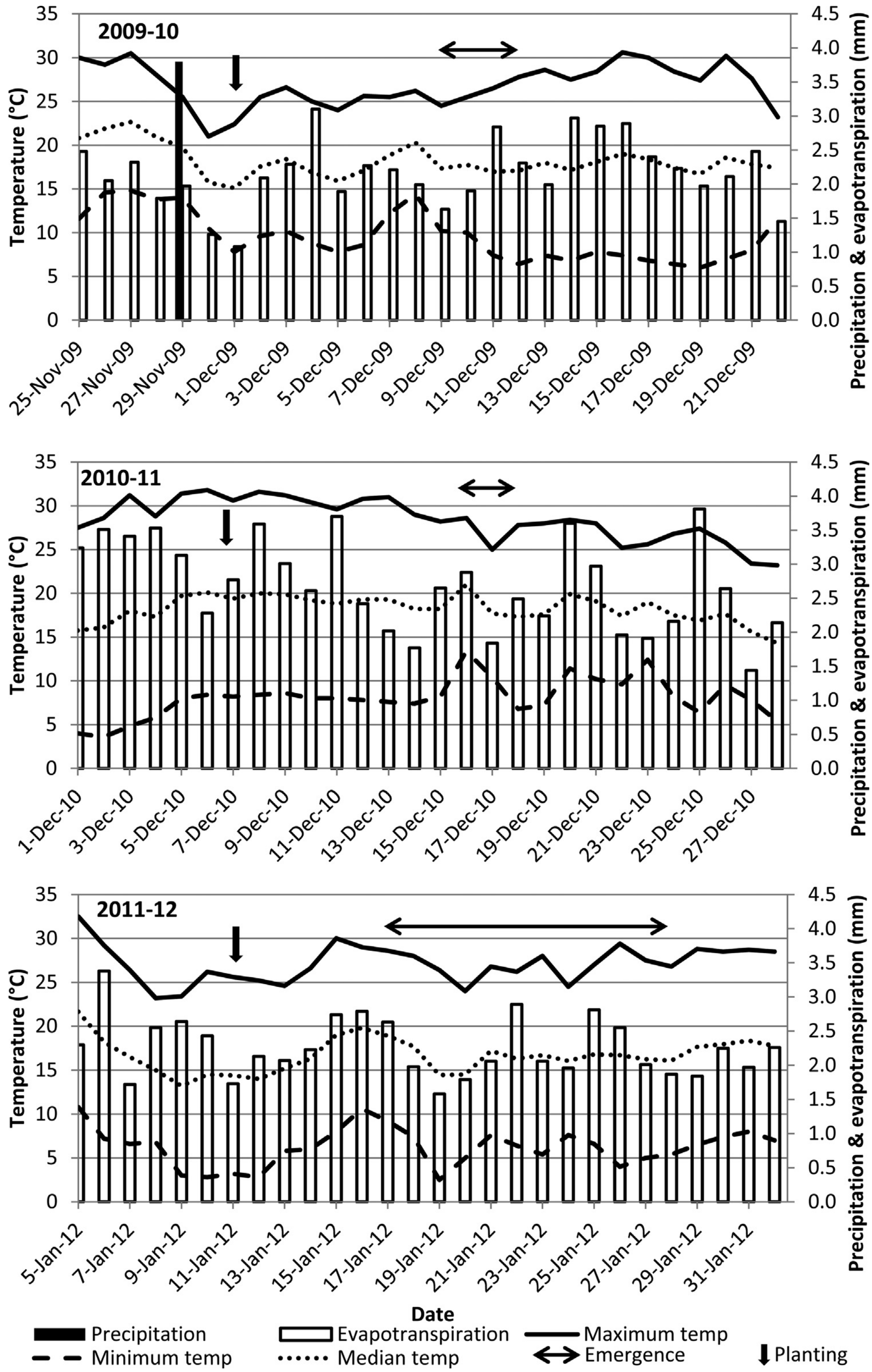


**Fig. 2.** Climatic conditions one week before and three weeks after sowing during the study period. Emergence dates correspond to 50% emergence.

differences under wet sowing for either genotype ($p > 0.60$). There was a dip in NDVI values after the frost event around 65 days after sowing.

During the 2011–12 season, all seed treatments had significantly greater NDVI slopes during early growth compared to the control under dry sowing regardless of genotype, while Dif-Mef and TMX-Dif-Mef exhibited the highest values among seed treatments (Fig. 4). During this phase, TMX-Dif-Mef and Dif-Mef NDVI slopes were significantly greater than Vit-Dac only for bread wheat cv. Roelfs ($p \leq 0.024$, data not shown); under durum wheat cv. Cirno they were not different ($p > 0.84$). The early growth slopes of TMX-Dif-Mef and Dif-Mef were significantly different only for Roelfs ($p = 0.0044$) but not for Cirno ($p = 0.58$). Under wet sowing, there were no significant differences among seed treatments ($p > 0.34$). The chronosequence appears truncated because data were not acquired during senescence.

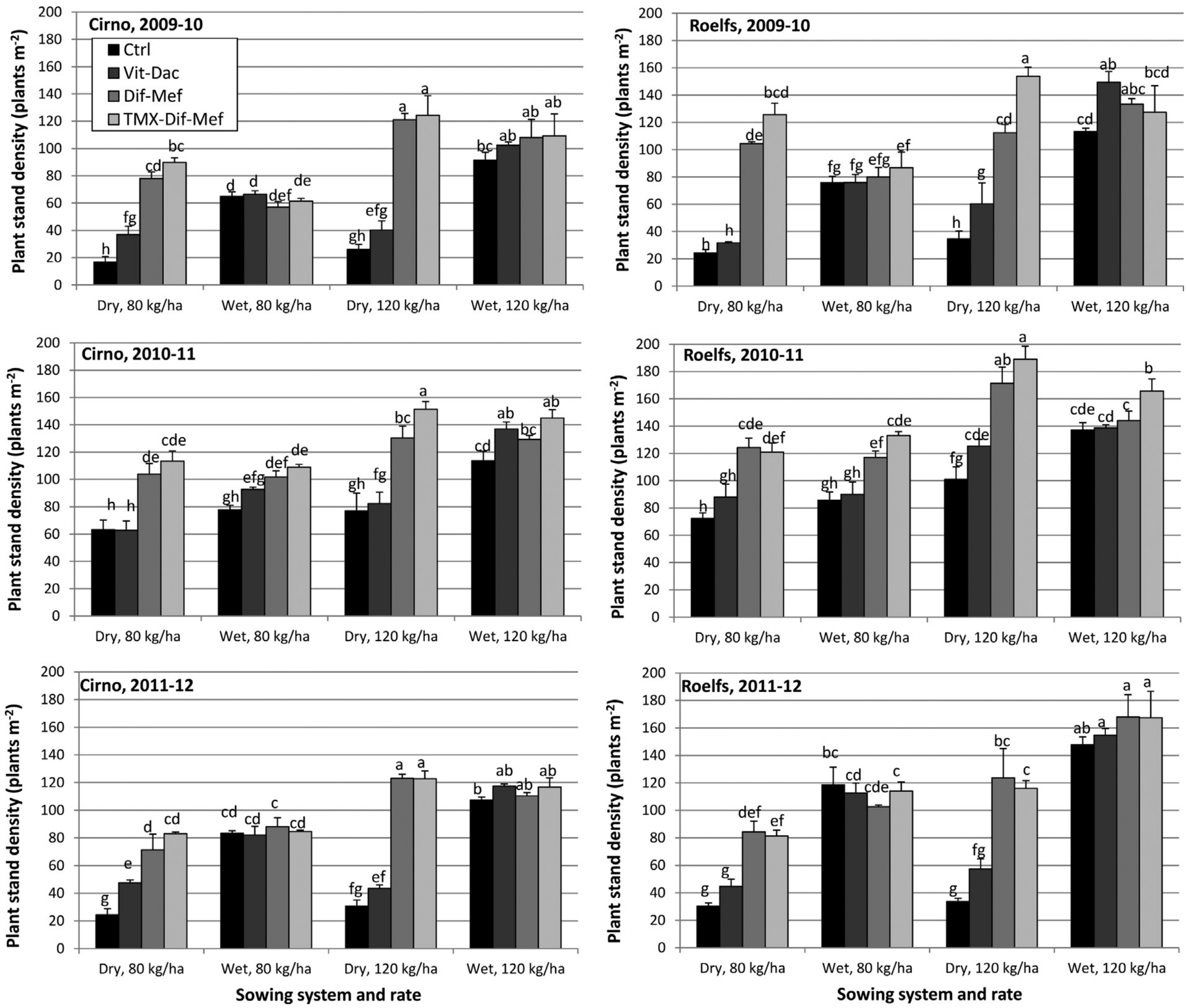


**Fig. 3.** Plant stand density of durum (cv. Cirno) and bread (cv. Roelfs) wheat under wet and dry sowing at two sowing rates (80 and 120 kg $ha^{-1}$) and four seed treatments over three seasons. Error bars represent positive standard errors of means. The seed treatments were 1. None (Ctrl), 2. Carboxin + thiram + chlorothalonil (Vit-Dac), 3. Difenoconazole + mefenoxam (Dif-Mef), and 4. Difenoconazole + mefenoxam + thiamethoxam (TMX-Dif-Mef). Within a graph, means followed by the same letter are not significantly different at $p < 0.05$.

During the plateau phase of NDVI curves, NDVI values under wet sowing were not significantly different between any treated seed and the control during two of the three seasons (Fig. 4). Under dry sowing with the durum wheat cv. Cirno, TMX-Dif-Mef had consistently higher NDVI numerical values than all other seed treatments, but was not significantly different than the other chemical treatments during any season. During two (2009–10 and 2011–12) of the three seasons, any chemical treatment was significantly higher than the control for cv. Cirno. Similarly, under dry sowing with the bread wheat cv. Roelfs, all chemical treatments were significantly higher than the control during two (2009–10 and 2011–12) of the three seasons. There were no significant differences among any of the chemical treatments during two (2009–10 and 2010–11) of the three seasons.

### 3.5. Grain yield

Under dry sowing at both sowing rates, the seed treatment TMX-Dif-Mef gave significantly higher yields during two of the three seasons compared to the control and Vit-Dac (Fig. 5). During 2009–10, the highest yields were achieved using durum wheat cv. Cirno under dry sowing conditions with Dif-Mef or TMX-Dif-Mef. A similar result was observed for the bread wheat cv. Roelfs under dry sowing; that is, significantly higher yields were observed using Dif-Mef or TMX-Dif-Mef at both sowing rates during 2009–10 compared to the control and Vit-Dac. Seed treatments did not generally affect grain yield during 2010–11 under any sowing system or genotype. During 2011–12 under dry sowing, the seed treatment TMX-Dif-Mef generally gave significantly higher yields than any other seed treatment within a sowing rate. Under wet sowing during this same season, TMX-Dif-Mef generally gave numerically higher yields than other seed treatments, although the difference was not always significant.

Under dry sowing, averaged over all seasons, rates, and genotypes, the seed treatment TMX-Dif-Mef yielded 7094 kg $ha^{-1}$, a 9.76% increase over the Vit-Dac yield of 6463 kg $ha^{-1}$, although it should be emphasized that the 2010–11 season did not produce the increased yields observed in other seasons. There were significant seed treatment × genotype × sowing system interactions over all three seasons ($p < 0.0017$), the highest order significant interaction observed during all three seasons.

Under wet sowing, TMX-Dif-Mef seed treatment effects were not consistent. In some cases, TMX-Dif-Mef significantly increased yield compared to the control (e.g., Roelfs, 2011–12, Fig. 5), and in some cases TMX-Dif-Mef decreased yields (e.g., Cirno, 2009–10). In most cases, there was no significant difference

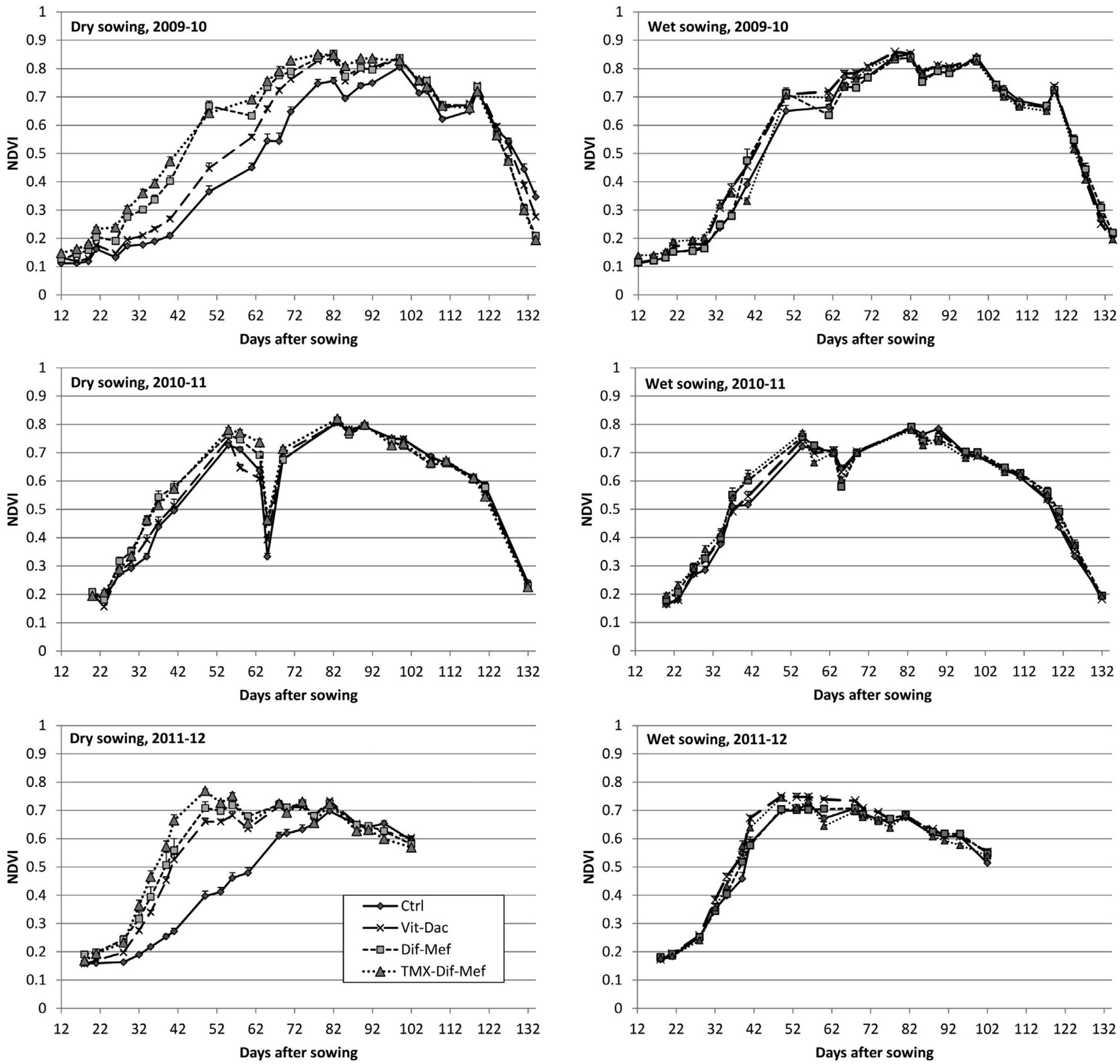


**Fig. 4.** Normalized Difference Vegetation Index (NDVI) vs. days after sowing for durum (cv. Cirno) wheat grown on dry- and wet-sown beds under four seed treatments over three growing seasons. The seed treatments were 1. None (Ctrl), 2. Carboxin + thiram + chlorothalonil (Vit-Dac), 3. Difenoconazole + mefenoxam (Dif-Mef), and 4. Difenoconazole + mefenoxam + thiamethoxam (TMX-Dif-Mef). Error bars represent positive standard errors of means. Means were averaged over two sowing rates (80 and 120 kg ha$^{-1}$).

between TMX-Dif-Mef and the control under wet sowing. The same may be said for comparisons of TMX-Dif-Mef to Vit-Dac.

## 4. Discussion

Clear and consistent differences in plant stand density, early season growth, and grain yield were observed between dry sowing and wet sowing. Since both Dif-Mef and TMX-Dif-Mef contain difenoconazole and mefenoxam, systemic fungicides, the results suggest that difenoconazole and/or mefenoxam suppressed a soil pathogen active under dry sowing but not under wet sowing. Indeed, mefenoxam was the only pesticide used with action against oomycetes, e.g. *Pythium*, though it may be noted that these are no longer classified as fungi. Factors that delay emergence can lead to *Pythium* infection. It is known that reduced wheat emergence in cold soils, even under dry conditions, can be due to *Pythium* infection (Smiley et al., 1996b). However, if *Pythium* were responsible for seedling non-emergence under dry sowing, root rot incidence would also be expected to have been higher under dry sowing compared to wet sowing; but this was not observed. Indeed, root rot scores were slightly lower under dry sowing than wet sowing, although not significantly so (Section 3.3). Indications that Dif-Mef and TMX-Dif-Mef pest control were not responsible for the results include root rot scores were not different among any of the treatments and there were generally no differences in plant stand densities among any seed treatments on wet-sown plots. The addition of the systemic insecticide TMX did not consistently increase plant stands compared to the Dif-Mef treatment and there was no history of Scarabaeidae larvae on the plots. However, it is possible that a pathogen or pest was active under dry sowing that was not active under wet sowing. Since the water requirements of bacteria are higher than those of fungi (Kouyeas, 1964; Griffin, 1969; Orchard and Cook, 1983; Guo et al., 2013), it may be that the prolonged presence of wet soils under wet sowing conditions prior to planting favored beneficial bacterial populations that out-competed pathogenic fungal or oomycete populations.

Another possibility is that there were some physical or environmental limitations, such as excess moisture or low soil temperature, present under dry sowing that were not present

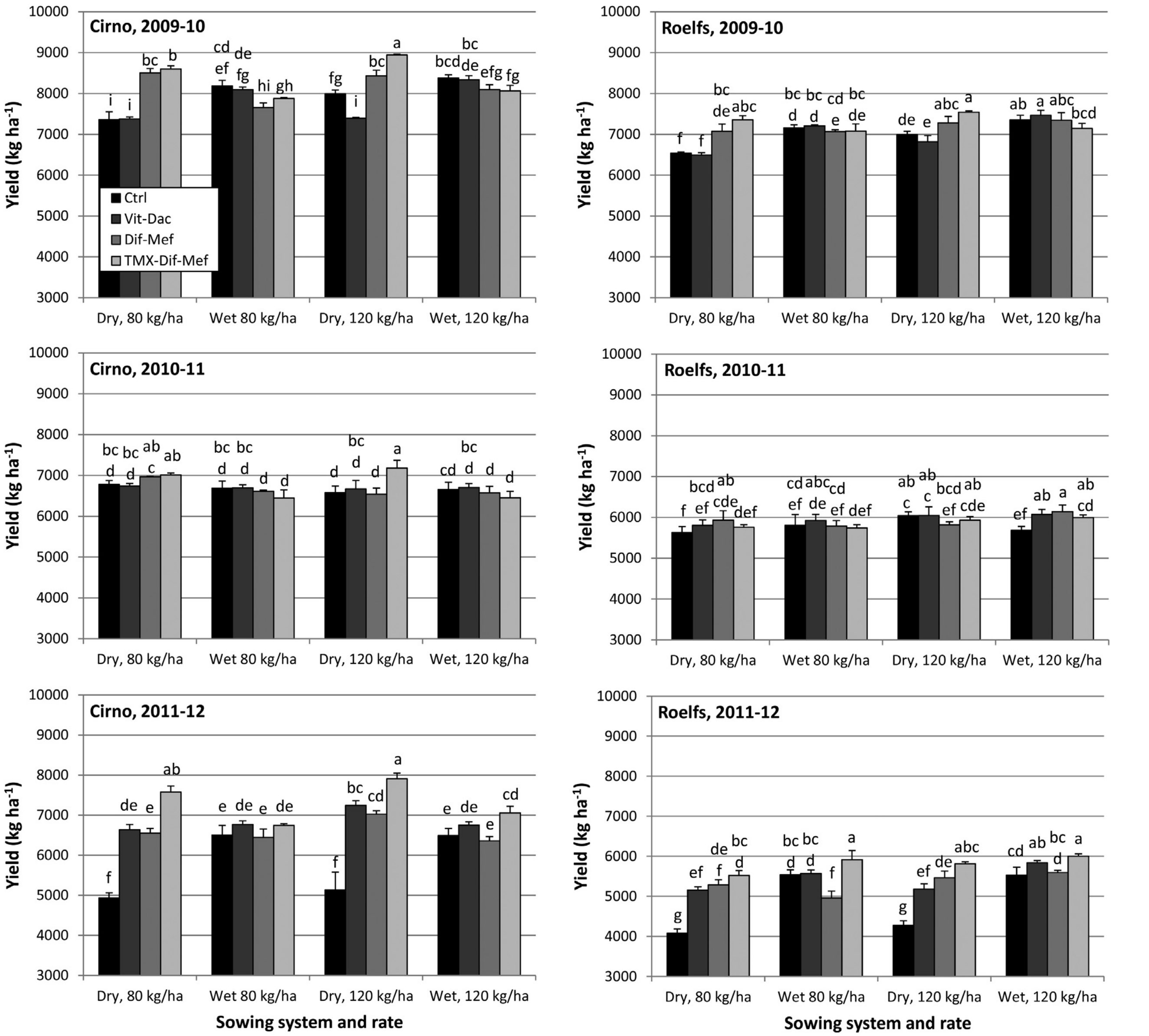


**Fig. 5.** Grain yield at 12% moisture of durum (cv. Cirno) and bread (cv. Roelfs) wheat under wet and dry sowing at two sowing rates and four seed treatments over three seasons. Error bars represent positive standard errors of means. The seed treatments were 1. None (Ctrl), 2. Carboxin + thiram + chlorothalonil (Vit-Dac), 3. Difenoconazole + mefenoxam (Dif-Mef), and 4. Difenoconazole + mefenoxam + thiamethoxam (TMX-Dif-Mef). Within a graph, means followed by the same letter are not significantly different at $p < 0.05$.

under wet sowing which led to non-emergence. It seems that the effect of seed treatment on plant stand density under dry sowing was more pronounced with lower temperatures during germination and emergence (Figs. 2–3). However, it remains unclear why Dif-Mef and TMX-Dif-Mef seed treatments would overcome physical limitations, if present, under dry sowing. Smiley et al. (1996a) investigated deeply seeded wheat in the Pacific Northwest and concluded that Dif-Mef improved emergence compared to carboxin + thiram and imidacloprid, but Babadoost and Islam (2003) found that mefenoxam alone did not have any effect on germination or vigor of pumpkin (*Cucurbita pepo* L.) seed. It is possible that the different physical conditions under dry sowing compared to wet sowing influenced microbial populations which induced a biological effect on germination and emergence.

The increased plant stand densities generally translated into greater NDVI slopes during early growth, with the notable exception of the 2010–11 season. Despite clear differences in plant stand density under dry sowing during 2010–11 (Fig. 3), those differences were not manifested in NDVI readings. The lowest plant stand density in 2009–10 and 2011–12 was approximately 20 plants $m^{-2}$, whereas in 2010–11 it was approximately 60 plants $m^{-2}$. The increased plant stand densities in 2010–11 resulted in greater ground coverage after tillering, which resulted in a lack of NDVI and yield differences that season. The frost in February 2011 may have reduced differences in yield that season.

Dif-Mef and TMX-Dif-Mef increased yields in two of three seasons under dry sowing compared to the control and Vit-Dac (Table 1). For cv. Cirno, dry sowing with TMX-Dif-Mef achieved significantly higher yields than wet sowing during all three seasons, regardless of sowing rate. Under wet sowing, however, there were few significant differences among seed treatments. The yield effects observed under dry sowing can be mainly explained by the altered plant stand densities observed in all seasons of the present study.

**Table 1**
Overview of results of seed treatment effects on select parameters under wet and dry sowing in Ciudad Obregón. Data are for three growing seasons. Fractional numbers indicate the number of years a positive significant response was consistently found.

| | Plant stand | | Initial growth | | Root rot | | Grain yield | |
|---|---|---|---|---|---|---|---|---|
| | DW[a] | BW[c] | DW | BW | DW | BW | DW | BW |
| Difenoconazole + mefenoxam vs. Control | | | | | | | | |
| Wet sowing | –[b] | – | – | – | – | – | – | – |
| Dry sowing | 3/3 | 3/3 | 2/3 | 2/3 | – | – | 2/3 | 2/3 |
| Difenoconazole + mefenoxam + thiamethoxam vs. Control | | | | | | | | |
| Wet sowing | 1/3 | 1/3 | – | – | – | – | – | 1/3 |
| Dry sowing | 3/3 | 3/3 | 2/3 | 2/3 | – | – | 2/3 | 2/3 |
| Difenoconazole + mefenoxam + thiamethoxam vs. Difenoconazole + mefenoxam | | | | | | | | |
| Wet sowing | – | – | – | – | – | – | – | 1/3 |
| Dry sowing | – | – | – | – | – | – | 1/3 | – |

[a] DW durum wheat.
[b] no positive effect found.
[c] BW bread wheat.

Under dry sowing with the durum wheat cv. Cirno, yield was significantly higher with TMX-Dif-Mef than with Dif-Mef alone in the majority of cases, but this was not the case with the bread wheat cv. Roelfs. Such a genotype × system × chemical interaction may demonstrate the utility of seed treatments to enhance yields for specific genotypes under specific agronomic environments. Although the mechanism by which crop enhancement effects occur is currently poorly understood, it may be related to hormetic or eustress effects, in which a toxin at low dosage produces a favorable effect (Tesche, 1992; Mattson, 2008; Belz et al., 2011). It is also possible that root mass increased due to TMX seed treatment (Macedo and Camargo e Castro, 2011; Perelló and Bello, 2011; Colman et al., 2012) or that the seed treatment altered the expression of genes that allow the seedling to overcome certain abiotic stresses. Larsen and Falk (2013) also considered the possibility that improved germination of treated wheat seed may be independent of soil or seed-borne disease when they observed increased cold stress tolerance with TMX-Dif-Mef. The crop enhancement effects of TMX are claimed to be most efficacious under abiotic stress, including drought, heat, cold, low pH, and soil salinity (Almeida et al., 2012a,b; Senn et al., 2004; Maienfisch, 2012).

TMX-Dif-Mef frequently resulted in higher plant stand densities, early growth rates, and sometimes yield than Dif-Mef alone, which may be attributable to abiotic stress resistance. There are biochemical bases by which one might expect TMX to induce resistance to abiotic stresses. A study by Ford et al. (2010) found that soil application of 4 mM clothianidin, an important metabolite of TMX in cotton (*Gossypium hirsutum*) plants (Nauen et al., 2003), altered the expression of 1790 genes in *Arabidopsis thaliana*, some of which may be related to water stress (Rajjou et al., 2006). Facile cleavage of clothianidin to the corresponding carboxylic acid resulted in salicylic acid-induced plant defense responses in *A. thaliana* (Ford et al., 2010). Salicylic acid-related processes primed seed metabolism, mobilized seed proteins, enhanced translation quality, and promoted antioxidant synthesis in *A. thaliana*, all of which increased seedling vigor and responses to water stress (Rajjou et al., 2006). It is conceivable that water-related stress may occur under conditions of dry sowing, where the seed is first placed in dry soil, then subjected to saturation and thus excess moisture, though it is acknowledged that most water-related stress studies refer to moisture deficit, not excess moisture.

More research is required to determine whether the observed effects of seed treatments are biological, physical, or genetic in nature. Soil sterilization under field conditions could help to distinguish between biological and physical factors and may help to uncover the underlying mechanism when combined with a detailed study of microbial populations during germination and emergence. An investigation of the seed treatment effects on a wider range of wheat genotypes would shed light on the genotype × chemical × system interactions observed in the present results.

## 5. Conclusion

In dry sowing systems, Dif-Mef and TMX-Dif-Mef seed treatments increased early season plant stands and grain yield. Dry sowing durum wheat cv. Cirno with TMX-Dif-Mef out-yielded all other wet sowing treatments. The trends were corroborated by increased early season NDVI slopes, and were not related to root rot. This is an important finding given dwindling water resources in the region, which will compel producers to use less water and may soon limit their ability to utilize wet sowing. Producers who wish to conserve water by transitioning into conservation agriculture with dry sowing may avoid yield losses, or even increase yield, with the use of Dif-Mef and TMX-Dif-Mef seed treatments. In wet sowing the effects of seed treatments on stand establishment, growth and yield were usually not significant. More research is required to understand the underlying mechanism of the observed effects of the seed treatments in dry sowing.

## Acknowledgements

This research was conducted under the WHEAT CGIAR Research Program. The authors wish to acknowledge funding by the Syngenta Corporation of the CIMMYT-developed project "Crop enhancement in interaction with crop management systems and genotypes: preparing an integrated yield increase." We thank Manuel de Jesus Ruiz Cano, Jesus Gutierrez Angulo, Juan de Dios Sanchez Lopez, Alvaro Zermeño, Juan Gastelum Flores, Beatriz Martínez Ortiz, and Ana Garcia Lopez for technical assistance.

## References

Almeida, A.d.S., Lauxen, L.R., Villela, F.A., Meneghello, G.E., Tillmann Â, M.A., 2012a. Physiological performance of wheat and barley seeds treated with bioactivator. Am. J. Exp. Agric. 2, 90–101.
Almeida, A.d.S., Villela, F.A., Tillmann Â, M.A., Meneghello, G.E., 2012b. Insecticide thiamethoxam: A bioactive action on carrot seeds (Daucus carota L.). In: Soloneski, S., Larramendy, M. (Eds.), Insecticides – Basic and Other Applications. InTech, Rijeka, Croatia, pp. 3–16.
Babadoost, M., Islam, S.Z., 2003. Fungicide seed treatment effects on seedling damping-off of pumpkin caused by *Phytophthora capsici*. Plant Dis. 87, 63–68.
Belz, R.G., Cedergreen, N., Duke, S.O., 2011. Herbicide hormesis – can it be useful in crop production? Weed Research (Oxford) 51, 321–332.

Bivand, R., Rowlingson, B., Diggle, P., Petris, G., Eglen, S., 2013. SPLANCS: Spatial and space-time point pattern analysis functions .

Borlaug, N.E., 2007. Sixty-two years of fighting hunger: personal recollections. Euphytica 157, 287–297.

Braun, H.J., Payne, T., 2012. Mega-Environment breeding. In: Reynolds, M.P., Pask, A. J.D., Mullan, D.M. (Eds.), Physiological breeding I: interdisciplinary approaches to improve crop adaptation. CIMMYT, Mexico, D.F.

Cheema, M.S., Muhammad, A., Ahmad, M.S., 1985. Impact of dry sowing methods on the yield of late sown wheat. J. Agric. Res. Pakistan 23, 175–178.

Colman, B.A., Masson, G.d.L., Missio, H.G., Nunes, A.d.S., Ceolin, A.C., 2012. Effect of addition insecticides to soybean seed treatment with biostimulant. Revista Verde de Agroecologia e Desenvolvimento Sustentável 7. Nota tecnica 45–48.

Dan, L.G.d.M., Dan, H.A., Braccini, A.L., Albrecht, L.P., Ricci, T.T., Piccinin, G.G., 2011. Performance of soybean seeds treated with insecticides and subjected to different storage periods. Revista Brasileira de Ciências Agrárias 6, 215–222.

Figueroa-López, P., Félix-Fuentes, J.L., Fuentes-Dávila, G., Valenzuela-Herrera, V., Chávez-Villalba, G., Mendoza-Lugo, J.A., 2010. CIRNO C2008 nueva variedad de trigo cristalino con alto rendimiento potencial para el estado de Sonora. Revista Mexicana de Ciencias Agrócolas 1, 739–744.

Figueroa-López, P., Fuentes-Dávila, G., Valenzuela-Herrera, V., Chávez-Villalba, G., Félix-Fuentes, J.L., Mendoza-Lugo, J.A., 2011. 'ROELFS F2007', nueva variedad de trigo harinero para el noroeste de Máxico. Revista Fitotecnia Mexicana 34, 221–223.

Ford, K.A., Casida, J.E., Chandran, D., Gulevich, A.G., Okrent, R.A., Durkin, K.A., Sarpong, R., Bunnelle, E.M., Wildermuth, M.C., 2010. Neonicotinoid insecticides induce salicylate-associated plant defense responses. Proc. Natl. Acad. Sci. U. S. A. 107, 17527–17532.

Govaerts, B., Verhulst, N., Sayre, K.D., Kienle, F., Flores, D., Limon-Ortega, A., 2009. Implementing conservation agriculture concepts for irrigated wheat based systems in northwest Mexico; a dynamic process towards sustainable production. In: Rattan, R.K., Mathur, V.C., Sharma, A.R., Manjaiah, K.M., Datta, S.P., Chakraborty, A., Chakraborty, D., Datta, K.K. (Eds.), 4th World Congress on Conservation Agriculture: Innovations for Improving Efficiency, Equity and Environment. M/s Print Process, New Delhi, India.

Griffin, D.M., 1969. Soil water in the ecology of fungi. Annu. Rev. Phytopathol. 7, 289–310.

Guo, Q., Wang, W., Liu, Y., Wen, X., Liao, Y., 2013. Effects of soil moisture and temperature on soil microbial ecological environment and soil respiration in rainfed wheat field. Res. Crops 14, 374–381.

Hobbs, P.R., 2007. Conservation agriculture: what is it and why is it important for future sustainable food production. J. Agric. Sci. 145, 127–137.

Hobbs, P.R., Sayre, K., Gupta, R., 2008. The role of conservation agriculture in sustainable agriculture. Philos. Trans. R. Soc. B – Biol. Sci. 363, 543–555.

Horii, A., McCue, P., Shetty, K., 2007. Enhancement of seed vigour following insecticide and phenolic elicitor treatment. Bioresour. Technol. 98, 623–632.

IUSS Working Group WRB, 2007. World reference base for soil resources 2006, first update 2007. In: WRB I.W.G (Ed.), World Soil Resources Reports No. 103. FAO, Rome, pp. 128.

Kanwar, R.S., Mann, S.S., Bhagat, S., 2013. Management of Heterodera avenae in wheat by dry sowing. Annals Plant Prot. Sci. 21, 460–462.

Kouyeas, V., 1964. An approach to the study of moisture relations of soil fungi. Plant Soil 20, 351–363.

Larsen, R.J., Falk, D.E., 2013. Effects of a seed treatment with a neonicotinoid insecticide on germination and freezing tolerance of spring wheat seedlings. Can. J. Plant Sci. 93, 535–540.

Lenth, R.V., 2013. LSMEANS: Least-squares means .

Macedo, W.R., Camargo e Castro, P.R.d., 2011. Thiamethoxam: molecule moderator of growth, metabolism and production of spring wheat. Pestic. Biochem. Physiol. 100, 299–304.

Macedo, W.R., Fernandes, G.M., Possenti, R.A., Lambais, G.R., Camargo e Castro, P.R. d., 2013. Responses in root growth, nitrogen metabolism and nutritional quality in *Brachiaria* with the use of *thiamethoxam*. Acta Physiol. Plant. 35, 205–211.

Maienfisch, P., 2012. Six-membered heterocycles: thiamethoxam and AKD 1022. In: Krämer, W., Schirmer, U., Jeschke, P., Witschel, M. (Eds.), Modern Crop Protection Compounds. Wiley-VCH Verlag GmbH & Co., Weinheim, pp. 1203–1225.

Mattson, M.P., 2008. Hormesis defined. Ageing Res. Rev. 7, 1–7.

McBeath, T.M., McLaughlin, M.J., Kirby, J.K., Armstrong, R.D., 2012. Dry soil reduces fertilizer phosphorus and zinc diffusion but not bioavailability. Soil Sci. Soc. Am. J. 76, 1301–1310.

Nauen, R., Ebbinghaus-Kintscher, U., Salgado, V.L., Kaussmann, M., 2003. Thiamethoxam is a neonicotinoid precursor converted to clothianidin in insects and plants. Pestic. Biochem. Physiol. 76, 55–69.

Oladeebo, J.O., Mkhonta, C.S., 2013. Does conservation agriculture matters in Swazis' economy? Evidence from maize producing farmers in Ngwempisi rural development area of Swaziland J. Env. Earth Sci. 3, 79–85.

Orchard, V.A., Cook, F.J., 1983. Relationship between soil respiration and soil moisture. Soil. Biol. Biochem. 15, 447–453.

Pask, A.J.D., Pietragalla, J., Mullan, D.M., Reynolds, M.P., 2012. Physiological Breeding II: A Field Guide to Wheat Phenotyping. CIMMYT, Mexico, D.F.

Perelló, A.E., Bello d, G.M., 2011. Suppression of tan spot and plant growth promotion of wheat by synthetic and biological inducers under field conditions. Ann. Appl. Biol. 158, 267–274.

Pinheiro, J., Bates, D., DebRoy, S., Sarkar, D., 2013. NLME: Linear and Nonlinear Mixed Effects Models .

Pretty, J., Toulmin, C., Williams, S., 2011. Sustainable intensification in African agriculture. Int. J. Agric. Sust. 9, 5–24.

R Core Team, 2013. R: A Language and Environment for Statistical Computing. R Core Team, Vienna, Autria.

Rajjou, L., Belghazi, M., Huguet, R., Robin, C., Moreau, A., Job, C., Job, D., 2006. Proteomic investigation of the effect of salicylic acid on Arabidopsis seed germination and establishment of early defense mechanisms. Plant Physiol. 141, 910–923.

SAS, 2013. SAS System for Windows 9.3. Cary, NC.

Saxton, A.M., 1998. A macro for converting mean separation output to letter groupings in Proc Mixed. Proceedings of 23rd SAS Users Group Intl., Cary, NC. SAS InstituteNashville, TN. , pp. 1243–1246.

Schillinger, W.F., Cook, R.J., Papendick, R.I., 1999. Increased dryland cropping intensity with no-till barley. Agron. J. 91, 744–752.

Senn, R., Hofer, D., Thieme, T., Zang, L., 2004. Method for Improving Plant Growth. US 6753296 B1. Syngenta Crop Protection, Inc. 6753296.

Smaling, E.M.A., Bouma, J., 1992. Bypass flow and leaching of nitrogen in a Kenyan Vertisol at the onset of the growing season. Soil Use Manag. 8, 44–47.

Smiley, R.W., Patterson, L.M., Rhinhart, K.E.L., 1996a. Fungicide seed treatment effects on emergence of deeply planted winter wheat. J. Prod. Agric. 9, 564–570.

Smiley, R.W., Patterson, L.M., Shelton, C.W., 1996b. Fungicide seed treatments influence emergence of winter wheat in cold soil. J. Prod. Agric. 9, 559–563.

Tesche, M., 1992. Immediate and long-term (memory) responses of Picea abies to a single growing season of $SO_2$-exposure or moderate drought. For. Ecol. Manag. 51, 179–186.

Soil Survey Staff, U.S.D.A., 2003. Keys to Soil Taxonomy. United States Department of Agriculture, Natural Resources Conservation Service, Washington, DC, pp. 332.

Verhulst, N., Deckers, J., Govaerts, B., 2009. Classification of the Soil at Cimmyt's Experimental Station in the Yaqui Valley Near Ciudad Obregón, Sonora, México., Cimmyt Report. CIMMYT, México, D.F.

Verhulst, N., Govaerts, B., Sayre, K.D., Sonder, K., Romero-Perezgrovas, R., Mezzalama, M., Dendooven, L., 2012. Conservation agriculture as a means to mitigate and adapt to climate change, a case study from Mexico. In: Wollenberg, E., Nihart, A., Tapio-Biström, M.-L., Grieg-Gran, M. (Eds.), Climate Change Mitigation and Agriculture. Earthscan, Oxon, pp. 287–300.