

QUANTIFYING YIELD LOSSES DUE TO BARLEY YELLOW DWARF ON WINTER  
WHEAT IN KANSAS USING DISEASE PHENOTYPIC DATA

by

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## ABSTRACT

Barley yellow dwarf (BYD) is one of the most important wheat diseases in the state of Kansas. Despite the development of cultivars with improved levels of resistance to BYD, little is known about the impact that this resistance has on yield loss from the disease. The intent of this research was to estimate yield loss in winter wheat cultivars in Kansas due to BYD and quantify the reduction in losses associated with resistant cultivars. During seven years, BYD disease incidence was visually assessed on numerous winter wheat cultivars in replicated field nurseries. Cultivars were planted about three weeks early to promote disease. When grain yields were regressed against BYD incidence scores, negative linear relationships significantly fit the data for each year and for the combined dataset covering all seven years. The models showed that, depending upon the year, 19-48% (average 33%) of the yields was explained by BYD incidence. For the combined dataset, 29% of the relative yield was explained by BYD incidence. The models predicted that cultivars showing high disease incidence had 25-86% (average 49%) less yield than a hypothetical cultivar that showed zero incidence. Using the models, the moderate level of resistance in the cultivar Everest was calculated to reduce yield loss from BYD by about 73%. Therefore, utilizing visual BYD symptom evaluations in Kansas, coupled with grain yields, is useful to estimate yield loss from the disease. Furthermore, linear models that incorporate those parameters can be used to calculate the impact of improving cultivar resistance to BYD on yield losses.

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## INTRODUCTION

Barley yellow dwarf (BYD) is caused by strains of the *Barley yellow dwarf virus* (BYDV) and *Cereal yellow dwarf virus* (CYDV) of the Luteoviridae family. The disease is capable of producing significant yield losses in several different crop species including wheat (*Triticum aestivum*) (4). It is considered one of the most economically important virus diseases of cereal crops in the world (9; 20; 27). In the state of Kansas, it is the fourth most important wheat disease in terms of average estimated yield losses (1).

The virus particles have an icosahedral shape and encase a single stranded positive RNA genome. The virus is phloem limited within the host plant and it cannot be transmitted without the aid of its hemipteran vector (13; 18). The serotype most common to Kansas agronomic cereal crops is PAV although the closely-related RPV strain of CYDV is also common (Bockus, unpublished). Both of these species are transmitted by either the bird cherry-oat aphid (*Rhopalosiphum padi*) or the greenbug (*Schizaphis graminum*) (28). The virus persists in the aphid vectors in a circulative, non-propagative manner (18).

BYD symptoms can vary depending on the host: however, even within a host such as wheat, symptoms can differ widely among cultivars. Visual foliar symptoms can vary between yellow leaf discolorations to oranges, reds, and purples. They begin at the leaf tip and spread toward the leaf base, particularly on the flag leaves (23). Stunting and a decrease in kernel size and kernel number per head can occur as well leading to an overall decrease in yield in the infected plants (17; 30). The stunting symptom is usually associated with early infections of the wheat plant by the pathogen.

BYDV is thought to over season in native perennial grasses and volunteer host plants which are considered to be the main viral/vector reservoir (19; 22). The virus is often spread in the fall when viruliferous aphid species fly or are blown from the reservoirs into winter wheat fields at emergence (23). Infection can also occur in the spring; however, those are significantly less important than the ones that occur in the fall (14). It was shown that the severity of BYD infection, and subsequent wheat yield loss, are positively correlated with how old the plant is at infection (14). Additionally, fall infections were shown to decrease the number of heads, number of seeds per head, as well as tiller height (16). A study looking at possible BYD control measures determined that a critical treatment time is during the emerging stages of the wheat life cycle, confirming the importance of limiting early infections by BYDV (23).

Yield has been shown to be greatly reduced when wheat is infected with BYDV. When comparing symptomatic with adjacent nonsymptomatic sites of winter wheat and barley in Virginia, BYD significantly reduced tiller height, head number, seed number, number of seed per head, 1000 seed weight, and yield (17). Seed number, number of seed per head and yield were reduced the most with yield reduced by 34%. Additionally, the amount of BYDV titer in wheat was shown to be directly correlated with the amount of resistance; resistant lines had significantly less titer (2). In the southeastern United States, researchers stated that data on BYD yield losses were sparse; however, they indicated that there were serious problems in many years due to the virus (15). Grain yield was shown to be “strongly associated with kernel number per plant” with a 36% decrease in research trials with BYD infection only and a 50% reduction in kernels when infected with BYD and infested with aphids (*R. padi*) at the same time (29). In the same study, it was shown that yield had been decreased by 46% in BYD infected wheat and 58% when wheat was infested with *R. padi* and simultaneously infected with BYDV. In a different

study, researchers concluded that yield had been decreased by a total of 34% in the host (16). In a 1990 report, regional yield losses in wheat in the U.S. due to the viruses were estimated at between 2% and 10% (25). In summary, there are numerous reports that point to the BYD viruses' capacity to cause 30-60% loss in wheat plants and up to 10% loss across large regions.

The purpose of this study was to estimate yield loss due to barley yellow dwarf in winter wheat cultivars in Kansas. Preliminary reports indicated 22-39% loss due to BYD in susceptible winter wheat cultivars in Kansas (5; 6). However, many cultivars that are grown in the state have some level of resistance to BYD (rating of 4 on a 1 to 9 scale where 1 is highly resistant and 9 is highly susceptible). There are no reports of how that resistance affects the yield loss. Therefore, this research sought to fill that gap and involved a novel method to estimate BYD yield loss for winter wheat cultivars in Kansas. BYD incidence ratings for numerous cultivars were collected over the course of seven years and regressed against their relative grain yields in replicated disease nurseries. Although many parameters can affect grain yields (inherent yielding ability, response to abiotic stresses, response to other diseases), the hypothesis was that, given enough data points, grain yield would be negatively associated with BYD incidence. Furthermore, if that is true, the resulting model equations could be used to calculate the impact that resistance has on yield loss from BYD.

## **MATERIALS & METHODS**

All experiments were conducted at the Plant Pathology Rocky Ford Experimental Field located near Manhattan, Kansas. The soil type at that location is a silty clay loam (pH = 6.5) and all experiments were sown in an area that had been fallowed the year before. A total of seven



experiments were conducted beginning in 2005 through 2013 (Table 1). Data from 2007 and 2009 were not available; therefore, those years were not included in the analyses. Each experiment used a randomized complete block design with five replications with 24-48 entries each year. Entries consisted of different winter wheat cultivars or advanced breeding lines (Table 1). The field plots consisted of single rows, each 2.3 m in length and 50 cm apart in all directions. On average, the planting date occurred three weeks early for North Eastern KS. Early planting facilitates BYD spread to winter wheat due to increased aphid presence and subsequent feeding at higher temperatures. Grain was harvested with a plot combine at the normal time for the region. To verify the species of virus present each year, several leaves were collected from the plot area after heading and subjected to Enzyme-Linked Immunosorbent Assay (ELISA) to test for the PAV species of BYDV and RPV species of *Cereal yellow dwarf virus*. PAV was the dominant species each year but RPV was also detected. These are the two species that are most common in wheat in Kansas with only trace amounts of other species detected (Bockus, unpublished).

Infection by BYDV occurred due to the natural activity of various aphid species (mostly the bird cherry-oat aphid, *Rhopalosiphum padi*) and was promoted by an early planting date. Disease incidence was visually observed and recorded weekly beginning after heading and ending at the onset of normal wheat senescence. At each rating date, the percentage of tillers with leaf symptoms was visually estimated for each plot. Fungicide treatments were used when needed to prevent leaf senescence from foliar diseases (leaf rust and tan spot). When foliar fungicides were used, they were applied at the early heading growth stage. Folicur 3.6F (38.7% tebuconazole) was applied at a rate of 292 ml/ha in 187 L water per hectare with a back-pack sprayer equipped with flat fan nozzles.

A total of 160 winter wheat cultivars or breeding lines were used in these experiments; however, the cultivars varied year to year (Table 1). Resistant and susceptible checks were included in each experiment and were selected based on the Kansas State University Cooperative Extension ratings (11). Ratings were on a 1-to-9 scale with 1 being highly resistant and 9 being highly susceptible. Resistant checks were those that had a rating of 4 and the susceptible checks were those with a rating of 9.

Disease incidence ratings were averaged across all rating dates and replications to determine the mean for each wheat entry. Mean grain yields for each entry were also calculated. For each experiment, mean incidence values were regressed against mean grain yield using the Excel software. To combine data across all seven years, incidence data were transformed by expressing them as a percentage of the entry showing the highest incidence. Similarly, mean grain yields were expressed as a percentage of the test average yield. Scatter graphs were constructed showing the linear model fit to the data utilizing Excel. Using the linear regression equations for each experiment, the potential yield loss for an experiment was calculated by comparison of the calculated yield of the entry showing the highest disease incidence with the calculated yield for a hypothetical entry that showed zero disease incidence. Data across all experiments were combined to produce an overall model for relative grain yield regressed against relative incidence of BYD.

## **RESULTS**

There was significant BYD pressure in the nurseries each year as indicated by the relatively high percentage of wheat plants displaying symptoms on susceptible cultivars (Fig. 1).

An example of the disease progress on moderately-resistant and susceptible cultivars is shown in Table 2. Depending upon the year, the most susceptible cultivars displayed between 29 and 83% incidence. This was due to the fact that the nurseries were planted 2-3 weeks earlier than the usual planting date for North East Kansas. Early planting of winter wheat cultivars in Kansas promotes BYD because the aphid vectors are more active in warmer temperatures. A continuum of BYD incidences, from relatively high to relatively low, was observed among cultivars for each year (Fig. 1). Some of the more resistant cultivars displayed less than 10% incidence, less than 20% of the symptoms seen on the susceptible cultivars in that experiment. The ELISA results showed that an average of 76% of the symptomatic plants had detectable BYDV or CYDV while none of the non-symptomatic plants had detectable virus.

For each of the seven years, data were incorporated into plots of the percentage symptomatic plants (*X* axis) and the grain yield (g/plot, *Y* axis) (Fig. 1). Linear regression models of these relationships significantly fit the data for all seven years (Table 3). The coefficients of determination ( $R^2$ ) ranged from 0.1924 in 2011 to 0.4788 in 2008 indicating that disease incidence values explained about 19 to 48% of the yield in these experiments. The *p*-values were highly significant (<0.0001) in 2005, 2006, 2008, and 2010 but were also significant (<0.02) for each of the other years. Equations of all of the regression lines for each year are given in Table 3.

The linear equations for each year were used to calculate the difference in yield between a cultivar that displayed the highest incidence that year with the yield of a hypothetical cultivar that was displaying zero symptoms. Using these differences, yield losses were calculated and are shown in Table 3.

To allow combining of data across years, yields were expressed as a percent of the test average for that year and BYD incidence as a percent of the highest-rating cultivar for that year. A graph was then created to compare relative grain yields with relative percent symptomatic plants across all the years (Fig. 2). Results from the combined data set displayed a highly significant ( $P < 0.0001$ ) linear relationship. A linear model was fit to the data and used as described above to calculate hypothetical yield loss for the data set combined over all seven years (Table 3). Using this equation, cultivars showing the highest disease incidence would be expected to have 48.9% less yield than a hypothetical cultivar that showed zero incidence.

## DISCUSSION

Yield losses in wheat due to BYD have been determined using several different methods. Some of these involve comparing yields of symptomatic areas and adjacent, non-symptomatic areas (16), maintaining disease-free checks (30), or using artificial inoculation methods (17; 26). With these techniques, large yield losses have been measured. For example, McKirdy and Jones (23) measured up to 43% loss, Hoffman and Kolb (17) up to 36%, Perry et al., (26) up to 35%, Herbert et al. (16) up to 34%, and Weisz et al., (30) up to 32%. Clearly, BYD can have a significant impact on wheat yields. In fact, for individual cultivars in some years, losses can exceed 60% (8). Similar losses (22-49%) have been reported for susceptible winter wheat cultivars in Kansas (5; 6; 7). However, the research reported here is the first use of visual disease phenotypic data, coupled with grain yields from many cultivars, to estimate yield losses.

Disease incidence (percentage of tillers expressing symptoms) was the method used to evaluate BYD in these experiments. Although many studies have shown a poor relationship

between BYD incidence and yield (10; 17; 30), results presented here showed a consistent statistically-significant relationship. Even within the citations listed above, there were years when symptom expression was negatively correlated with yield (17); therefore, there are environments where the association between these parameters is significant. Kansas appears to consistently be one of those environments because there was a significant relationship in each of the seven years of these experiments. Another difference between the studies cited above and this one is that data from 160 wheat cultivars and breeding lines and seven years (264 cultivar/years) went into our analyses compared with a relatively small number of cultivars in the earlier studies (11 or less). With such a large number of disease ratings correlated with yields, we were able to establish that barley yellow dwarf incidence can be useful to predict winter wheat yield losses in Kansas. Nevertheless, there are clearly environments where our models may not be valid.

Similar to results presented here, other studies have also shown a significant negative linear relationship between BYD incidence and grain yield (3; 24; 26). Nevertheless, there were important differences between the previous experiments and the current ones. Only a few cultivars were used in the previous experiments (6; 1; and 3, respectively) while 160 cultivars and breeding lines were used in our experiments. Additionally, the earlier studies used ELISA to detect the presence of the virus to estimate incidence while the current experiments used visual symptoms. Although it has been reported that visual symptoms underestimate the actual incidence of virus in plants (17), they were shown to be useful indicators of the impact that the virus has on yield under conditions in Kansas. Each method of determining BYD incidence has its own virtues. The use of ELISA or PCR would be the most accurate because the presence of the virus and virus titer are being measure in each plant. However, those methods are laborious

and may not be able to be utilized in breeding nurseries where there are large numbers of plots. Visual assessment of disease incidence is rapid and can be used in breeding nurseries where there are large numbers of entries. Despite the differences, our results corroborate these earlier findings that a significant negative linear relationship exists between BYD incidence and grain yield in some environments.

As noted above, there are reports that BYD incidence may not be correlated with grain yield and visual symptoms can underestimate actual virus incidence in plants. For that reason, researchers have used artificial inoculations or seed-treatment and foliar insecticides to obtain “healthy” checks to determine yield loss (17; 24; 30). While those methods have merit, they are costly or require much effort. For example, to artificially inoculate some plots with viruliferous aphids necessitates maintaining aphid colonies feeding on BYDV-infected plants and controlling the spread of aphids from inoculated plots to non-inoculated ones. Additionally, high levels of control of aphid transmission using seed-treatment and foliar chemicals can be difficult to achieve. In Kansas, treatment of seeds with insecticides results in about 50% control of BYD in some years (5; 6) but can also give no significant control in other years (Bockus, *unpublished*; 7). That method alone would not allow accurate assessment of the impact of BYD on grain yields in Kansas. To achieve “healthy” check plots in that state can require seed-treatment insecticides coupled with up to nine applications of foliar insecticides (7). Such elaborate methods to achieve “healthy” checks are often not feasible for breeding and disease-evaluation nurseries. The methods used here are more conducive to recurrent selection and BYD phenotyping experiments.

Using visual assessment of BYD incidence (symptoms) has proven to be useful in Kansas to help produce cultivars with improved levels of resistance to this important disease. Using this technique, the winter wheat cultivar Everest was developed and released in 2009 (12). Because

of selection for reduced BYD symptom expression, Everest is rated a 4 on the KSU Extension 1 to 9 scale, where 1 is highly resistant and 9 is highly susceptible (7). That moderate level of resistance was a major factor in the rapid grower adoption of Everest and it is now the number one cultivar grown in Kansas. Using the equations of the models generated here, Everest is calculated to have an average of 7.8% yield loss during the 4 years it was included in these experiments (2010-2013). During those same 4 years, the most susceptible cultivar had an average of 28.5% loss. Therefore, the resistance in Everest is estimated to have reduced yield loss from BYD by about 73% relative to more susceptible cultivars. In conclusion, the linear models developed here have utility in Kansas to: 1) quantify potential BYD yield losses in Kansas; 2) help develop cultivars with resistance to BYD; and 3) quantify the impact that improved levels of resistance has on yield loss from BYD.

**Table 1.** Winter wheat entries in seven barley yellow dwarf phenotyping experiments

Number	Year						
	2005	2006	2008	2010	2011	2012	2013
1	2137	2137	2137	2137	2137	282	Arrow
2	2163	2145	Abilene	Arkan	2145	625	Art
3	Akron	2174	Arkan	Armour	Above	894	Billings
4	Arkan	Above	Betty	Art	Arkan	947	Brawl CL
5	Auburn	Arkan	BondCL	Aspen	Art	2029	Byrd
6	Avalanche	Avalanche	Coker 9663	Betty	Bill Brown	2072	Centerfield
7	B-02	B-04	Danby	Bill	CJ	2089	Clara CL
8	B-03	B-06	Dominator	Coker 9663	Coker 9663	2100	Coker 9553
9	B-04	Betty	Duster	Danby	Cutter	2101	Duster
10	B-05	Coker 9663	Endurance	Dominator	Deliver	2110	Endurance
11	B-06	Cutter	Fuller	Duster	Endurance	2121	Everest
12	B-07	Dominator	Heyne	Endurance	Everest	2124	Garrison
13	B-08	Dumas	Jagger	Everest	Hawken	2126	Greer
14	B-09	Heyne	5391	Fuller	Hitch	2127	Jackpot
15	B-10	Intrada	5392	Hatcher	Ike	2129	Karl 92
16	B-11	Jagalene	5393	Hawken	Karl 92	2130	McGill
17	B-12	Jagger	5405	Heyne	Larned	2134	MFA 2018
18	Betty	Kaskaskia	5406	Hitch	Millenium	2139	MFA 2525
19	Coker 9474	KS010525-1-1	5407	Jagger	NuFrontier	2141	P 25R30
20	Coker 9663	KS010525-1-3	Karl 92	Karl 92	NuHills	2149	P 25R39
21	Custer	Lakin	Larned	Keota	Overland	Art	P 25R40
22	Dumas	Larned	McCormick	Larned	Overley	Coker 9663	P96134A3-2-2
23	Intrada	McCormick	MFA 2020	Neosho	P 25R47	Everest	Ripper
24	Jagalene	Millenium	Neosho	OK Bullet	P96134A3-2-2	Karl 92	Robidoux
25	Jagger	NuFrontier	Newton	Overland	Pat	-	Ruby Lee
26	Kaskaskia	NuHills	NuFrontier	Overley	Roane	-	SY Gold
27	Larned	NuPlains	OK Bullet	P 25R47	Sabbe	-	SY Wolf
28	McCormick	Overley	Overley	P96134A3-2-2	Shocker	-	T153
29	Millenium	P96134A3-2-2	P96134A3-2-2	Post Rock	Smokey Hill	-	T154
30	NuHills	Platte	Pioneer 25R47	Protection	Snowmass	-	T158



31	NuPlains	Prairie Red	Platte	Ripper	Sturdy 2K	-	T163
32	Overley	Prowers 99	Post Rock	Roane	TAM 110	-	TAM 110
33	P961341A3-2-2	Roane	Protection	RonL	TAM 111	-	TAM 113
34	Prairie Red	Stanton	Roane	Santa Fe	TAM 304	-	Thunder CL
35	Red Chief	T81	RonL	Shocker	Tiger	-	WB Cedar
36	Roane	TAM 107	Santa Fe	Smokey Hill	Trego	-	WB Stout
37	Stanton	Tomahawk	Shocker	Spartan	-	-	-
38	T81	Truman	Stanton	Stanton	-	-	-
39	Tomahawk	U4808-1-1-17-8	Sturdy 2K	T-81	-	-	-
40	USG 3209	U4808-1-1-25-6	TAM 112	TAM 110	-	-	-
41	-	U4808-4-3-19-3	Tarkio	TAM 111	-	-	-
42	-	U4808-4-3-20-5-7	Truman	TAM 112	-	-	-
43	-	-	-	TAM 203	-	-	-
44	-	-	-	TAM 304	-	-	-
45	-	-	-	Tarkio	-	-	-
46	-	-	-	Truman	-	-	-
47	-	-	-	Wesley	-	-	-
48	-	-	-	Winterhawk	-	-	-

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**Table 2:** Progression of barley yellow dwarf incidence in moderately resistant and susceptible cultivars during 2012<sup>a</sup>

<b>Cultivar<sup>b</sup></b>	<b>April 13</b>	<b>April 20</b>	<b>April 30</b>	<b>May 9</b>	<b>Mean</b>
Everest	13.4 <sup>c</sup>	22.2	28.8	22.6	21.8
Art	40.8	51.2	65.0	55.2	53.1
LSD ( $P=0.05$ ) <sup>d</sup>	10.7	12.9	12.2	11.1	9.9

<sup>a</sup>Visual assessment of the percentage tillers in a plot showing symptoms.

<sup>b</sup>Everest is moderately resistant and Art is susceptible.

<sup>c</sup>Each value is the average of four replications

<sup>d</sup>Analysis of variance followed by Fisher's protected least significant difference at  $P = 0.05$ .

**Table 3.** Linear regression models for seven years of Barley yellow dwarf incidence regressed against grain yield and for a data set combined over all seven years

<b>Year</b>	<b>N<sup>a</sup></b>	<b>P-value<sup>b</sup></b>	<b>R<sup>2</sup><sup>c</sup></b>	<b>Equation<sup>d</sup></b>	<b>Yield loss (%)<sup>e</sup></b>
2005	40	< 0.0001	0.4390	Y = -4.11X + 195.6	85.7
2006	42	< 0.0001	0.3676	Y = -3.17X + 279.4	55.2
2008	42	< 0.0001	0.4788	Y = -2.65X + 187.9	63.2
2010	48	< 0.0001	0.3748	Y = -1.61X + 266.2	40.4
2011	32	0.0120	0.1924	Y = -0.939x + 312.6	25.0
2012	24	0.0134	0.2473	Y = -2.18X + 255.6	45.2
2013	36	0.0023	0.2188	Y = -1.38X + 119.4	33.2
Combined	264	<0.0001	0.2891	Y = -0.659X + 134.9	48.9

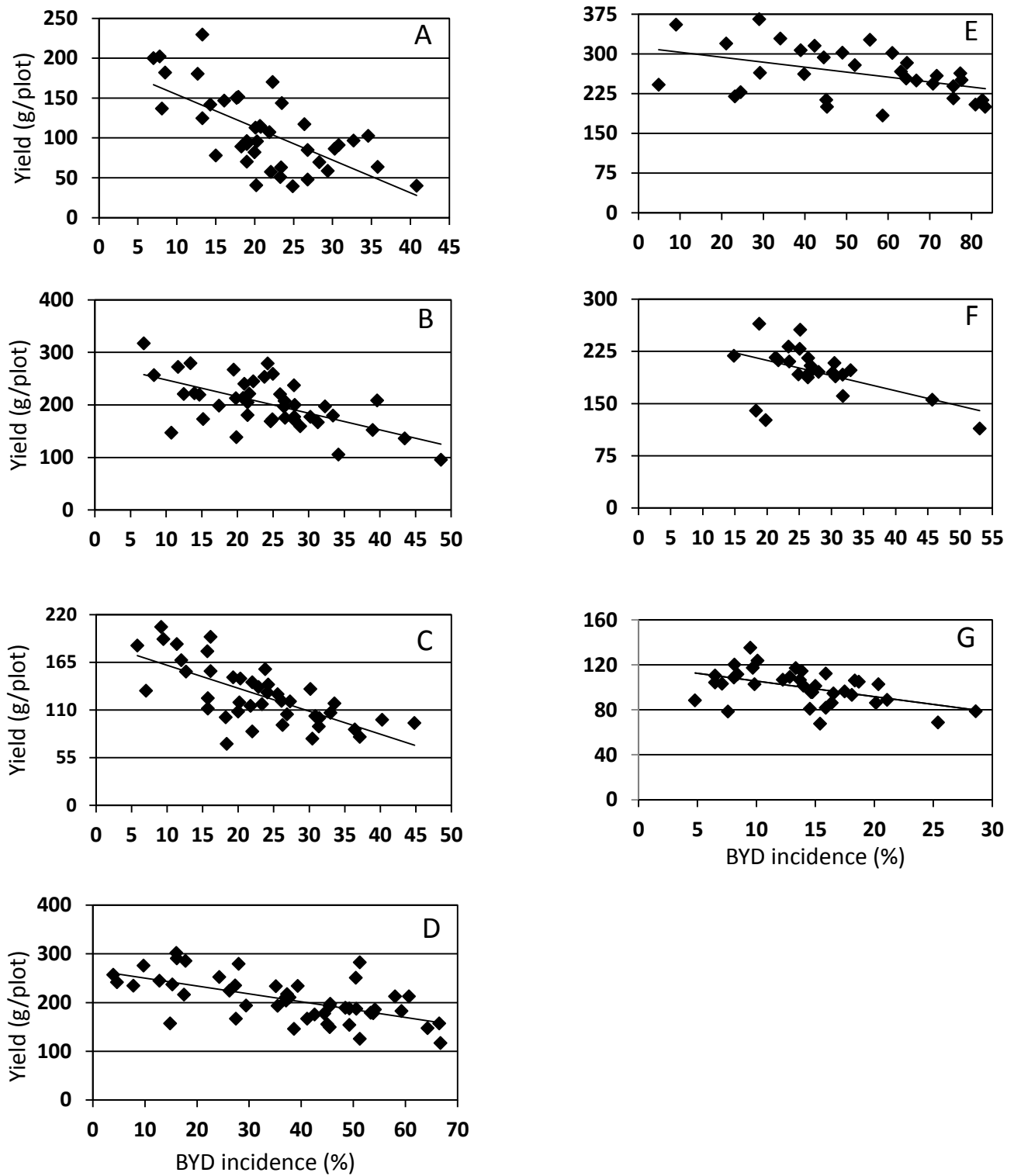
<sup>a</sup>Number of cultivars included in each field experiment.

<sup>b</sup>Significance of the slope parameter for the equation.

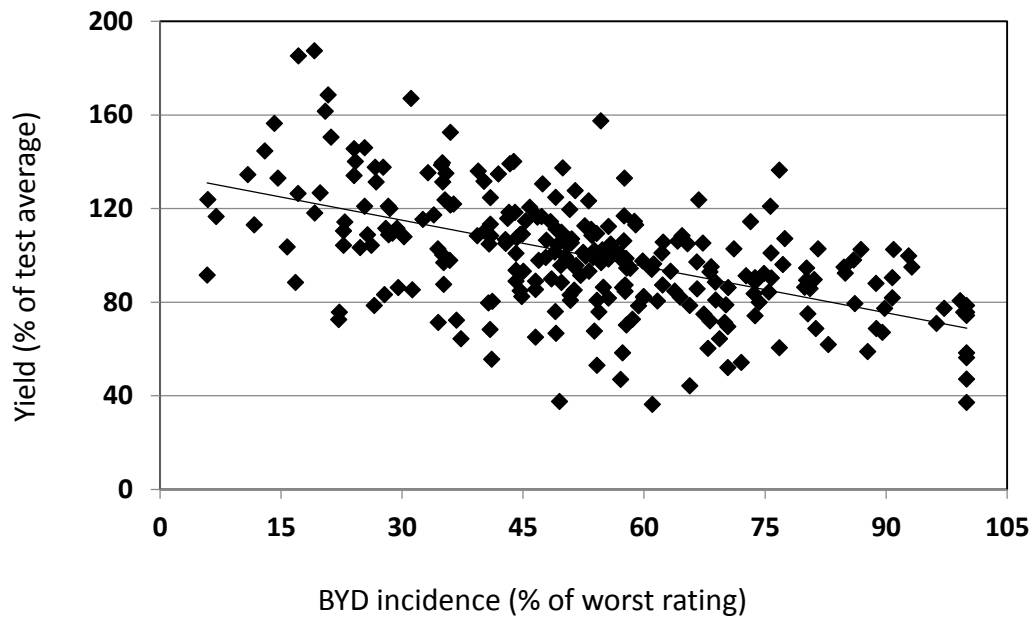
<sup>c</sup>Relationship between incidence of barley yellow dwarf and grain yield.

<sup>d</sup>Equation describing the trend line for incidence of barley yellow dwarf regressed against grain yield.

<sup>e</sup>Calculated from the incidence of the cultivar with the highest incidence that year compared with a hypothetical cultivar that had zero incidence.



**Fig. 1.** Incidence of Barley yellow dwarf regressed against grain yield for 2005 (A), 2006 (B), 2008 (C), 2010 (D), 2011 (E), 2012 (F), and 2013 (G).



**Fig. 2.** Relative incidence of Barley yellow dwarf regressed against relative grain yield for the combined data set for 2005, 2006, 2008, 2010, 2011, 2012, and 2013.

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