Impact of Temperatures on Hessian Fly (Diptera: Cecidomyiidae) Resistance in Selected Wheat Cultivars (Poales: Poaceae) in the Great Plains Region

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ABSTRACT Changes in temperature can result in fundamental changes in plant physiology. This study investigated the impact of different temperatures from 14 to 26°C on the resistance or susceptibility to the Hessian fly, *Mayetiola destructor* (Say), of selected wheat cultivars that are either currently popular in the Great Plains area or soon to be released to this region. We found that many wheat cultivars including 'Bill Brown,' 'Byrd,' 'Endurance,' 'Fuller,' 'GA-031257-10LE34,' and 'KS09H19-2-3' were susceptible to Hessian fly infestation at ≥20°C, but became resistant at a certain lower temperature, depending on different cultivars. These cultivars were classified as Hessian fly susceptible according to the traditional standards, and their impact on Hessian fly management needs to be reevaluated. However, many wheat cultivars that were resistant at ≤20°C became destabilized at a certain higher temperature. Phenotypic variations among the resistant cultivars at different temperatures were also observed, suggesting potential different resistance mechanisms. Studies on the genetic and molecular mechanisms associated with resistance at different temperatures are needed, which may lead to improved wheat cultivars with more durable resistance to Hessian fly infestation.

KEY WORDS Hessian fly, *Mayetiola destructor*, plant resistance, temperature effect, wheat

The Hessian fly, *Mayetiola destructor* (Say), is a serious pest of wheat in the United States and worldwide (Hatchett et al. 1987, Buntin 1999, Pauly 2002, Stuart et al. 2012). Since its introduction to the United States during the American Revolution ≈1,779, major Hessian fly outbreaks have been recorded, and localized serious damage due to this pest occurs every year. In recent years, heavily infested fields have occurred more frequently and on a larger scale, especially in the southern part of the United States, including Oklahoma, Texas, Georgia, and Louisiana (Colyer et al. 1989, Royer 2005, Watson 2005, Comis 2007, Knutson and Swart 2007, Smith 2007, Huang et al. 2011). This recent increase in Hessian fly incidence may be a result of climate change or the widely adopted no-till cultivation practice.

Major control measures for the Hessian fly include 1) late planting (Best Pest Management Planting Date) to avoid infestation from the fall generation; 2) timely destruction of volunteer wheat; 3) seed treatment with systemic pesticides; and 4) deployment of resistant wheat cultivars (Buntin and Bruckner 1990, Zelarayan et al. 1991, Buntin 1992, Buntin et al. 1992). Each control measure has its limitations. Late planting can only be adopted in cooler wheat growing regions because Hessian fly emergence can occur in late fall in warmer regions. In addition, many farmers plant wheat early for cattle grazing. Destruction of volunteer wheat is hard to coordinate within large wheat growing areas. The effect of seed treatment lasts only for 2–4 wk. Chemical application beyond seed treatment is generally not effective because of the nature of Hessian fly damage. Hessian fly larvae are hard to see because they are very small and live inside wheat plants. Once the damage is visible, it is too late to apply chemical pesticides because the damage is irreversible (Byers and Gallun 1971). Deployment of resistant cultivars is highly effective and cost efficient. However, resistance conferred by specific resistance genes is usually short-lived, lasting for only 6–8 yr (Gould 1998).

To date, 34 Hessian fly resistance genes have been identified and are named as *H1–H34* (Li et al. 2013). All known resistance genes are inherited as major dominant traits except *h4,* which is recessive, *H7H8,* which are dominant but must be inherited together to be effective (Amri et al. 1990), and *H34,* which is a newly identified gene that has not yet been well characterized (Li et al. 2013). All resistance genes are antibiotic to Hessian fly larvae, so that larvae die within plants without developing into second-instar larvae (Stuart et al. 2012). Wheat genes with resis-

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tance to Hessian fly are temperature-sensitive. Resistance is lost when temperature is above a certain degree depending on different resistance genes (Sosa and Foster 1976, Sosa 1979, Tyler and Hatchett 1983, Ratanatham and Gallun 1986). Among the known resistance genes, *H18* is the most sensitive, and it loses resistance when air temperature is >18°C (Cambron et al. 1996). Other known resistance genes are also temperature-sensitive, but the resistance remains as long as temperature is <22°C (Buntin et al. 1990). While screening wheat breeding lines for Hessian fly resistance, we observed that some lines were highly sensitive to changes in greenhouse temperatures, and often yield very inconsistent resistance results to Hessian fly from year to year. The objectives of this study were to determine the ranges of temperatures under which selected elite wheat cultivars in the Great Plains lose resistance; and to determine the duration under these temperatures to lose or gain resistance. We observed that many wheat cultivars that were categorized as susceptible previously are actually resistant under lower temperatures.

Materials and Methods

Hessian Fly. The Great Plains (GP) Hessian fly biotype was used in this study. Biotype GP was derived from a colony collected from Ellis County, KS, in 1988 (Gagne and Hatchett 1989). Since then, the insects have been maintained on susceptible wheat seedlings ('Newton' or 'Karl 92') in the greenhouse at 20°C. Biotype GP is avirulent to all known major Hessian fly resistance genes (Ratcliffe et al. 1994).

Wheat Cultivars. Each year scientists at the Kansas State Experimental Station, Kansas State University, Manhattan, KS, assemble a set of recently released or soon-to-be-released wheat cultivars from public and private breeders in the Great Plains area for testing resistance to various diseases and insects. Fifty-seven wheat cultivars from the 2013 set were used to study the impact of different temperatures on wheat resistance to Hessian fly. In addition to the 57 wheat cultivars, cultivars Newton and Karl92 were also used as susceptible controls in all experiments.

Infestation and Phenotyping. Approximately 12 wheat seeds were planted in each test. Wheat lines were planted in flats (54 by 36 by 8 cm) filled with PRO-MIX 'BX' potting mix (Hummert Inc., Earth City, MO). Each flat was divided into 24 sections with a divider and each cultivar was planted into each section. The flats were initially placed in a greenhouse to geminate. After 7 d, when the second leaf was beginning to emerge, the plants were infested with Hessian fly eggs by confining fly adults in a tent that enclosed the wheat seedlings. Female flies oviposit on the adaxial surface of plants in a free-choice manner, and eggs hatch in 3–5 d, depending on temperatures. When egg density reached ≈8 per plant, adult flies were removed from the tent, and the flats were transported into growth chambers programmed at various temperatures with a photoperiod of 14:10 (L:D) h. Neonate larvae migrate along the leaf, enter into a plant, and live between the first and second leaf under this condition.

Twenty-one days after infestation, plants were categorized and recorded as susceptible or resistant. During phenotyping, the first leaf sheath was separated from the second leaf sheath, and the dissected plant was examined under a microscope. Dead larvae (reddish and skinny) can be easily distinguished from live larvae (white and fat) by differences in color and size. Plants were considered resistant if they contained dead larvae and had grown normally. Plants were considered susceptible if they contained live larvae and were stunted. Plants with no dead or live larvae were categorized as escapes.

Temperature Duration Test. For cultivars requiring temperatures <20°C to exhibit Hessian fly resistant trait, we determined the duration of lower temperatures necessary to exhibit resistance. Wheat seedlings were infested as stated previously, seedlings with eggs were initially cultured at 20°C for 3 d, and the temperature was adjusted to 14°C before egg hatching. Egg hatch and larval migration were monitored on an hourly basis to determine the time when larvae reached the feeding site. When an average of four larvae reached the feeding site on a plant, the time was recorded as the initial attack time (Time 0). Seedlings were continuously cultivated under 14°C for 12, 24, 48, 72, and 120 h, and then transferred to a growth chamber at 20°C until scoring.

Statistical Analyses. Data were fitted to a logistic regression model using the numbers of resistant and susceptible plants as the response variable and temperature and wheat cultivar as the explanatory variables (factors). A deviance test with chi-square distribution was used to obtain P values. For each cultivar tested, a logistic model with deviance test was also used to assess whether the percentage of resistant plants differed significantly across different temperatures. After Bonferroni (1935) correction, a test was considered significant at the 0.05 level if the $P < 0.0028$ for cultivars tested at lower temperatures; and if the $P < 0.0011$ for cultivars tested at higher temperatures.

Results

Phenotypes of Selected Wheat Cultivars at 20°C. Cultivars were first tested for Hessian fly resistance at 20°C, the temperature at which routine screening was carried out. Of the 57 cultivars, 41 had ≥90% resistance (Table 1). 'Everest' showed 77% resistance, cultivars including 'Hatcher,' 'GA-031257-10LE34,' 'WB-Redhawk,' 'KS09H19-2-3,' 'Bill Brown,' and 'Thunder CL' had 10–40% resistance, and cultivars including 'Danby,' 'Brawl CL Plus,' 'Byrd,' 'Denali,' 'Endurance,' 'Fuller', 'GA-045710-10E46,' 'LCSMint,' and 'Santa Fe' had <10% resistance. The susceptible control cultivars Newton and Karl92 had 0% resistance.

Some Cultivars Susceptible at ≥20°C Become Resistant at Lower Temperatures. Cultivars with <90% resistant plants at 20°C were retested at 14°C (range of 14–16°C), 16°C (16–18°C), and 18°C (18–20°C). As shown in Table 2, the impact of lower temperatures

Table 1. Resistance or susceptibility of selected wheat cultivars to Hessian fly from the United States Great Plains at 20–22°C

Cultivar	Replicate 1			Replicate 2			Replicate 3			Total			SD
	R	S	%R	R	S	%R	R	S	%R	R	S	%R	
1863	11	0	100	8	0	100	10	0	100	29	0	100	0
2135	11	0	100	11	0	100	12	0	100	34	0	100	0
2166	10	0	100	9	0	100	10	0	100	29	0	100	0
2525	11	0	100	10	0	100	8	0	100	29	0	100	0
25R30	10	0	100	9	0	100	8	0	100	27	0	100	0
25R32	11	0	100	7	0	100	11	0	100	29	0	100	0
25R39	10	0	100	10	0	100	9	0	100	29	0	100	0
Armour	7	0	100	11	0	100	8	0	100	26	0	100	0
Art	12	0	100	11	0	100	11	0	100	34	0	100	0
Billings	11	0	100	11	0	100	12	0	100	34	0	100	0
Centerfield	10	0	100	8	0	100	9	0	100	27	0	100	0
CJ	11	0	100	9	0	100	12	0	100	32	0	100	0
Clara CL (W)	9	0	100	9	0	100	11	0	100	29	0	100	0
Deliver	7	0	100	9	0	100	11	0	100	27	0	100	0
Duster	10	0	100	11	0	100	8	0	100	29	0	100	0
GA-031086-10E26	12	0	100	10	0	100	11	0	100	33	0	100	0
Gallagher	12	0	100	9	0	100	11	0	100	32	0	100	0
Garrison	7	0	100	11	0	100	8	0	100	26	0	100	0
Greer	10	0	100	12	0	100	8	0	100	30	0	100	0
Jackpot	9	0	100	11	0	100	10	0	100	30	0	100	0
LCH08-109	11	0	100	8	0	100	10	0	100	29	0	100	0
LCH08-80	12	0	100	9	0	100	11	0	100	32	0	100	0
LCH09-19	10	0	100	11	0	100	9	0	100	30	0	100	0
LCS08-12	10	0	100	9	0	100	11	0	100	30	0	100	0
OK09915C	11	0	100	9	0	100	10	0	100	30	0	100	0
PostRock	8	0	100	11	0	100	10	0	100	29	0	100	0
SY Southwind	9	0	100	11	0	100	8	0	100	28	0	100	0
T153	8	0	100	8	0	100	10	0	100	26	0	100	0
T154	9	0	100	9	0	100	11	0	100	29	0	100	0
T158	10	0	100	8	0	100	10	0	100	28	0	100	0
TAM 111	7	0	100	12	0	100	9	0	100	28	0	100	0
TAM 113	11	0	100	9	0	100	10	0	100	30	0	100	0
WB4458	9	0	100	11	0	100	10	0	100	30	0	100	0
WB-Cedar	10	0	100	10	0	100	11	0	100	31	0	100	0
WB-Grainfield	10	0	100	9	0	100	12	0	100	31	0	100	0
Winterhawk	11	0	100	11	0	100	9	0	100	31	0	100	0
LCH09-43	10	1	91	10	0	100	11	0	100	31	1	97	5
SY Gold	8	0	100	9	1	90	11	0	100	28	1	97	8
SY Wolf	6	1	86	9	0	100	11	1	92	26	2	93	7
TAM 304	11	0	100	7	2	78	10	0	100	28	2	93	13
TAM 401	8	2	80	11	0	100	10	1	91	29	3	91	10
Everest	*9*	*1*	*90*	*7*	*3*	*70*	*8*	*3*	*73*	*24*	*7*	*77*	*11*
Hatcher	*6*	*3*	*67*	*2*	*8*	*20*	*3*	*8*	*27*	*11*	*19*	*37*	*25*
GA-031257-10LE34	*5*	*5*	*50*	*3*	*8*	*27*	*3*	*7*	*30*	*11*	*20*	*35*	*13*
WB-Redhawk	*5*	*4*	*56*	*2*	*8*	*20*	*3*	*8*	*27*	*10*	*20*	*33*	*19*
KS09H19-2-3	*4*	*6*	*40*	*1*	*4*	*20*	*3*	*8*	*27*	*8*	*18*	*30*	*10*
Bill Brown	*0*	*9*	*0*	*3*	*7*	*30*	*0*	*11*	*0*	*3*	*27*	*10*	*17*
Thunder CL	*0*	*8*	*0*	*2*	*8*	*20*	*1*	*10*	*9*	*3*	*26*	*10*	*10*
Danby (W)	*1*	*8*	*11*	*0*	*9*	*0*	*0*	*12*	*0*	*1*	*29*	*3*	*6*
Brawl CL Plus	*0*	*10*	*0*	*0*	*8*	*0*	*0*	*9*	*0*	*0*	*27*	*0*	*0*
Byrd	*0*	*8*	*0*	*0*	*8*	*0*	*0*	*12*	*0*	*0*	*28*	*0*	*0*
Denali (Kansas)	*0*	*11*	*0*	*0*	*7*	*0*	*0*	*11*	*0*	*0*	*29*	*0*	*0*
Endurance	*0*	*5*	*0*	*0*	*9*	*0*	*0*	*11*	*0*	*0*	*25*	*0*	*0*
Fuller	*0*	*9*	*0*	*0*	*7*	*0*	*0*	*10*	*0*	*0*	*26*	*0*	*0*
GA-045710-10E46	*0*	*11*	*0*	*0*	*7*	*0*	*0*	*10*	*0*	*0*	*28*	*0*	*0*
LCSMint	*0*	*8*	*0*	*0*	*7*	*0*	*0*	*10*	*0*	*0*	*25*	*0*	*0*
Santa Fe	*0*	*7*	*0*	*0*	*11*	*0*	*0*	*10*	*0*	*0*	*28*	*0*	*0*
Newton	*0*	*13*	*0*	*0*	*9*	*0*	*0*	*11*	*0*	*0*	*33*	*0*	*0*
Karl92	*0*	*9*	*0*	*0*	*11*	*0*	*0*	*10*	*0*	*0*	*30*	*0*	*0*

R, number of resistant plants; S, number of susceptible plants; T, total number of plants; %R, percent resistant plants (mean); SD, standard deviation. Cultivars in italic were with <80% resistant plants.

was very different on the Hessian fly resistance of different wheat cultivars. The cultivar Everest, which showed a moderate 77% resistance, remained moderately resistant at all temperatures with a slight increase in resistance at the 14–16°C range. Similarly, WB-Redhawk, which had a low-level 33% resistance, remained low, but with a slight increase at the 14–16°C range. Lower temperatures have no significant impact on resistance in Everest ($P = 0.634$) or WB-Redhawk ($P = 0.432$).

Conversely, lower temperatures had a dramatic impact on resistance of several cultivars. Hatcher, GA-

Table 2. Wheat cultivars susceptible at 20–22°C became resistant to Hessian fly infestation at lower temperatures

Cultivar	14–16°C		16–18°C		18–20°C		20–22°C	
	T	%R/SD	T	%R/SD	T	%R/SD	T	%R/SD
Everest	33	88/7	33	81/8	35	77/1	31	77/11
Hatcher	34	97/2	33	76/4	33	85/16	*30*	*37/25*
GA-031257-10LE34	33	91/9	33	85/14	34	65/14	*31*	*35/13*
Bill Brown	34	97/5	29	97/6	34	94/10	*30*	*10/17*
Thunder CL	34	94/5	32	72/11	31	81/2	*29*	*10/10*
Fuller	32	94/10	30	97/6	34	71/2	*26*	*0/0*
Endurance	35	94/10	31	97/10	32	94/1	*25*	*0/0*
Byrd	30	93/10	*32*	*6/10*	*31*	*3/6*	*28*	*0/0*
Denali (Kansas)	29	52/14	*32*	*6/9*	*30*	*0/0*	*29*	*0/0*
KS09H19-2-3	31	87/2	*33*	*6/10*	*31*	*10/16*	*26*	*30/10*
Brawl CL Plus	*34*	*18/5*	*32*	*3/5*	*30*	*0/0*	*27*	*0/0*
GA-045710-10E46	*30*	*40/9*	*33*	*9/9*	*33*	*18/14*	*28*	*0/0*
Danby (W)	*35*	*43/9*	*32*	*6/10*	*23*	*0/0*	*30*	*3/6*
LCSMint	*29*	*10/10*	*31*	*17/10*	*33*	*15/16*	*25*	*0/0*
Santa Fe	*33*	*36/4*	*32*	*31/11*	*33*	*18/10*	*28*	*0/0*
WB-Redhawk	*35*	*43/4*	*35*	*26/11*	*39*	*28/2*	*30*	*33/19*
Newton	*61*	*15/4*	*59*	*0/0*	*56*	*0/0*	*33*	*0/0*
Karl92	*56*	*14/6*	*58*	*0/0*	*61*	*0/0*	*30*	*0/0*

T, total number of plants tested; %R, percent resistant plants (mean); SD, standard deviation. Cultivars with <50% resistant plants are in italic. Each treatment was repeated three times.

031257-10LE34, Bill Brown, Thunder confidence limits, Fuller, and Endurance became moderately to highly resistant (71–94%) beginning at 18–20°C. Byrd, Denali, and KS09H19-2-3 also became moderately to highly resistant (52–93%) at 14–16°C. All these differences in Hessian fly resistance were significant at the 0.05 level.

The impact of lower temperature on Brawl confidence limits Plus, 'GA-045710-10E46, Danby, LCSMint, and Santa Fe was less significant. These cultivars were susceptible at 20–22°C, and remained susceptible or had a very low level of resistance until the temperature reached 14–16°C, when resistance increased but did not exceed 50%. The susceptible control cultivars Newton and Karl92 remained susceptible until the temperature reached 14–16°C, when some (<20%) plants became resistant. Lower temperature had no significant impact on resistance in LCSMint ($P = 0.108$) and Brawl confidence limits Plus (0.004) to Hessian fly infestation. However, lower temperatures significantly impacted GA-045710-10E46, Danby, and Santa Fe at the 0.05 level, even though resistance did not exceed 50%.

Some Cultivars Resistant at ≤20–22°C Become Susceptible at Higher Temperatures. Wheat cultivars with ≥70% resistant plants (Table 1) were retested at 22–24°C, 24–26°C, and 26–28°C (Table 3). Everest remained moderately resistant at all temperatures. Resistance in 'TAM 401' became destabilized with only 56% resistance at 22–24°C, and essentially lost resistance at ≥24°C. Resistance in 'Clara confidence limits' became destabilized at 24–26°C with <50% resistant plants, and essentially lost resistance ≥26°C. 'LCH09-19,' 'GA-031086-10E26,' '25R30,' and 'Art', became susceptible, with resistance of <50%. 'SY Southwind,' 'SY Wolf,' 'SY Gold,' 'PostRock,' 'OK09915C,' 'Jackpot,' 'Greer,' '25R32,' and 'WB-Cedar' also became destabilized and lost resistance, with percentages of resistant plants decreasing to 50–80%. Higher temperatures significantly impacted resistance in wheat cultivars at the 0.05 level, with the exception of SY Wolf and WB-Cedar. Other cultivars retained high levels (>80%) of resistance at all tested temperatures.

Time Required for Phenotype Changes Under Selected Temperatures. Phenotype switching from susceptible to resistant occurred at different durations at 14–16°C for different cultivars (Table 4). Bill Brown and Hatcher needed only 12 h at 14–16°C to trigger resistance in ≥50% plants. It took 48 h for Thunder confidence limits, 72 h for Byrd and KS09H19-2-3, and 120 h for GA-031257-10LE34, Fuller, and Endurance to exhibit resistance in ≥50% plants under the same conditions.

Phenotypic Variations Among Infested Seedlings of Different Cultivars. For susceptible plants, Hessian fly feeding symptoms were rather typical (Fig. 1A and S). The elongation of the second leaf sheath was inhibited and the third leaf failed to grow out or extremely stunted if it grew out at all. Susceptible plants were all dead after larvae inside pupated (data not shown). Physical appearance varied for plants with resistance to Hessian fly infestation. Two of the most commonly observed resistant phenotypes are shown in Fig. 1. For type 1 (R1) plants, the elongation of the second leaf sheath was inhibited as observed in susceptible plants, but the third leaf grew out relatively normally 2 wk after Hessian fly infestation (Fig. 1A and R1 in the middle). Elongation of the third leaf sheath and growth of fourth leaf were normal in type 1 plants when the plants were continuously cultured in growth chambers (Fig. 1, R1 on the right). For type 2 plants (R2), elongation of the second leaf sheath and growth of third leaf were similar to uninfested control plants (Fig. 1, R2). Fly larvae in resistant plants were dead in resistant plants without apparent growth or development, whereas larvae in susceptible plants developed into second instars 2 wk after infestation (Fig. 1B).

Table 3. Wheat cultivars with Hessian fly resistance at 20–22°C lose resistance at higher temperatures

Cultivar	20–22°C		22–24°C		24–26°C		26–28°C	
	T	%R/SD	T	%R/SD	T	%R/SD	T	%R/SD
TAM 401	32	91/10	34	56/16	34	*23/3*	*36*	*16/8*
Clara CL (W)	29	100/0	33	82/9	33	*43/7*	*32*	*18/8*
LCH09-19	30	100/0	32	100/0	28	97/5	31	*10/8*
GA-031086-10E26	33	100/0	29	100/0	27	93/6	31	*15/15*
25R30	27	100/0	31	100/0	30	90/9	31	*27/15*
Art	34	100/0	31	97/6	29	83/4	28	*48/21*
SY Southwind	28	100/0	33	92/8	28	61/6	31	**50/13**
SY Wolf	28	93/7	33	88/3	34	77/25	30	**56/13**
SY Gold	29	97/6	33	85/4	33	82/2	32	**57/16**
PostRock	29	100/0	30	100/0	34	92/8	27	**59/8**
OK09915C	30	100/0	29	100/0	32	88/4	30	**60/6**
Jackpot	30	100/0	27	100/0	28	94/6	28	**62/11**
Greer	30	100/0	32	100/0	32	94/5	35	**63/3**
Everest	31	77/11	27	66/4	31	77/6	32	**72/10**
25R32	29	100/0	30	100/0	31	100/0	31	**74/10**
WB-Cedar	31	100/0	26	100/0	33	92/8	33	**79/3**
25R39	29	100/0	26	100/0	30	97/6	28	81/17
TAM 111	28	100/0	32	100/0	33	86/8	34	82/9
WB-Grainfield	31	100/0	27	100/0	30	97/4	30	94/5
Winterhawk	31	100/0	26	100/0	30	100/0	31	94/5
Armour	26	100/0	28	100/0	28	97/4	27	96/6
T158	28	100/0	32	100/0	27	100/0	31	97/6
TAM 304	30	93/13	27	100/0	26	97/5	29	97/6
WB4458	30	100/0	33	97/4	30	97/5	29	97/5
1863	29	100/0	27	100/0	25	100/0	30	97/5
CJ	32	100/0	24	100/0	26	100/0	28	97/5
Gallagher	32	100/0	26	100/0	29	100/0	29	97/5
LCH08–80	32	100/0	25	100/0	29	100/0	26	97/5
Billings	34	100/0	26	100/0	29	100/0	33	97/4
LCH08-109	29	100/0	31	100/0	28	100/0	26	100/0
LCH09-43	32	97/5	26	100/0	26	100/0	25	100/0
LCS08-12	30	100/0	32	100/0	32	100/0	32	100/0
T153	26	100/0	27	100/0	26	100/0	27	100/0
T154	29	100/0	30	100/0	29	100/0	30	100/0
2135	34	100/0	28	100/0	29	100/0	31	100/0
2166	29	100/0	29	100/0	31	100/0	28	100/0
2525	29	100/0	24	100/0	23	100/0	29	100/0
Centerfield	27	100/0	29	100/0	30	97/6	25	100/0
Deliver	27	100/0	27	100/0	34	97/5	31	100/0
Duster	29	100/0	29	100/0	29	100/0	28	100/0
Garrison	26	100/0	32	100/0	29	100/0	26	100/0
TAM 113	30	100/0	30	100/0	33	97/4	28	100/0
Newton	33	0/0	34	0/0	33	0/0	34	0/0
Karl92	30	0/0	34	0/0	37	0/0	37	0/0

T, total number of plants tested; %R, percent resistant plants (mean); SD, standard deviation.
Cultivars with <50% resistant plants are in italic, or 80% resistant plants are in bold. Each treatment was repeated three times.

Table 4. Times (h) exposed to 14–16°C for wheat cultivars to exhibit Hessian fly resistance at 20°C

Cultivar	12 h		24 h		48 h		72 h		120 h	
	T	%R/SD	T	%R/SD	T	%R/SD	T	%R/SD	T	%R/SD
Bill Brown	*32*	*50/9*	*32*	*97/5*	*32*	*97/5*	*29*	*93/6*	*34*	*97/5*
Hatcher	*34*	*71/6*	*34*	*76/5*	*31*	*68/2*	*32*	*94/10*	*34*	*97/2*
Thunder CL	26	27/16	30	27/2	*31*	*52/8*	*31*	*97/6*	*34*	*94/5*
Byrd	30	0/0	30	0/0	32	6/10	*32*	*75/11*	*30*	*93/10*
KS09H19-2-3	29	14/12	29	17/14	34	12/10	*35*	*86/6*	*31*	*87/2*
GA-031257-10LE34	33	30/5	31	39/7	32	34/7	36	36/5	*33*	*91/9*
Fuller	32	3/5	30	3/5	29	3/6	33	30/3	*32*	*94/10*
Endurance	30	3/6	32	3/5	32	6/10	29	31/6	*35*	*94/10*
Newton	56	0/0	64	5/5	58	10/9	61	10/4	61	15/4
Karl92	59	2/3	60	5/8	60	10/6	55	15/4	56	14/6

Seedlings infested with Hessian fly larvae were continuously cultivated at 14–16°C for 12, 24, 48, 72, and 120 h, and were then transferred to 20°C for phenotyping.
Italics indicate cultivars with >50% resistant plants under that duration exposed to 14–16°C. Each treatment was repeated three times.
T, total number of plants tested; %R, percent resistant plants (mean); SD, standard deviation.

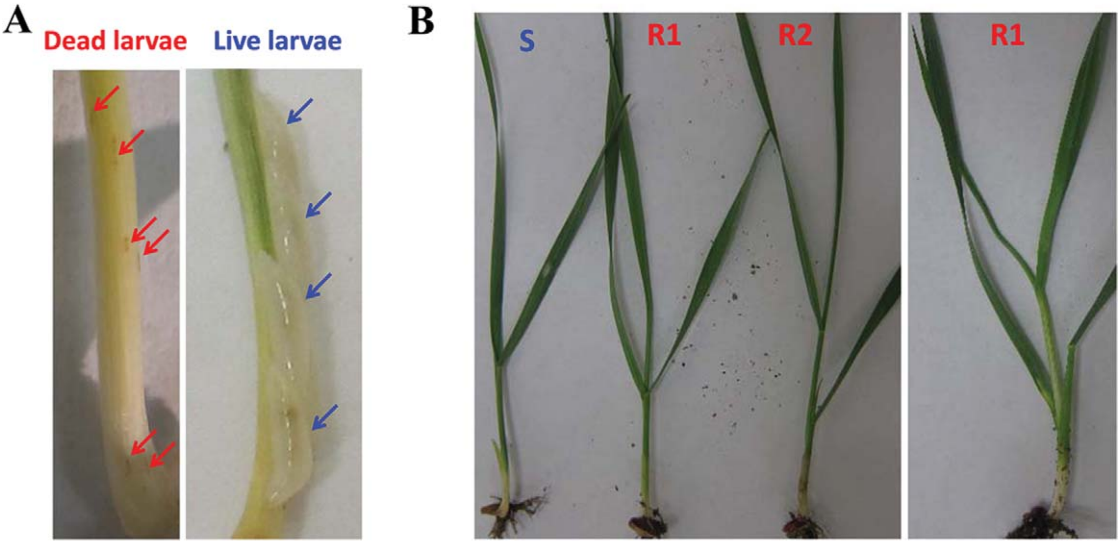

Fig. 1. Variations in phenotypes of susceptible and resistant wheat seedlings. (**A**) Skinny dead larvae (indicated by red arrows on the left side) in resistant plants, and fat live larvae (indicated by blue arrows on the right) in susceptible plants. (**B**) Plant appearance of susceptible and resistant plants after Hessian fly attack. The plant indicated by the letter S represents the appearance of a typical susceptible plant 2 wk after the initial Hessian fly attack. The elongation of the second leaf sheath is inhibited, and often the third leaf fails to grow out. The plant indicated by R1 in the middle represents the appearance of wheat seedlings of some resistant cultivars 2 wk after Hessian fly attack. The elongation of the second leaf sheath was inhibited, but the third leaf grows out. After another week, the plant indicated by R1 in the middle develops into a plant indicated by R1 on the right, with elongation of the third leaf sheath and growth of the fourth leaf. The plant indicated by R2 is a different type of appearance of resistant seedlings. The plant grows relatively normally with elongation of the second leaf sheath and growth of the third leaf.

Most cultivars tested with resistance to Hessian fly at low temperatures (<20°C) had the type 1 phenotype (Bill Brown, Denali, Fuller, GA-031257-10LE34, Hatcher, and Thunder confidence limits; Fig. 2). Exceptions were Byrd and Endurance, which exhibited the type 2 phenotype. In contrast, most wheat cultivars with Hessian fly resistance at ≥20°C exhibited the type 2 phenotype, but there were also exceptions (Fig. 2).

Discussion

The impact of temperature on various plant traits attracts a great deal of attention in both the public and the scientific community because of climate change. The impact of temperature on the effectiveness of wheat resistance to Hessian fly is particularly important for the management of this insect pest for several reasons. First, the loss of wheat resistance to Hessian

Type 1 Resistance Phenotype (R1)	Type 2 Resistance Phenotype (R2)		
Art	1863	Duster	SY Southwind
Bill Brown	2135	**Endurance**	SY Wolf
Clara CL	2166	GA-031086-10E26	T153
Denali	2525		T154
Everest	25R30	Garrison	T158
Fuller	25R32	Greer	TAM 111
GA-031257-10LE34	25R39	Jackpot	TAM 113
Gallagher	Armour	LCH08-80	TAM 304
Hatcher	Billings	LCH09-19	WB4458
LCH08-109	**Byrd**	LCH09-43	WB-Cedar
OK09915C	Centerfield	LCS08-12	WB-Grainfield
TAM 401	CJ	PostRock	Winterhawk
Thunder CL	Deliver	SY Gold	

Fig. 2. Wheat cultivars with the two types of phenotypic appearance of resistant plants after Hessian fly attack. The bold and shaded cultivars were resistant to Hessian fly infestation only under low (<20°C) temperatures.

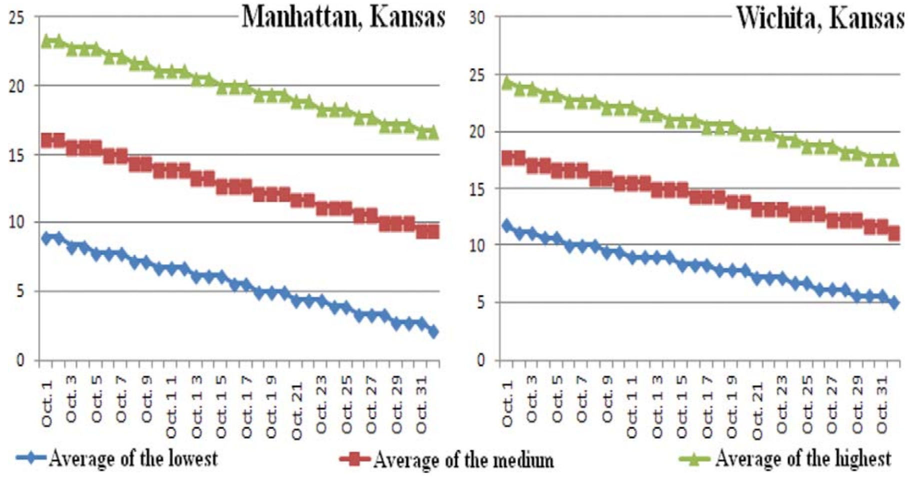

Fig. 3. The 2002–2012 temperatures in October in Manhattan and Wichita, KS, from the National Weather Service center of the National Oceanic and Atmospheric Administration (http://www.weatherpages.com/wxhistory.html).

fly at higher temperatures poses a threat to the overall strategy of using plant resistance for controlling this pest in a global warming scenario. Historically, plant resistance has played a crucial role in reducing wheat yield loss due to Hessian fly infestation (Buntin et al. 1992). Second, even a slight temperature change that results in a resistant plant becoming susceptible or vice versa presented a major challenge in breeding Hessian fly-resistant wheat and in basic research to understand the Hessian fly resistance mechanism. Greenhouse evaluation of breeding lines for fly resistance often yields inconsistent results from year to year, and these results also complicate efforts to map and clone Hessian fly resistance genes in wheat. Third, the existence of a wide range of wheat cultivars with resistance at lower temperatures was unknown, and the impact of this type of resistance on Hessian fly population dynamics remains to be evaluated.

Historically, the resistance strategy for controlling Hessian fly has overall been less successful in the southern United States than in the northern United States. One hypothesis for this phenomenon is that Hessian fly has more generations in the South, and therefore may overcome plant resistance more quickly. According to this study and previous reports (Buntin et al. 1990), the differences in temperatures during the wheat growing seasons between the South and the North could be another major factor. For example, the historical average temperatures in October in the Kansas City (MO) area is 9–19°C (medium average at the end of October–medium average at the beginning of October), whereas that in the Dallas (TX) area is 14–25°C, according to the National Weather Service center of the National Oceanic and Atmospheric Administration (http://www.weatherpages.com/wxhistory.html). These 5–6°C differences mean that many wheat cultivars that are resistant to Hessian fly infestation in Kansas may become susceptible or less resistant in Texas. The impact of temperature on the effectiveness of controlling Hessian fly damage using resistant wheat cultivars needs to be fully evaluated from region to region.

Many wheat cultivars popular in the Great Plains area, including Bill Brown, Byrd, Fuller, and Endurance, were classified as susceptible to the Hessian fly by traditional standards. Yet these cultivars become fully resistant to Hessian fly infestation at temperatures below 14–20°C, depending on the specific cultivar (Table 2). Historically, average medium temperatures are 10–15°C in Kansas (Fig. 3), which is in the right range for these wheat cultivars to exhibit Hessian fly resistance. The existence of cultivars resistant to Hessian fly <20°C in the Great Plains may have played an unrecognized role in controlling this pest. The potential benefit of wheat cultivars with resistance at lower temperatures needs to be investigated.

Wheat resistance to Hessian fly is currently thought to be conferred by major dominant resistance genes with antibiosis (Stuart et al. 2012). These resistance genes interact with Hessian fly avirulence genes in a typical gene-for-gene fashion. The genetic mechanism of wheat resistance to Hessian fly infestation at low temperatures is not yet known. Phenotypically, most cultivars with resistance at the lower temperature ranges appear to have a greater defense cost in terms of wheat growth inhibition (Figs. 1 and 2), which could indicate a different resistance mechanism. It remains to be determined if single major genes or multiple minor genes are responsible for Hessian fly resistance at lower temperatures. Further investigation into the genetic and molecular mechanisms involved in low temperature-associated resistance is needed, and could lead to new ways to generate more durable resistance of wheat cultivars for Hessian fly management.

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References Cited

Amri, A., T. S. Cox, J. H. Hatchett, and B. S. Gill. 1990. Complementary action of genes for Hessian fly resistance in the wheat cultivar 'Seneca'. J. Hered. 81: 224–227.

Bonferroni, C. E. 1935. Il calcolo delle assicurazioni su gruppi di teste, pp. 13–60. *In* Studi in Onore del Professore Salvatore Ortu Carboni. Rome: Italy.

Buntin, G. D. 1992. Assessment of a microtube injection system for applying systemic insecticides at planting for Hessian fly control in winter wheat. Crop Prot. 11: 366–370.

Buntin, G. D. 1999. Hessian fly (Diptera: Cecidomyiidae) injury and loss of winter wheat grain yield and quality. J. Econ. Entomol. 92: 1190–1197.

Buntin, G. D., and P. L. Bruckner. 1990. Effect of planting date on Hessian fly infestation and production of triticale. Appl. Agric. Res. 5: 82–88.

Buntin, G. D., P. L. Bruckner, J. W. Johnson, J. E. Foster. 1990. Effectiveness of selected genes for Hessian fly resistance in wheat. J. Agric. Entomol. 7: 283–291.

Buntin, G. D., S. L. Ott, and J. W. Johnson. 1992. Integration of plant resistance, insecticides, and planting date for management of the Hessian fly (Diptera: Cecidomyiidae) in winter wheat. J. Econ. Entomol. 85: 530–538.

Byers, R. A., and R. L. Gallun. 1971. Ability of the Hessian fly to stunt winter wheat. I. Effect of larval feeding on elongation of leaves. J. Econ. Entomol. 65: 955–958.

Cambron, S. E., H. W. Ohm, R. H. Ratcliffe, and F. L. Patterson. 1996. A second gene for resistance to Hessian fly in Iumillo durum wheat. Crop Sci. 36: 1099–1101.

Comis, D. 2007. Virulent Hessian flies renew attack on U.S. wheat. United States Department of Agriculture Agricultural Research – News and Events (January 24). (http://www.ars.usda.gov/is/pr/2007/070124.htm).

Colyer, P. D., P. R. Vernon, and T. Riley. 1989. First report of Hessian fly (Diptera: Cecidomyiidae) on winter wheat in Louisiana. J. Agric. Entomol. 7: 147–148.

Gagne, R. J., and J. H. Hatchett. 1989. Instars of the Hessian fly (Diptera: Cecidomyiidae). Ann. Entomol. Soc. Am. 82: 73–79.

Gould, F. 1998. Sustainability of transgenic insecticidal cultivars: integrating pest genetics and ecology. Annu. Rev. Entomol. 43: 701–726.

Hatchett, J. H., K. J. Starks, and J. A. Webster. 1987. Insect and mite pests of wheat, pp. 625–675. Wheat and Wheat Improvement. Agronomy Monograph No. 13. American Society of Agronomy, Madison, WI.

Huang, F., S. Harrison, B. R. Leonard, M. Ghimire, and P. P. Price. 2011. Hessian fly, an emerging wheat pest in Louisiana. Louisiana Agriculture Mag. (http://text.lsuagcenter.com/en/communications/publications/agmag/Archive/2011/Winter/Hessian-Fly.htm).

Knutson, A., and J. Swart. 2007. Hessian fly outbreaks showing up in many north Texas wheat fields. Southwest Farm Report. (http://southwestfarmpress.com/grains/041807-hessian-fly/).

Li, C., M. S. Chen, S. Chao, J. Yu, and G. Bai. 2013. Identification of a novel gene, H34, in wheat using recombinant inbred lines and single nucleotide polymorphism markers. Theor. Appl. Genet. (in press).

Pauly, P. J. 2002. Fighting the Hessian fly. Environ Hist. 7: 385–507.

Ratanatham, S., and R. L.Gallun. 1986. Resistance to Hessian fly (Diptera: Cecidomyiidae) in wheat as affected by temperature and larval density. Environ. Entomol. 15: 305–310.

Ratcliffe, R. H., G. G. Safranski, F. L. Patterson, H.W. Ohm, and P. L. Taylor. 1994. Biotype status of Hessian fly (Diptera: Cecidomyiidae) populations from the eastern United States and their response to 14 Hessian fly resistance genes. J. Econ. Entomol. 87: 1113–1121.

Royer, T. A. 2005. Hessian fly reported in Oklahoma wheat. (http://www.ento.okstate.edu/Pddl/2005/PDIA4-1.pdf).

Smith, R. 2007. Widespread outbreaks of Hessian fly reported in wheat. Southwest Farm Report. (http://southwestfarmpress.com/grains/051807-hessian-fly/).

Sosa, O., Jr. 1979. Hessian fly resistance of wheat as affected by temperature and duration of exposure. Environ. Entomol. 8: 280–281.

Sosa, O., Jr., and J. E. Foster. 1976. Temperature and the expression of resistance in wheat to the Hessian fly. Environ. Entomol. 5: 333–336.

Stuart, J. J., M. S. Chen, M. O. Harris, and R. H. Shukle. 2012. Gall midges (Hessian Flies) as plant pathogens. Annu. Rev. Phytopathol 50: 339–357.

Tyler, J. M., and J. H. Hatchett. 1983. Temperature influence on expression of resistance to Hessian fly (Diptera: Cecidomyiidae) in wheat derived from *Triticum taushii.* J. Econ. Entomol. 76: 323–326.

Watson, S. 2005. Hessian fly problems have been increasing in recent years in the Central Plains. Wheat Farmer/Row Crop Farmer 9: 4–5.

Zelarayan, E. L., G. D. Buntin, J. W. Johnson, P. L. Bruckner, and P. L. Raymer. 1991. Integrated management for Hessian fly in Triticale. J. Prod. Agric. 4: 629–633.

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