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A Simple Method for the Assessment of Fusarium Head Blight Resistance in Korean Wheat Seedlings Inoculated with *Fusarium graminearum*

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Fusarium head blight (FHB; scab) caused mainly by *Fusarium graminearum* is a devastating disease of wheat and barley around the world. FHB causes yield reductions and contamination of grain with trichothecene mycotoxins such as deoxynivalenol (DON) which are a major health concern for humans and animals. The objective of this research was to develop an easy seed or seedling inoculation assay, and to compare these assays with whole plant resistance of twenty-nine Korean winter wheat cultivars to FHB. The clip-dipping assay consists of cutting off the coleoptiles apex, dipping the coleoptiles apex in conidial suspension, covering in plastic bag for 3 days, and measuring the lengths of lesions 7 days after inoculation. There were significant cultivar differences after inoculation with *F. graminearum* in seedling relative to the controls. Correlation coefficients between the lesion lengths of clip-dipping inoculation and FHB Type II resistance from adult plants were significant ($r=0.45$; $P<0.05$). Results from two other seedling inoculation methods, spraying and pin-point inoculation, were not correlated with adult FHB resistance. Single linear correlation was not significant between seed germination assays (soaking and soak-dry) and FHB resistance (Type I and Type II), respectively. These results showed that clip-dipping inoculation method using *F. graminearum* may offer a real possibility of simple, rapid, and reliable for the early screening of FHB resistance in wheat.

Keywords : Fusarium head blight (FHB), seed germination assay, seedling inoculation assay, wheat

Fusarium head blight (FHB) is a major disease problem on the wheat and barley crops around the world. FHB can be associated with at least seventeen *Fusarium* species, although most is caused by *Fusarium graminearum*, *Fusarium culmorum*, *Fusarium avenaceum*, *Fusarium poae* and *Microdochium nivale* (Parry et al., 1995). FHB significantly reduces wheat grain yield and quality (Bai and Shaner, 1994). Yield losses results from reduction in the number of kernels and shriveled kernels. Grain quality is reduced due to accumulation of trichothecene mycotoxins, such as deoxynivalenol (DON), which pose a significant risk to human and animal health (McMullen et al., 1997).

The most practical and effective way to protect wheat from FHB is to develop resistant varieties. However, conventional breeding programs have been limited by a lack of effective resistance genes (Bai and Shaner, 1996; Rudd et al., 2001). Two major types of FHB resistance have been classified. Type I resistance is a reduction in initial infection after spray inoculation and Type II resistance is reduced spread of disease symptoms in the spike after point inoculation of a single floret on the wheat head (Schroeder and Christensen, 1963). The Chinese cultivar Sumai 3 exhibits Type II resistance, and is considered the most effective source of resistance, despite lacking complete resistance to FHB (Anderson et al., 2001). Quantitative trait loci (QTL) have been identified that confer Type I and Type II resistance (Buerstmayr et al., 2003; Waldron et al., 1999). Wheat breeding programs select for both Type I and Type II resistance to increase FHB (Liu and Anderson, 2003; Rudd et al., 2001).

Unfortunately, the evaluation of FHB resistance has been slow due to the necessity to avoid escapes by evaluating resistance in whole plants over several years and in environment factors (Browne and Cooke, 2004). Therefore, there has been interest in developing *in vitro* assays to provide methods for prescreening FHB resistance

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such as: detached leaf assay (Browne et al., 2005; Browne and Cook, 2005b; Browne, 2007; Diamond and Cooke, 1999), seedling resistance (Mesterhazy, 1995; Snijders, 1990), seed germination assay (Browne and Cook, 2005a; Browne, 2007, 2009), coleoptiles assay (Li et al., 2010; Wu et al., 2005) and response to the *Fusarium* mycotoxin deoxynivalenol (DON) (Buerstmayr et al., 1996). The detached leaf assay was successful in identification of an important component of the resistances of FHB in European wheat germplasm and may have utility as a mechanism for discrimination among different resistances in breeding program (Browne et al., 2004). The *in vitro* evaluation of partial disease resistance (PDR) against FHB has been related to an important component of whole plant FHB resistance in wheat cultivars. *Microdochium majus* is used in the detached leaf assays to detect leaf symptoms for observing PDR components (incubation period, latent period and lesion length) (Diamond and Cooke, 1999; Browne and Cooke, 2004). Lesion length is one of the components of PDR measured as an indicator of fungal pathogenicity and aggressiveness. PDR components detected in the *M. majus* detached leaf assay have been correlated to FHB resistance in wheat inoculated with *F. culmorum* and *F. graminearum* (Browne and Cooke, 2004; Browne et al., 2005; Browne, 2007; Diamond and Cooke, 1999). FHB resistance in seed germination assay was highly correlated in *F. graminearum*, *F. avenaceum*, *F. culmorum*, *M. majus* and *Microdochium nivale* indicating common resistance between *M. Majus* and other *Fusarium* spp. in the *in vitro* assay (Browne and Cooke, 2005a). Browne (2009) reported that PDR components (incubation periods, longer latent periods and shorter lesion lengths) in the detached leaf assay and higher germination rates in the seed germination assay were related to greater FHB resistance (Type II). However, the exotic wheat germplasms which provide highly effective resistances to FHB resistance do not appear to be detected in the detached leaf (Browne and Cook, 2004; Browne et al., 2005) or seed germination assays (Browne and Cook, 2005a).

There is still a need for a simple, rapid and reliable pre-screening method for FHB resistance. The objectives of this work were to develop alternative seedling stage screening methods and evaluate the use of a rapid seedling assay for FHB resistance on twenty-nine Korean winter wheat cultivars.

Materials and Methods

Plant materials. Twenty-nine winter wheat cultivars developed in Korea were used for these experiments. The

Korean cultivars were obtained from the National Institute of Crop Science, RDA.

Evaluation of FHB resistance in the greenhouse. FHB resistance (Type I and Type II) was evaluated in greenhouses at the National Institute of Crop Science, at Iksan. Seeds of wheat cultivars were surface-sterilized in 1% (v/v) sodium hypochlorite, rinsed three times in sterile distilled water, and placed on sterile moist filter paper in a Petri dish. Seeds were vernalized for 3 weeks at 4°C prior to germination. After vernalization, seedlings were planted into Sunshine Mix #1 (SunGro, Canada) in 15 cm round plastic pots in a greenhouse. Twenty seeds were planted for each line; each pot contained five seeds. To evaluate Type II resistance, a single central floret of the spikelet of the main stem was inoculated at anthesis with 10 µl of a macroconidial spore suspension (4×10^4 conidia/ml) of *F. graminearum* using a syringe. Wheat heads were covered with plastic bags for 3 days to maintain moisture. FHB disease severity was assessed as the percentage of spikelets on the inoculated spikes with visually detectable disease symptoms at 20 days after inoculation. Type I resistance was evaluated by spraying wheat with a macroconidial suspension (4×10^4 conidia/ml). Pots were covered in plastic cylinder for 3 days. The FHB disease severity was evaluated visually 20 days after the initial inoculation. Disease severity was measured as the percentage of symptomatic spikelets per spike.

Fusarium inoculums. Wheat spikes with head blight symptoms were collected from the wheat fields in Jeonbuk province in Korea during the spring 2009. Diseased wheat seeds were sterilized in 4% (v/v) sodium hypochlorite for 1 min, rinsed in sterile water for 2 min, and placed on potato dextrose agar (PDA) selective medium. The culture plates were incubated at 25°C for 7 days. Following mycelia growth, fungal plugs were transferred into carnation leaf agar (CLA) and plates were incubated under UV light at 25°C to induce spore formation (Leslie and Summerell, 2006). Conidia were washed with sterile water, and diluted to a concentration of 4×10^4 conidia/ml. To induce adhesion and spore germination, the inoculum contained 5.0% sucrose and 0.05% Silwet L-77 (Lehle Seeds, Round Rock, TX, USA). Single-spore culture of *F. graminearum* was used for in this study.

Seed germination assay. Two method of inoculation, soaking and soak-dry were compared through seed germination assay (Table 1). In soaking experiment, thirty seeds of wheat cultivars were surface-sterilized in 1% (v/

Table 1. Screen method of seeds germination and seedlings inoculated in *Fusarium graminearum*

Inoculation method		Treatment	Research methodology
Seed	Soaking	Sowing after imbibing seeds with conidial suspension (petridish/pot)	Germination (%)
	Soak-dry	Sowing after imbibing seeds with conidial suspension and dry	Germination (%)
Seedling	Spraying	Spray inoculation in 10 days old seedling	FHB incidence (%)
	Pin-point	Pin-point wounding inoculation after germination (5 days)	FHB incidence (%)
	Clip-dipping	Inoculation after seedling leaf-cutting and dipping (3 days)	Lesion length (cm)

v) sodium hypochlorite and rinsed in sterile distilled water for three times. Seeds were imbibed in 15 ml of a 4×10^4 conidia/ml with *F. graminearum* for 15 min. Controls were imbibed with sterile distilled water only. Seeds were then plated onto sterile moist filter paper in Petri dishes (10 seeds per Petri dish) at 15°C in a growth chamber with a 12/12 light/dark cycle. After one day, the seeds were planted in the greenhouse with each pot containing five seeds. The experiment was repeated twice. The number of seeds germinated was recorded as seedlings with coleoptiles length >1 cm at 7 days after inoculation. The number of germinating seeds inoculated with *F. graminearum* was divided by the number of germinated control seeds, and the results from two experiments were averaged. In soak-dry experiments, thirty seeds of wheat cultivars were sterilized and rinsed three times in distilled water. Seeds were imbibed in 15 ml of a *F. graminearum* suspension (4×10^4 conidia/ml). Controls were imbibed with sterile distilled water only. After 15 min excess conidia suspension was decanted off and the seeds were dried using filter paper. The seeds were plated onto sterile moist filter paper in Petri dishes and placed in a growth chamber. The experiment was repeated twice. The fraction of seeds germinating was calculated as described above.

Seedling inoculation assay. Three methods of inoculation, pin-point, foliar-spraying, and clip-dipping were compared through seedling assay (Table 1). In pin-point experiments, wheat seeds were germinated in Petri dishes on a stack of filter paper saturated with sterile distilled water. Three day old seedling stems (about 2–3 cm) were wounded by pin-point and inoculated with 10 ul of a suspension of conidia (4×10^4 conidia/ml). Controls were inoculated with sterile distilled water only. Twenty seedlings were inoculated and grown in a growth chamber under 12 h photoperiod cycle. The lengths of brown lesions on diseased stems were measured 7 days post inoculation.

For foliar-spraying and clip-dipping assays, 20 seeds of each cultivars were imbibed in a letter envelop (10×5 cm, length×height) and placed in plastic basket (15×8×10 cm, length×width×height) which has small hole can through

the water (see attached picture). These baskets were placed in plastic containers (60×40×14 cm) with enough water for seed germination. 10 day old seedlings were inoculated with a conidia suspension of *F. graminearum* by foliar-spraying or clip-dipping methods. In foliar-spray experiments, a conidial suspension (4×10^4 conidia/ml) was sprayed on both sides of leaves using an atomizer. Inoculated plants were placed in a moist chamber at 25°C for 3 days and then returned to greenhouse for disease evaluation. Controls were sprayed with sterile distilled water only. The disease incidence was determined as the percentage of infected seedlings with visually necrotic lesion and/or sporulation of fungal disease symptoms. In clip-dipping experiments, the tip of the coleoptiles were cutoff and then dipped in 20 ml of a suspension of conidia (4×10^4 conidia/ml) for 3 times. Inoculated seedlings were covered in plastic bag to maintain high humidity for 3 days and then moved to greenhouse for disease evaluation. Controls were dipped with sterile distilled water only. Lesions on the inoculated leaves were measured at 7 days post inoculation.

Statistical analysis. Statistical analysis of heading date, stem length and grain morphologies were done using SAS software (SAS Institute, NC, USA) using Fisher's least significant difference (LSD) procedure. Correlations between FHB resistances (Type I and Type II) and seedling assays (seed germination and seedling inoculation assay) were estimated using SAS CORR procedure.

Results

Evaluation of FHB resistance (Type I and Type II).

Twenty-nine Korean winter wheat cultivars were evaluated for Type I and Type II FHB resistance (Table 2). Spikes were spray inoculated of conidial suspension to test for Type I resistance, or the central spikelet of each plant was point inoculated with *F. graminearum* to test for Type II resistance. The symptoms of FHB disease showed after 7 days post inoculation, and disease resistance was scored two weeks later. For Type I FHB resistance, the fraction of plants showing disease symptoms ranged from 29.8%

Table 2. Percentage of Fusarium head blight (FHB) severity in the greenhouse evaluation for Type I and Type II resistance

Cultivars	Pedigree	FHB severity (%)	
		Spraying inoculation (Type I)	Point inoculation (Type II)
Ol	Norin72/Norin12	54.9	66.0
Geuru	Strampelli//69D-3607/Chokwang	52.5	22.8
Dahong	Norin72/Wonkwang	36.2	55.1
Chungkye	Norin4/Sharbati-Sonora	45.1	39.5
Eunpa	Chukoku81/Tob-CNO//Yuksung3///Suwon185	44.8	57.2
Tapdong	Chukoku81//Shinkwang/Toropi	74.6	42.9
Namhae	Ol/Calidad	43.8	17.3
Uri	Geuru/Ol	36.7	59.3
Olgeuru	Geuru'S/Chokwang//Seohae143	49.0	62.5
Alchan	Suwon210/Tapdong	68.1	95.6
Gobun	Eunpa/Tapdong//Eunpa/Shannung6521	67.9	24.1
Keumkang	Geuru'S/Kanto75//Eunpa	49.4	72.8
Seodun	Geuru/Genaro81	29.8	42.8
Saeol	Sirogane//Norin43/Sonalika	57.2	100.0
Jinpoom	Geuru/Genaro81	32.1	51.2
Milseong	Norin43/Sonalika	67.6	78.8
Joeun	Eunpa/Suwon242	74.3	51.5
Anbaek	Sae/Geuru	37.1	36.8
Jopoom	SW88416-B-0/SW89277	48.0	100.0
Shinmichal	Olgeuru//Kanto107/Baihuo	77.2	60.4
Jonong	Suwon234/SW80199-B-Y14-0	55.7	63.9
Jokyung	Seri82/Keumkang	54.0	81.8
Younbaek	Keumkang/Tapdong	64.9	48.3
Shinmichal1	Alchan/Kanto107//Baihuo	61.0	31.3
Dabun	Keumkang/SW97027	44.1	19.3
Baekjoong	Keumkang/Olgeuru	58.3	34.2
Jeokjoong	Keumkang/Tapdong	54.2	48.6
Sukang	Suwon266/Asakaje	59.6	24.4
Hanbaek	Shann7859/Keumkang//Guamuehill	43.4	48.8
Mean		53.1	53.0

to 77.2% and for Type II resistance from 17.3% to 100%. Namhae, Dabun, Geuru, Sukang and Gobun were the most resistant cultivars, and Saeol, Jopoom, and Alchan were the most susceptible cultivars for Type II resistance (Table 2). Although there was no significant correlation between FHB severity for Type I and Type II evaluation, Namhae and Dabun showed a high level of FHB resistance (Type I and Type II).

Seed germination assay. Seed germination rates, after imbibing seeds with conidial suspensions of *F. graminearum*,

showed large variations between cultivars (Table 3). Overall, exposure to *F. graminearum* reduced mean seed germination 65.5% relative to the water controls. The most resistant cultivars, those with the greatest germination as a percentage of water control from both soaking and soak-dry assays, were Dahong, Jonong, Gobun, and Baekjoong. Uri, Tapdong, Hanbaek and Jokyung were amongst the more susceptible cultivars. There was not a statistically significant correlation between either seed germination assay with FHB resistance (Type I or Type II). Correlation coefficients between the mean data from the two seed

Table 3. Disease reactions of seed germination assay inoculated with *Fusarium graminearum*

Cultivars	Seed germination assay		
	Soaking (%)	Soak-dry (%)	Mean ^a
Ol	45.5	71.4	58.4
Geuru	80.0	57.1	68.6
Dahong	90.5	100.0	95.2
Chungkye	73.7	68.4	71.1
Eunpa	50.0	64.7	57.4
Tapdong	22.7	72.7	47.7
Namhae	55.5	100.0	77.8
Uri	33.3	40.0	36.7
Olgeuru	46.2	91.7	68.9
Alchan	26.1	92.9	59.5
Gobun	69.6	93.3	81.4
Keumkang	33.3	81.3	57.3
Seodun	55.6	90.9	73.2
Saeol	52.6	66.7	59.6
Jinpoom	68.0	78.6	73.3
Milseong	71.4	66.7	69.0
Joeun	84.6	63.2	73.9
Anbaek	73.9	63.6	68.8
Jopoom	55.6	50.0	52.8
Shinmichal	61.5	64.7	63.1
Jonong	70.8	93.3	82.1
Jokyung	66.7	35.3	51.0
Younbaek	60.9	58.8	59.8
Shinmichal1	76.5	50.0	63.2
Dabun	39.3	93.3	66.3
Baekjoong	73.1	78.6	75.8
Jeokjoong	72.7	66.7	69.7
Sukang	71.4	64.3	67.9
Hanbaek	61.1	40.0	50.6
Mean	60.1	71.0	

^athe mean data of seed germination assay both soaking and soak-dry

germination assays (soaking and soak-dry) and FHB Type II resistance were significant ($r=-0.37$; $P<0.05$).

Seedling inoculation assay. Seedlings inoculated with *F. graminearum* conidia by spraying, pin-point and clip-dipping showed clear disease symptoms within a few days after inoculation (Table 4). Stems and coleoptiles began to turn brown 3 days post inoculation and brown lesions developed by 7 days to varying degrees as a results of the seedling resistance. Brown lesions started from the pin-

Table 4. Disease reactions of seedling assay inoculated with *Fusarium graminearum*

Cultivars	Seedling inoculation assay		
	Spraying (%)	Pin-point (%)	Clip-dipping (cm)
Ol	5.0	23.3	2.2±0.7
Geuru	45.5	48.0	3.6±1.3
Dahong	50.0	10.0	2.2±0.6
Chungkye	42.1	23.3	2.2±1.0
Eunpa	66.7	43.3	3.0±1.0
Tapdong	12.5	33.3	1.6±0.6
Namhae	25.0	10.0	0.9±0.3
Uri	46.7	10.0	2.1±1.0
Olgeuru	53.3	15.0	2.2±0.5
Alchan	10.5	23.3	4.2±1.5
Gobun	11.8	11.1	1.0±0.6
Keumkang	50.0	18.5	2.7±0.7
Seodun	100.0	10.0	2.0±0.5
Saeol	75.0	25.0	3.0±1.1
Jinpoom	5.9	18.5	2.1±0.7
Milseong	30.0	14.8	1.3±0.3
Joeun	85.7	63.3	2.2±1.4
Anbaek	50.0	50.0	2.1±1.4
Jopoom	26.3	23.1	2.2±0.5
Shinmichal	11.8	31.0	1.9±0.4
Jonong	25.0	50.0	3.2±1.7
Jokyung	40.0	27.6	2.4±0.7
Younbaek	90.0	32.0	2.0±0.9
Shinmichal1	-	36.4	1.8±0.8
Dabun	23.5	18.5	1.9±1.0
Baekjoong	-	44.0	2.2±0.4
Jeokjoong	-	42.9	2.6±0.5
Sukang	-	19.2	1.1±0.5
Hanbaek	-	33.3	2.8±1.0
Mean	23.5	27.9	2.2

point area and cut tips of coleoptiles inoculated with soaked in conidial suspension (Fig. 1). Resistance in the clip-dipping assay was determined by the length of the lesions. The average lesion lengths across the twenty-nine cultivars for clip-dipping ranged from 0.9 cm to 4.2 cm (Table 4). Namhae, Gobun, Sukang and Milseong were amongst the most resistant for clip-dipping inoculation experiment. The more susceptible cultivars Alchan, Geuru and Jonong showed an average lesion length of 4.2 cm, 3.6 cm, and 3.2 cm, respectively. Correlation between pin-point and Type

Fig. 1. Clip-dipping inoculation assay using wheat seedling inoculated with *Fusarium graminearum*. (A) Wheat seedling grown in the letter envelopes and then removed the coleoptiles for inoculation. (B) The disease symptoms on the wheat coleoptiles 7 days after inoculation (left: inoculated with water control, right: inoculated with *F. graminearum*). (C) Various length of lesion after inoculation (Control, low, medium, high, respectively).

II resistance were not significant. However, correlation coefficients between the lesion lengths of clip-dipping inoculation and FHB Type II resistance was significant ($r=0.45$; $P<0.05$) (Fig. 2).

Discussion

Developing FHB resistant varieties of wheat is the most practical approach for minimizing economic losses from this disease. Type I resistance is resistance to the initial infection, and it is assayed by inoculating wheat spikes. FHB Type II resistance is defined as resistance to the spread of the pathogen within the spike, and is determined by point inoculation of a single spikelet (Schroeder and Christensen, 1963). In addition to measuring visual symptoms of infection on spikes, the levels of *Fusarium*-damaged kernels (FDK) and the mycotoxin deoxynivalenol (DON) may also be used to measure resistance to FHB (Mesterhazy et al., 1999). Field and greenhouse screening has limitations associated with time, space, and environmental factors for evaluating FHB resistance. In this study, we evaluated seed and seedling assays for rapid and mass screening of Korean wheat cultivars for resistance to FHB.

Seed germination and seedling inoculation assay was selected on basis of their similarity to adult plant spike inoculation using spraying and point inoculation assays for Type I and Type II resistance. Using point inoculation (Type II) on adult spikes, five cultivars (Namhae, Dabun, Geuru, Sukang and Gobun) showed resistance to *F. graminearum* with less than 25% FHB. Seedling inoculation assays for Type II resistance (pin-point and clip-dipping) detected resistance in four of the five cultivars (Namhae, Dabun, Sukang and Gobun). Pin-point and cut-dipping seedling

inoculation assays showed similar results for the different cultivars, however the clip-dipping assay permitted greater numbers of cultivars to be screened. Therefore, it has been proposed that cut-dipping inoculation assay was an efficient method for seedling stage compared to FHB resistance.

Seed germination assays exhibited higher FHB incidence compare to spike inoculation method, however results from these assays were poorly correlated with the degree of resistance in adult plants. Similarly, FHB resistance determined by spray inoculation of seedlings was also poorly correlated with resistance in adult plants. Thus, these assays both were not proper methods to selection for FHB resistance.

Detached leaf assay have been used primarily for screen for FHB in wheat cultivars. In these assays, partial disease resistance (PDR) components measured after exposure to *Fusarium* spp. and *Microdochium majus* included incubation period, latent period and lesion length (Diamond and Cooke, 1999; Browne and Cooke, 2004; Browne, 2007). PDR components detected in the *M. majus* detached leaf assay have been correlated to FHB in wheat inoculated with *F. graminearum*. Using the detached-leaf assay, whole-plant FHB resistance has related to an important PDR component in commercially grown European wheat cultivars and the 30 USA soft red winter wheat entries in the 2002 Uniform Southern FHB Nursery (Browne and Cook, 2004; Browne et al., 2005; Diamond and Cook, 1999). In addition, resistance determined *in vitro* using a seed-germination assay correlated with whole-plant FHB resistance ratings among European wheat cultivars. The PDR component incubation period was highly related to FHB disease incidence while resistance detached in the seed germination assay related to a greater decline in the

Fig. 2. Single linear correlation between clip-dipping inoculation assay and spraying inoculation (Type I) (A) and point inoculation (Type II) (B) with *F. graminearum*.

level of FDK and a smaller reduction in DON than would be expected from the reduction in FHB diseases on the wheat spike (Browne, 2007). Browne (2009) showed that higher germination rates in the seed germination were greater FHB resistance measured by single point inoculation strongly related to resistance assessed in spike (Type II). In this report, FHB resistance determined by seed soaking or soak-dry methods did not predict Type I resistance or Type II resistance in adult plants. The average of germination rate both the soaking and soak-dry experiments were negatively related with Type II resistance in Korean wheat cultivars.

Clip-dipping inoculation promotes the interaction between wheat tissues and fungus. The coleoptiles tips are removed, allowing fungi to penetrate plant tissues. Thus, disease development by pathogenic fungi with wounding is manifested through appearance of symptoms such as discolored, necrotic area on the affected coleoptiles tips. FHB disease symptom showed that wheat cultivars with resulted in various lesion lengths in a growth chamber under strictly controlled conditions. Wu et al. (2005) observed that disease lesion length on coleoptiles inoculation of wheat with *F. graminearum* isolates was significantly correlated with

disease development on adult plants of the same genotype under field conditions. Our results show that the lesion lengths of clip-dipping inoculation was also significant correlated to FHB Type II resistance ($r=0.45$; $P<0.05$), however results were not significantly correlated with type I resistance in the field condition. It has been proposed that the individual PDR components not only influence the total level of resistance but also have a variable individual influence on disease development parameters.

This study was conducted to develop an easy seedling method to test large numbers of wheat cultivars for FHB resistance. The clip-dipping inoculation assay is a simple method to use for the FHB severity. It also proved to be a high-throughput method and reliable pre-screening method for FHB resistance. Using this technique, it should be possible to screen large populations during seedling stage in Korean wheat breeding programs, and develop wheat cultivars with improved FHB resistance.

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