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Relationship between resistance and tolerance responses of barley DH lines to *Pyrenophora graminea*

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Abstract

Barley leaf stripe (BLS), caused by the fungus *Pyrenophora graminea*, is an important seed-borne disease that causes *substantial crop yield losses* globally. The recent BLS score scale does not adequately reflect a cultivar's true performance, as it neglects its tolerance. Since, resistance reveals the ability of a plant to reduce the extent of pathogen infection, whereas, tolerance is the plant's ability to yield, in spite of being infected. Consequently, a more informative BLS score system is essential to address this issue. For this goal, 40 doubled haploid (DH) lines were tested in this work using both resistance and tolerance traits. Data showed significant differences among DH lines with a wide spectrum of disease responses ranging from high to low levels based on the percentage of leaf infected rating scale. However, even though B08-AS-3 genotype had a high BLS infection level (94 %), it had a moderate grain yield per plant (7.8g), similarly with B08-AS-11, 15 and 18 lines. Furthermore, the most resistant lines B08- AS-5 and 12 did not give a high yield. A correlation (r =0.59, P<0.01) was found between resistance and tolerance, indicating that the reaction of barley genotypes to this disease should therefore consider not only the resistance rating of a line, but also its tolerance which can reflect a genotype's true performance.

Keywords: Barley (*Hordeum vulgare* L.), leaf stripe, resistance, tolerance

1. Introduction

Pyrenophora graminea [anamorph *Drechslera graminea* (Rabenh. ex Schltdl.) Ito], the causal agent of barley leaf stripe disease, is one of the most widely distributed seed-borne pathogens globally. It is considered economically important as it can cause *substantial* reduction in crop yield [1,2].

The fungus can infect barley plants during seed germination, and hyphae accelerate its intercellular spread within the coleorhizae, the embryo, the roots and scutellar node, then symptoms are becoming visible on leaves as yellow and brown-colored longitudinal stripes [3,4]. Yield loss in susceptible cultivars could exceed 70% [1] and is attributed to a reduction in the number of spikes, grain size and tillers [5,6].

The use of barley resistant cultivars remains the most appropriate and economical way to control this disease. However, currently there are lacking germplasm resources with resistance to BLS [7], consequently, screening of a bigger number of genotypes is desirable to find new resistant sources.

Both incidence (diseased plants) and severity (symptoms) are the commonly used measures to estimate BLS disease [8]. However, this strategy is useful in determining the plant reaction but without measuring the plant yield, despite being infected.

Resistance and tolerance are known to be the two important mechanisms to protect plants against pathogens [9]. Resistance is typically defined as the ability of a plant to restrict the infection by the pathogen, or restrict its growth throughout the plant, by dissimilarity, tolerance is usually measured as the ability of the plants to give yield, although being infected [10]. Thus, it is important to understand the complex relationships between barley plant resistance, and tolerance because not all genes that confer biochemical resistance (in terms of reducing symptoms) will necessarily increase yield [11]. Hence, accurate measurement under BLS conditions is crucial to enhance our capacity for selection based on a cultivar's true performance.

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No studies on the reactions between BLS and barley resistance/tolerance were so far stated in the literatures. In this work, we used two barely DH populations to determine their performance under BLS artificial infection using the both parameters resistance and tolerance.

2.Materials and Methods

2.1. *Plant material*

A total of 40 barley double haploid (DH) lines produced according to Kasha and Kao [12] were used in this investigation. They were created through five resistant-by-susceptible barley crosses made among six parents possessing different BLS reactions. Arabi Abiad is a Syrian local cultivar, PK130-36 was received from Pakistan, Igri is a German cultivar, IC-9 was developed at the International Center for Agricultural Research in Dry Areas (ICARDA), Arrivate was received from USA and CI5791 is an Ethiopian cultivar. Barley spikes were manually emasculated and pollinated with fresh pollen. Then, they were treated with 2,4-dichlorophenoxyacetic acid (2,4-D). Tillers were collected from donor plants at mid- to late-uninucleate stage of microspore development.. Anthers were transferred to FW culture medium [4]. Haploid plants were vernalized for 8 weeks. Later, haploid seedlings were treated with colchicine for chromosome doubling.

2.2. Seed inoculation with P. graminea

The most Syrian virulent isolate PgSy3 was used in this work [1]. Inoculation was achieved according to Hammouda [13]. Seeds were surface-sterilized in 2% sodium hypochlorite for 5 min, dried for 3-4 h, then placed on 8-day-old mycelial culture growing on potato dextrose agar (PDA, DIFCO, Detroit, MI, USA) medium in Petri dishes and incubated for 14 days at 6 °C in the dark. As a control, seeds were incubated on PDA medium alone.

2.3. Field experiments

Inoculated and un-inoculated seeds of the DH lines and parents were planted under field conditions in Syria, at a site of 970 m altitude (550 mm rainfall average) with three replicates. The location of the experiment was selected to be favorable for the development of BLS disease. Each plot consisted of 1 x 1 m with a 1 m buffer. Each plot consisted of 5 rows 20 cm apart with 50 seeds sown per row. Cultural practices were as previously described [1].

2.4. Disease evaluation

BLS assessment was visually recorded as infected leaf area per plant expressed as a proportion of the total area using a scale described by Delogu et al. [14] where; resistant (R) (0-11%), moderately resistant (MR) (12-26%), susceptible (S) (27-78%) and highly susceptible (HS) (79-100% of infected plants).

2.5. Yield estimation

Three central rows of each replicate plot were harvested at maturity stage to measure grain yield (g/plant) where; High (H) > 10 - Medium (M): 7-10 and Low (L) less than 7.

2.6. Statistical analyses

Data was analyzed using the STAT-ITCF statistical programme. Variations between means were evaluated using Newman-Keuls test at 5% probability level [15].

3. Results and Discussions

In the present work, six barley parents with different levels of resistance to *BLS* were used. Data showed that the disease caused more severe infection on the highly susceptible parent ‘Arabi Abiad’ as compared with the resistant ones ‘Igri’. Furthermore, the disease symptoms (Fig. 1 and 2) were typically detected in infected plants which is in agreement with our earlier field observations [1].

According to the scale of Delogu et al. [14], reactions of the 40 progeny lines to BLS were categorized into four groups (Fig. 3). However, significant differences ($P < 0.05$) in mean severity values were detected among different lines, and a continuum of genotypic reactions to the virulent strain SY3from highly resistant to highly susceptible was observed (Fig. 3).

Data revealed that B08-AS-3 had a high BLS infection level (94 %), it had a moderate grain yield per plant (7.8g), similarly with B08-AS-11, 15 and 18 lines. However, the most resistant lines B08- AS-5 and 12 did not give a high yield. Furthermore, the correlation between resistance and tolerance to BLS was r =0.59, P<0.01 (Fig. 4), indicating that reaction of barley genotypes to this disease should therefore consider not only the resistance rating of a line, but also its tolerance which can reflect a genotype’s true performance.

Figure 1. BLS symptoms on the resistant (left) and susceptible and highly susceptible (right) barley DH lines.

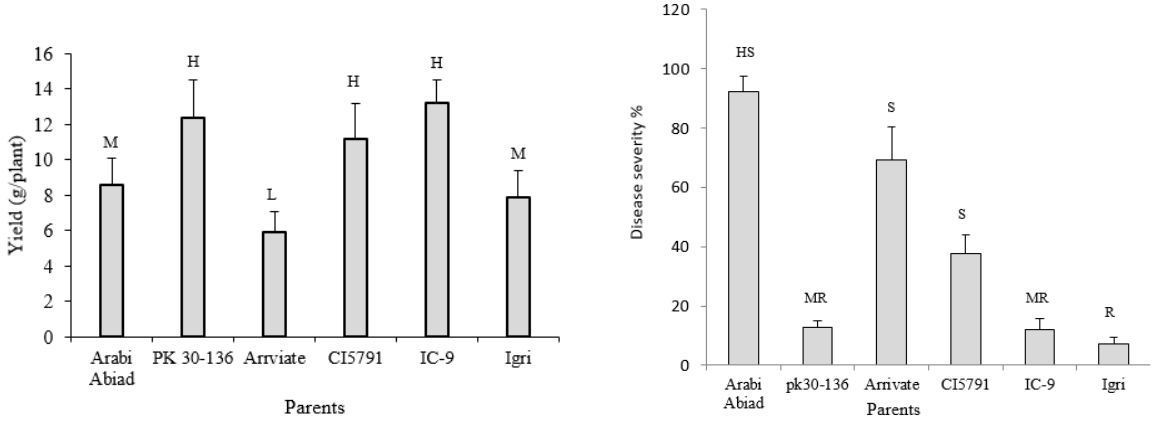

Figure 2. Yield (g/plant; H; High, M: moderate and L: low; see the text), and frequency of BLS reactions incited on the barley parents according to the scale of Delogu et al. (1989) where; resistant (R) (0-11%), moderately resistant (MR) (12-26%), susceptible (S) (27-78%) and highly susceptible (HS) (79-100% of infected plants).

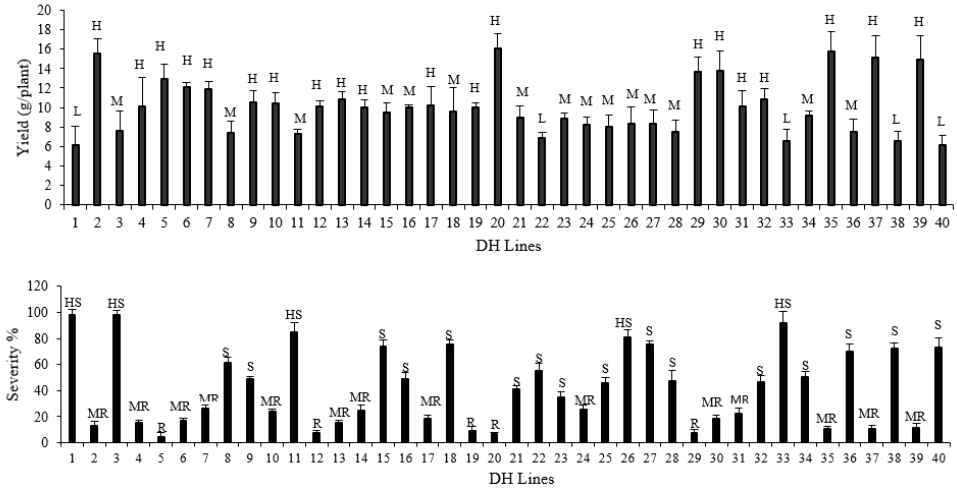

Figure 3. Yield (g/plant; H; High, M: moderate and L: low; see the text), and frequency distribution of BLS infection response of 40 DH barley according to the scale described by Delogu et al. (1989). Data were obtained from three crosses of barley parents.

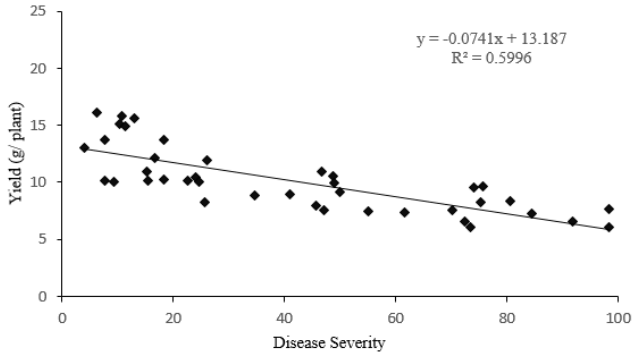

Figure 4. Relationship between BLS severity and yield (g/plant) of 40 barley DH lines

Our findings are in agreement with those of Politowski and Browning [16] on oat and Mikaberidze and McDonald [9] on wheat.

It has been reported that barley plant may be able to compensate the reduction in one yield component and continue to yield well in spite of fungal pathogen infection [17,18]. Van der Meijden et al. [19] suggested that the evolution of tolerance or resistance to plant damage would depend on the quantity of nutrients obtainable and on the growth rate of plant. In many plant-fungal interactions, it has been revealed that plants can compensate for reduced CO_2 fixation in infected tissues by increasing photosynthesis in healthy parts of infected leaves and/or in uninfected leaves. For instance, tolerance of *Senecio vulgaris* to the fungi *Puccinia lagenophorae* and *Coleosporium tussilginis* has been revealed to be associated with higher CO_2 fixation at the total plant scale [20]. Furthermore, Scholes et al. [21] confirmed that the presence of green photosynthetic areas in oat leaves inoculated with fungus *Puccinia coronata* could compensate for the decrease of photosynthetic activity in other plant parts of the same leaf injured by the pathogen. Here, the effect of BLS on grain yield may be attributed to reduced area of photosynthetically which enhances respiration and transpiration [22].

On the other hand, the ability of barley genotypes to yield well when damaged by BLS may indicate a level of tolerance to the disease. Here, it might explain that in spite of BLS infection, barley plants have the ability to develop yield components to compensate for initial tiller reduction and also by inherent tolerance to the disease. However, identification of new sources of resistance to BLS and their introduction into cultivated crops is a very important element of breeding programs [7]. Therefore, barley breeders need to identify whether a BLS yield loss amount can be attributed to resistance or tolerance in the tested lines. One modern helping method is to use molecular markers which can alleviate some of these issues. Moreover, DH lines with low disease levels were associated with high yield although some of them are not the highest yielding.

4. Conclusion

The current work revealed that accurate measurement and classification of barley cultivars performance under BLS infection is crucial for selection. A relationship was found between resistance and tolerance (r =0.59, P<0.01), this might propose that resistance and tolerance can be combined to achieve an additive effect for obtaining a cultivar's true performance during barley- *P. graminea* evaluations.

Compliance with Ethics Requirements. The authors declare that they comply with the Ethics requirements of the journal. The authors declare that they have no conflicts of interest and that all procedures involving human or animal subjects (if any) comply with specific regulations and standards.

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