

Identification of Resistance Sources to Cereal Cyst Nematode, *Heterodera avenae* Wollenweber in Barley (*Hordeum vulgare* L.)

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Abstract

A field study was carried out to identify the sources of resistance for cereal cyst nematode in Barley at three locations; Ludhiana, Durgapura and Hisar during three crop seasons from 2015-18. Out of 625 screened entries at three locations, none of the entry was found to be resistant to all the three populations, however, the four lines i.e. RD-2977, BH-959, RD-2794 and RD-2957 were found resistant to Jaipur and Hisar CCN population only. Further, PL-874 (2015-16), RD-2927 (2016-17) and BH-946 (2017-18) were found moderately resistant to Ludhiana and Jaipur populations. These results clearly indicate that Ludhiana population was more virulent than other two populations. Therefore, plant breeders can use PL-874, RD-2927 and BH-946 to breed barley varieties resistant against Ludhiana CCN populations.

Keywords: Barley, cereal cyst nematode, resistance.

1. Introduction

Barley (*Hordeum vulgare* L.) is one of the oldest cereal crops evolved as a source of food grain, feed and malting grain and is largely referred as a poor man's food (Baik and Ullrich, 2008). During 2017-18, barley was grown in an area of 0.68 million ha and gave total production of 1.7 million tons with a productivity of 26.3 q/ha (Anonymous, 2018). It is gaining importance in nutraceutical diets and has potential health benefits due to soluble fiber β -glucan (Behall *et al.*, 2004; Johansson *et al.*, 2013), which helps in lowering cholesterol level, improving lipid metabolism, and reducing glycemic index (Granfeldt *et al.*, 1994; Delaney *et al.*, 2003; Brennan and Cleary 2005; Keenan *et al.*, 2007; Johansson *et al.*, 2013). Several biotic and abiotic factors hinder the production of barley. Cereal cyst nematode (CCN), *Heterodera avenae* is one of the biotic root sedentary endoparasite which causes 'molya' disease in barley and wheat in northern parts of India and delimit the production substantially under severe infestations.

It was first reported from Sikar district of Rajasthan by Vasudeva in 1958. Presently, it has been distributed throughout India of major wheat growing regions. The disease is characterized by yellowing of plants, stunted patchy growth with reduced number of tillers, delayed emergence of ears, reduced number and size of spikelets and grains. Presence of white glistening cysts seen by mid February on roots is only confirmation of CCN infestation, (Kort, 1972). Economically it can cause up to 97.28 million rupees in wheat (Jain *et al.*, 2007). Even though, CCN can be managed through crop rotation with non-host crop, (Handa, 1983; Handa *et al.*, 1975) but many a times it's not economical to farmers. The development of resistant cultivar is the most promising method for managing *H. avenae*. Several sources of resistance found associated with a single dominant gene, are available in barley (Yadav *et al.*, 1987) but breeding for resistance is complicated due to the presence of different pathotypes of *H. avenae* with

different levels of virulence. Nevertheless, a number of resistant barley cultivars have been developed in India but either they are not under cultivation or specific to a particular region (Bhatti and Dahiya, 1992; Bishnoi and Yadav, 2002; Kaur and Dhindsa, 2006; Shekhawat *et al.*, 2018). Hence, identification of new resistance source to develop resistant cultivars is most important method to manage CCN. Therefore, an attempt was made to screen barley elite lines at three different locations i.e. Ludhiana, Durgapura (Jaipur) and Hisar for respective CCN pathotypes.

2. Materials and methods

Location of Experiment and Plant material: The current study was conducted in three location *viz.*, Wheat section, Department of Plant Breeding and Genetics, PAU, Ludhiana (Ludhiana CCN population), Rajasthan Agricultural Research Institute, Durgapura, Jaipur (Jaipur CCN population) and Department of Plant Breeding and Genetics, CCSHAU, Hisar (Hisar CCN population) during the wheat crop season 2015-16, 2016-17 and 2017-18. A total 210, 198 and 217 different elite barley lines breeding were received from ICAR-Indian Institute of Wheat and Barley Research, Karnal during 2015-16, 2016-17 and 2017-18, respectively for CCN screening.

Nematode Culture and Screening: CCN pure culture maintained at respective locations was used in the experiment. Three seeds of each lines were sown in 12.5cm diameter and 11 cm deep pots, filled with 1000 cm³ sandy loam soil infested with two cysts of the nematode/250 cm³ (averaging 250 eggs/cyst), in first fortnight of November during 2015-16, 2016-17 and 2017-18. After two weeks of germination, thinning was ensured to single seedling per pot. Three replications of each line were properly maintained. Pots were irrigated as and when required, to maintain proper growth and development. The cyst from each pot was determined after 90 days of sowing in March 2016, 2017 and 2018 by following Cobb's decanting and sieving technique. The soil from each pot was washed and sieved through 20 and 60 mesh sieves. The residue collected from 60 mesh sieve was observed under binocular stereoscopic microscope for counting cysts number per plant. The reaction of each line against CCN was categorized according to the scale suggested by the All India Coordinated Wheat and Barley Improvement Project, ICAR-IIWBR, Karnal (Table 1). CCN screening of barley test entries in pot experiment was conducted at all three locations but in sick fields, this experiment was only carried out at PAU Ludhiana and RARI, Durgapura, Jaipur only.

Table 1: CCN rating scale in wheat (Given by the All India Coordinated Wheat and Barley Improvement Project, Karnal, Haryana)

Rating	No. of Cysts per plants	Host reaction
1	0	Highly Resistant (HR)
2	0.1 to 4	Resistant (R)
3	4.1 to 9	Moderately Resistant (MR)
4	9.1 to 20	Susceptible (S)
5	>20	Highly Susceptible (HS)

3. Result and discussion

Among three sets of barley elite breeding lines screened at different locations (Ludhiana, Jaipur and Hisar) in three years (2015-16, 2016-17 and 2017-18) shown lot of variability in resistance with respective to local CCN populations. Out of 210 in 2015-16, 19 lines were found resistant to only Hisar CCN population and 5, 13 and 1 entries were found moderately resistant to all three respective CCN populations (Table 2 & Fig 1). Remaining entries were found either susceptible or highly susceptible. The highest percentage of resistant lines (10%) was found against Hisar CCN population. The moderately resistant lines 2% and 6% were found against Ludhiana and Jaipur CCN population, respectively (Fig 2). However, BCU-7643 and PL-874 lines showed moderately resistant to both Ludhiana and Jaipur CCN population (Table 2). In 2016-17 there was a 1% and 7% of resistant lines in Jaipur and Hisar CCN population (Fig 2). Amongst one ninety eight lines, only one and thirteen lines were found resistant to CCN of Jaipur and Hisar population. Ten and eight lines were moderately resistant to CCN population of Ludhiana and Jaipur, respectively. Nevertheless RD-2927 was found moderately resistant to both Ludhiana and Jaipur CCN population and DWRB-73 was found moderately resistant to Jaipur and Hisar CCN population (Table 2 & Fig 1). Left over entries was fallen into susceptible or highly susceptible categories. The same trend continued in the 2017-18 also, out of two hundred and seventeen lines tested, the eight and thirty lines were found resistant to Jaipur and Hisar CCN population and eleven, ten and two lines found moderately resistant to Ludhiana, Jaipur and Hisar CCN populations (Table 2 & Fig 1). Only 4 % and 15 % of tested lines were found resistant to Jaipur and Hisar CCN populations but none of the line was found resistant to Ludhiana CCN population (Fig 2). However, the four lines i.e. RD-2977, BH-959, RD-2794 and RD-2957 were found resistant to Jaipur and Hisar CCN population. Two lines PL-898 and BH-946 were found moderately resistant to CCN population of Ludhiana and Jaipur (Table 2). Remaining all the barley entries/lines was either susceptible or highly susceptible to all the geographical

Table 2: Resistant and moderately resistant identified barley elite breeding lines/entries found during three seasons.

SN	Host Reaction	Lines/Entries
1	Resistant	2015-16 Hisar: HUB-242, HUB-246, JB-322, JB-325, JB-328, RD-2925, UPB-1054, DWRB92, BCU-7594, BCU-7602, BCU-7615, BCU-7624, BCU-7625, BCU-7732, DWRB-127, HUB-241, HUB-242, KB-1367, RD-2903. 2016-17 Jaipur: BH-1018 Hisar: DWRB-163, JB-347, KWS IRINA, PL-895, PL-897, PL-898, RGT PLANET, TRAVELLER, DWRB-73, DWRB-91, DWRB-64, PL-751, BK-1508 2017-18 Jaipur: RD-2977, RD-2984, AZAD, BH-959, RD-2794, RD-2949, RD-2957, HUB-247 Hisar: Andreia, Danielle, PL-898, RD-2977, BH-946, BH-959, RD-2794, RD-2957, DWRB-185, DWRB-186, Explorer, HUB-264, JB-360, NDB-1699, PL-902, Planet, RD-2948, RD-2969, RD-2972, RD-2975, RD-2978, RD-2980, RD-2981, RD-2988 Xanadu, DWRB-137, K-603, RD-2035, DWRB-170, VLB-151
2	Moderately Resistant	2015-16 Ludhiana: BCU-7643, PL-874, RD-2901, VLB-130, VLB-137 Jaipur: DWRB-156, NDB-1650, RD-2943, RD-2552, RD-2715, RD-2849, BCU-7643, BCU-7719, BH-1001, PL-874, RD-2900, RD-2910, RD-2914. Hisar: BCU-7601 2016-17 Ludhiana: KB-1521, NDB-1665, RD-2927, RD-2954, TRAVELLER BHS-352, BHS-400, KB-1318, BCU-7746, BHS-430 Jaipur: DWRB-162, HBL-778, RD-2917, RD-2927, RD-2960, RD-2966 DWRB-73, RD-2912 2017-18 Ludhiana: BHS-464, JB-362, PL-898, RD-2973, RD-2986, Traveller, KB-1531, RD-2947, DWRB-147, DWRB-150, HUB-113 Jaipur: HUB-253, RD-2974, RD-2976, BH-946, DWRB-123, BH-1009, RD-2955, PL-890, VLB-154, BHS-380 Hisar: DWRB-181, HBL-113

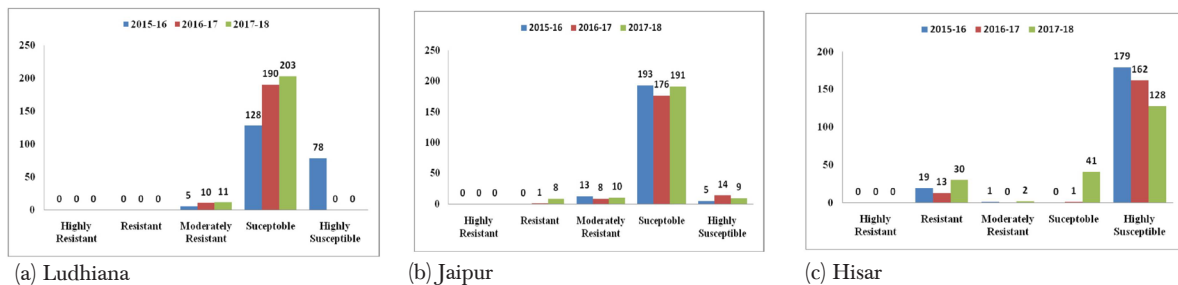


Fig.1: Reaction of barley elite lines/entries against CCN populations during three cropping seasons: (a) Ludhiana (b) Jaipur (c) Hisar

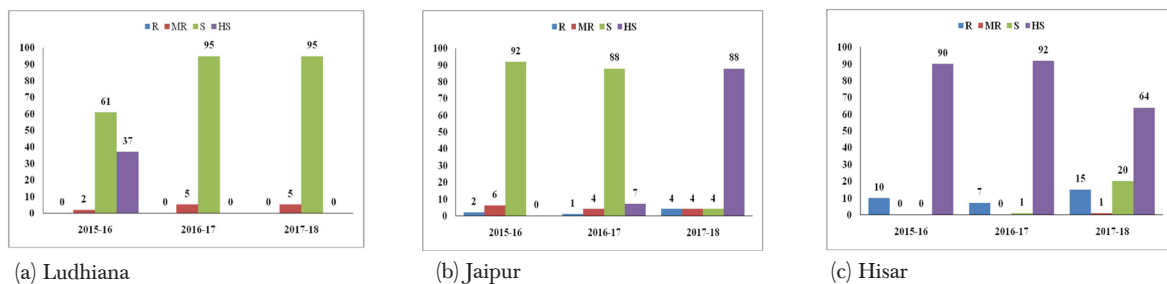


Fig.2: Percent reaction of barley elite lines/entries against CCN populations during three cropping seasons: (A) Ludhiana (B) Jaipur (C) Hisar

isolated CCN population. In all the three locations, none of the barley lines were found resistant to all three CCN populations, specifically for Ludhiana populations. Existence of multiple pathotypes in India was proved by the several researchers based on host differentials. Mathur *et al* (1974) confirmed the presence of five pathotypes of CCN from Rajasthan and Haryana based on assortment of cereals and grasses. Similarly, Swarup *et al* (1979) differentiated the Punjab CCN pathotype from Rajasthan and Haryana pathotypes. Bishnoi and Bajaj (2004) studied the CCN pathotype complex of eight Indian populations based on morphometric characteristics and international host differentials in which they concluded that Ambala, Himachal Pradesh and Punjab CCN populations were different (*H. filipjevi*) from Delhi, Jaipur, Mahendergarh, Sirsa and Udaipur CCN populations. Apart from this, biochemical and molecular studies were also proved the presence of different pathotypes. The study of isozyme, malate dehydrogenase pattern differentiated the Ambala, Himachal Pradesh and Punjab CCN population from Delhi, Jaipur, Naranaul, Sirsa and Udaipur CCN populations by detecting single band of MDH with Rf value 0.34. (Bishnoi *et al* 2004). A molecular characterization of Delhi and Punjab (Hoshiarpur and Ludhiana) population using PCR-RFLP of ribosomal DNA revealed, existence of intraspecific variability in CCN and occurrence of *H. filipjevi* populations at Punjab (Rao *et al* 2004). These results clearly indicate that Ludhiana CCN population was entirely different pathotype and more virulent than other two CCN population of Jaipur and Hisar. Therefore, plant breeders can use PL-874, RD-2927 and BH-946 to develop resistant variety against Ludhiana CCN populations.

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