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Resistance to Blast (*Magnaporthe grisea*) in a Mini-Core Collection of Finger millet Germplasm

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Abstract. Blast caused by Pyricularia grisea [teleomorph: Magnaporthe grisea] is an economically important and widespread disease of finger millet in the world. Host resistance is the most economical and effective means of combating this disease as finger millet is predominantly grown by resource-poor and marginal farmers. At International Crops Research Institute for the Semi-Arid Tropics (ICRISAT), we evaluated a finger millet mini-core collection of 80 germplasm accessions (about 1% of the total germplasm collection representing major trait variability) for blast resistance both in field and greenhouse. Field evaluation was done using a refined screening technique that included new improved rating scales for leaf, neck and finger infection. Sixty six of the 80 accessions showed combined resistance to leaf, neck and finger blast in two seasons (2009 and 2010) of field screening. A highly significant and positive correlation was found between neck and finger blast ratings (r = 0.92), whereas small but significant correlations were found between leaf blast and neck blast (r = 0.25) and between leaf blast and finger blast (r = 0.30). These accessions were also screened for leaf blast resistance in the greenhouse by artificial inoculation of seedlings to confirm field observations. Fifty-eight of the 80 accessions were resistant to leaf blast in the greenhouse screen as well. These resistant accessions represented one wild (africana) and four cultivated races (vulgaris, plana, elongate and compacta) of finger millet that originated from

13 countries in Asia and Africa and exhibited considerable diversity for agronomic traits, such as maturity period, plant height and panicle type. These blast resistant accessions from the mini-core collection would be useful in finger millet disease resistance breeding programs.

#### Introduction

Finger millet [Eleusine coracana (L.) Gaertn.], a widely grown traditional and highly nutritious grain cereal cultivated in the semi-arid areas of Eastern and Southern Africa and South Asia, is a staple food for millions of poor people. The precise data on world area under finger millet are not available, because it is frequently reported with other millets including pearl millet (in the FAO database). However, as per the estimate by the Consultative Group on International Agricultural Research (CGIAR), finger millet contributes to about 10 per cent of the total area (34.6 m ha) planted to millets (FAO 2004). In India, finger millet ranks next to pearl millet and is cultivated on 2.6 m ha area with a production of about 3.0 m t (www.indiastat.com). Finger millet is being increasingly recognized as a highly nutritious food for the weak and immuno-compromised people (Takan et al. 2012). The grains are rich source of protein (Malleshi and Klopfenstein 1998), fiber, minerals (calcium, iron, zinc, and manganese) and amino acids (tryptophan, cystine, and methionine), which are crucial to human health and growth, and these are deficient in most cereals. The nutritional quality of finger millet grain makes it an ideal food for expectant women, lactating mothers, children, the sick, and diabetics (National Research Council 1996). Thus, finger millet plays the key role in the livelihood of small-holder farmers in the semi-arid areas of Africa and Asia. However,

diseases and insects, in addition to abiotic stresses, are major impediments toward realizing the high yield potential of finger millet cultivars.

Among the diseases, blast caused by *Pyricularia grisea* (Cooke) Sacc. [teleomorph: Magnaporthe grisea (Hebert) Barr.] is very prominent and affect the productivity, utilization and trade of finger millet within Eastern and Southern Africa (Mgonja et al. 2007) and South Asia. The average loss due to blast has been reported to be around 28-36% (Nagaraja et al. 2007), and in certain areas yield losses could be as high as 80-90% (Vishwanath et al. 1986; Rao 1990). The disease affects the crop at all growth stages, and neck and panicle blast are the most destructive form of the disease (Pande et al. 1995; Takan et al. 2004; Takan et al. 2012). The most susceptible stage for leaf blast is seedling stage, whereas for neck and finger blast is pre-flowering stage. Growing cultivars with durable resistance is the best means of combating the blast disease of finger millet. Resistance is often assessed at the seedling stage, which did not correlate well with neck and finger infection. Hence, neck and finger blast are more destructive than leaf blast were considered important parameters for blast resistance (Nagaraja et al. 2007). Resistance in finger millet to M. grisea is often evaluated in the field under natural infection (Somashekhara et al. 1991; Takan et al. 2004; Mgonja et al. 2007; Nagaraja et al. 2007 & 2010) and no systematic artificial inoculation was made. Screening under natural infection condition may provide escapes and the true resistance may not be identified (Thakur et al. 2009). The neck and finger blast are routinely assessed at the dough stage of the crop as per cent disease incidence *i.e.*, number of plant infected (incidence does not differentiates levels of susceptibility – number and size of lesions across test lines).

Finger millet core collection (about 10% of the total germplasm collection) consists of 622 accessions representing geographical regions and biological races from the entire germplasm collection available at ICRISAT (Upadhyaya et al. 2006). Further, a mini-core (10% of core and 1% entire collection) was developed (Upadhyaya and Ortiz 2001) to facilitate easy evaluation of germplasm accessions for various traits, including disease resistance. This mini-core evaluation approach has successfully been used to identify resistance sources for various diseases (Pande et al. 2006; Sharma et al. 2010 & 2012; Sharma et al. 2012), drought (Kashiwagi et al. 2005; Krishnamurthy et al. 2010) and salinity (Vadez et al. 2007). Similarly, a finger millet mini-core consisting of 80 accessions (representing genetic diversity of the core collection and entire collection) developed at ICRISAT (Upadhyaya et al. 2010) was used in this study. The prime objective of this study were to develop a comprehensive disease severity assessment (rating scales) for leaf, neck and finger blast based on the qualitative and quantitative differences of lesions observed on finger millet plants infected with M. grisea and to evaluate the mini-core collection of finger millet in field and greenhouse by artificial inoculation in order to identify accessions having resistance to blast disease that could be utilized in resistance breeding programs.

#### Materials and methods

Seed source

Seed of the 80 germplasm accessions of the finger millet mini-core comprising of 4 cultivated races – *vulgaris* (53), *plana* (13), *elongata* (6) *compacta* (7) and one wild race *africana* (1), and sub races – *liliacea* (5), *stellata* (7), *incurvata* (23) and *digitata* (18) in race *vulgaris*; *seriata* (2), *confundere* (9) and *grandigluma* (2) in race *plana*; *laxa* (1), *reclusa* (4), and *sparsa* (1) in race *elongata* was obtained from Genebank, ICRISAT, Patancheru, India. The race *compacta* has no subraces (Prasada Rao and deWet 1997).

The finger millet mini-core was developed from a core collection of 622 accessions (<u>www.icrisat.org</u>). The core collection was evaluated for 5 qualitative and 15 quantitative traits at five agroecologically diverse locations in India. The hierarchical cluster analysis of data using phenotypic distances resulted in 40 clusters. A mini-core collection of 80 accessions (sampling 1 to 4 accessions from 40 clusters) was formed using the sampling strategy of 10% or a minimum of one accession from each cluster (Upadhyaya et al. 2010).

# **Pathogen and Inoculum preparation**

Inoculum was prepared from a single-spore representative culture of *M. grisea* isolated from blast infected samples collected from the finger millet fields during the rainy season 2008 at ICRISAT, Patancheru, India. Mass multiplication of fungal spores for inoculation was achieved by growing the fungus (9 discs/plate) on oat meal agar (OMA) medium at  $26\pm1^{\circ}$ C for 10 days. Spores were harvested by flooding the plates with sterilized distilled water and scrapping the growth by a spatula. The spore suspension was adjusted to desired concentration  $(1\times10^5 \text{ spores/ml})$  with the help of hemocytometer and a drop of a surfactant (Tween 20) was

added to ensure the uniform dispersal of spores. The suspension was used for field and greenhouse inoculations.

# **Field screening**

The 80 finger millet mini-core accessions along with checks (VR 708, VL 149, RAU 8 and PR 202) were evaluated in the finger millet blast nursery during the 2009 and 2010 rainy seasons in field by artificial inoculation at ICRISAT, Patancheru, India. Each accession was grown in two rows of 2 m length with row-to-row spacing of 60 cm and plant-to-plant spacing within the row of 10 cm with two replications in a randomized complete block design (RCBD). In order to increase the disease pressure, the susceptible check variety (VR 708) was planted on every 5<sup>th</sup> row alternatively. Plants were thinned to 20 plants/row at 15 days after emergence. Thirty day-old-seedlings were inoculated by spraying the inoculum (1×10<sup>5</sup> spores/ml) on the foliage using a Knapsack power sprayer during the evening hours. High humidity (>90% relative humidity) and leaf wetness was provided by sprinkler irrigation twice a day for 30 min each between 10:00 a.m. and 12:00 noon, and 4:00 p.m. and 6:00 p.m. to promote disease development. For neck and finger blast, plants were spray-inoculated at pre-flowering stage with an aqueous conidial suspension and procedure for providing favourable conditions as explained above up to physiological maturity.

#### **Disease severity assessment**

Leaf blast severity: Infected leaves exhibiting uniform lesion types were photographed to make color plates to aid in the classification of disease development. The leaf blast severity

was recorded at 10 days after inoculation (DAI) using a progressive 1 to 9 scale, where 1 = no lesions to small brown specks of pinhead size (0.1-1.0 mm), less than 1% leaf area affected; 2 = typical blast lesions covering 1-5% leaf area covered with lesions; 3 = 6-10%, 4 = 11-20%, 5 = 21-30%, 6 = 31-40%, 7 = 41-50%, 8 = 51-75% and many leaves dead; and 9 = typical blast lesions covering >75% leaf area or all the leaves dead (Fig. 1).

**Neck blast severity:** Based on the relative lesion size on the neck a 1 to 5 progressive rating scale was developed where, 1 = no lesions to pin head size of lesions on the neck region, 2 = 0.1 to 2.0 cm size of typical blast lesion on the neck region, 3 = 2.1 to 4.0 cm, 4 = 4.1 to 6.0 cm, and 5 = >6.0 cm size of typical blast lesion on the neck region (Fig. 2). Data were recorded in field at the physiological maturity on 10 randomly selected individual plants of each accession.

**Finger blast severity:** The finger blast severity estimate was recorded as visual percentage of blasted florets across all tillers of a plant (Fig. 3) on the same 10 randomly selected plants that were earlier rated for the neck blast severity in each row.

**Agronomic traits:** Data were also recorded for agronomic traits, such as days to flowering (DF) (time of full panicle emergence in 50% of the plants in a row), plant height (measured from the base of the plant to the tip of the panicle at maturity), and spike type (compactness of the panicle at maturity *i.e.* top curved, incurved and long open) during 2010 by following the finger millet descriptor (IBPGR 1985).

# **Greenhouse screening**

Resistance to leaf blast in mini-core germplasm accessions along with checks was confirmed under greenhouse screening. The seedlings were raised in 15-cm diameter plastic pots (10 seedlings/pot) filled with sterilized soil-sand-farm yard manure mix (2:1:1 by volume) in a greenhouse bay maintained at  $28\pm2^{\circ}$ C. The 12 day-old seedlings were inoculated by using an atomizer to spray the conidial suspension on the leaves of finger millet plants. The conidial inoculum was applied just until the beginning of runoff from the foliage. Inoculated plants were placed in a moist chamber at  $23\pm1^{\circ}$ C. After inoculation for 48 h in the moist chamber, the plastic pots containing 10 inoculated finger millet plants, were transferred to a greenhouse bay and exposed to high humidity (>90% RH) under misting for 10 days. Ten seedlings of each accession were tested in three replications (10 seedlings/pot) in a completely randomized design (CRD) and repeated twice to confirm the results. Leaf blast severity was recorded for 10 days after inoculation.

#### Data analysis

Data on leaf, neck and finger blast severity were recorded from each plot in the field experiments during 2009 and 2010. Statistical analysis was performed following the Residual Maximum Likelihood (REML) on GENSTAT statistical package (edition 14.0; Rothamsted Experiment Station, Herpenden, Herts AL52JQ, UK) for both years separately and on the combined data. For combining data across 2 years, Bartlett's test for homogeneity of error variance was done, which indicated that the error variances were homogeneous. Here, the year (environment) were considered as a fixed and genotype as random effect. Variance

components due to genotype ( $\sigma_g^2$ ), genotype × environment (years) interaction ( $\sigma_{ge}^2$ ) and their standard errors were determined. The significance of the years (environment) was assessed using the Wald statistic (Wald 1943) that asymptotically follows a  $\chi^2$  distribution. Environment-wise best linear unbiased predictors (BLUPs) for the leaf, neck and finger blast severity of mini-core accessions were calculated. The associations between pairs of variables such as – leaf, neck and finger blast, plant height, days to flowering and spike type were determined in terms of Pearson's correlation coefficients using the PROC CORR procedure in SAS (SAS Institute Inc. 2008. SAS/ STAT® 9.2 User's Guide. Cary, NC).

# Results

# **Development of new rating scales**

For assessing leaf blast severity, a 1-9 rating scale was developed based on per cent leaf area covered with lesions (Fig. 1). The 1-to-9 rating scale was classified into four general categories of resistant (R) (1.0-3.0 score, Fig. 1-1 to 1-3), moderately resistant (MR) (3.1-5.0 score, Fig. 1-4 and -5), susceptible (S) (5.1-7.0 score, Fig. 1-6 and -7) and highly susceptible (HS) (>7.0 score, Fig. 1-8 and -9). This rating scale was used both in field and greenhouse screens. For assessing neck blast severity, a 1-5 rating scale was developed based on the lesion size on the neck region just below the fingers (Fig. 2). Although the size of lesions on neck region varied almost continuously, four general classes (1.0-2.0 = resistant; 2.1-3.0 = moderately resistant; 3.1-4.0 = susceptible and 4.1-5.0 = highly susceptible) were distinguished based on the relative size of lesions (Figs. 2.1-2.5). The finger blast severity

percentage was classified into resistant (1.0-10%), moderately resistant (10.1-20%), susceptible (20.1-30%) and highly susceptible (>30%) (Fig. 3).

#### **Field resistance to blast**

Of the 80 finger millet mini-core germplasm collection evaluated for blast resistance (leaf, neck and finger) in field during the rainy seasons 2009 and 2010, blast severity was recorded on 78 accessions in 2009 (two accessions could not get established) and on all accessions in 2010. The REML analysis indicated significant (P<0.001) variation among the accessions for leaf, neck and finger blast reactions in both the environments (2009 and 2010 seasons) separately as well as in the pooled data (Table 1), indicating high variation among the genotypes for blast resistance. However, in the pooled analysis, variance component due to genotype ( $\sigma_g^2$ ) was non-significant for leaf blast, but highly significant for neck and finger blast. Although there was significant interaction between accession and years, the variance components due to genotypes ( $\sigma_g^2$ ) was very high compared to G×E interaction ( $\sigma_g^2$ ) for neck and finger blast, indicating that differences in the blast severity were mainly contributed by the accessions (Table 1).

Based on mean leaf blast severity for the two experiments (2009 & 2010), 78 accessions were found resistant (score 1.0-3.0 on 1-9 scale) and 2 moderately resistant (score 3.1-5.0). Based on mean neck blast severity, 68 accessions were resistant (score 1.0-2.0 on a 1-5 scale), 6 moderately resistant (score 2.1-3.0), 2 susceptible (score 3.1-4.0) and 4 highly susceptible (score >4.0) compared to 4.6 and 4.4 score in susceptible checks VR 708 and VL

149, and 1.5 and 2.0 in resistant checks PR 202 and RAU 8, respectively. Sixty-nine accessions were resistant (finger blast severity 1-10%), 4 moderately resistant (10.1-20%), 2 susceptible (20.1-30%) and 5 highly susceptible (>30%) to finger blast compared to 76.8 and 40.7% severity in susceptible checks VR 708 and VL 149, and 7.0 and 9.6% in resistant checks PR 202 and RAU 8, respectively (Table 2).

Out of eight mini-core accessions, sixty-six were found to have combined resistance to all three phases of blast (leaf, neck and finger) in both the years and data of those accessions along with checks are presented in Table 2. These resistant accessions belong to four cultivated races of finger millet, *vulgaris* (43 out of 53), *plana* (10 out of 13), *compacta* (7), *elongata* (5 out of 6) and wild race *africana* (1) (Table 2). The finger millet mini-core accessions originated from 13 countries, which is an ideal pool of geographical diversity of resistance sources. Of the sixty six resistant mini-core accessions, 21 accessions originated from Zimbabwe, 11 from India, 10 from Uganda, each 5 from Kenya and Nepal, 4 and 3 from Malawi and Zambia, respectively, and one each from Burundi, Germany, Nigeria, Senegal and United States of America (USA) and 2 were of unknown origin (Table 2).

## Correlation between disease severity and agronomic traits

A significant moderate correlation was observed between leaf blast with neck (r = 0.25, P < 0.001) and finger blast (r = 0.30, P < 0.001) whereas, neck and finger blast ratings had a high correlation (r = 0.92, P < 0.001) (Table 2; Fig. 4). The blast resistant accessions exhibited wide diversity for agronomical traits such as days to 50% flowering (DF), plant height and spike

type. The significant differences were observed for DF and plant height between the mini-core accessions. The DF ranged between 45 to 92 days with the mean of 70.2 days and plant height ranged between 70 to 137 cm (mean 105.8 cm). Diversity for spike type such as top curved (TC), incurved (I) and long open (LO) was also observed in mini-core accessions (Table 2). Leaf, neck and/or finger blast severity was negatively correlated with plant height (r = -0.21, -0.26 and/or -0.27) and DF (r = -0.19, -0.55 and/or -0.57) whereas, it was positively correlated with spike type (r = 0.17, 0.06 and/or 0.07).

#### **Confirmation of leaf blast resistance under greenhouse conditions**

Leaf blast severity scores of 80 accessions in greenhouse screen showed that 58 accessions as resistant, 20 moderately resistant and 2 susceptible compared with 8.0 and 7.1 score on susceptible checks VR 708 and VL 149, and 3.0 and 2.4 on resistant checks PR 202 and RAU 8, respectively (Table 2). Significant and moderate correlation (r = 0.44, *P*<0.0001) was found between greenhouse and field screening for leaf blast.

# Discussion

In this study, the rating scales were developed or refined for accurately assessing the leaf, neck and finger blast under field and greenhouse conditions. Leaf blast severity in finger millet has been routinely assessed using the rice blast rating scale (Takan et al. 2004; Mgonja et al. 2007; Nagaraja et al. 2007 & 2010; Takan et al. 2012). A literature search revealed that there is no specific rating system to finger millet for recording the leaf blast severity. Thus, we developed a new 1-9-class leaf blast rating scale and classified these based on per cent leaf area diseased, which is illustrated by colour plates (Fig. 1). The neck and finger blast are routinely assessed at the dough stage of the crop as disease incidence *i.e.*, percentage of ears showing infection on the neck and fingers over total number of neck and fingers in a row (Somashekhara et al. 1991; Mgonja et al. 2007; Nagaraja et al. 2007 & 2010). This type of scoring does not provide the extent of infection and the damage caused by the pathogen in the lines being evaluated. To overcome this limitation, we developed a new rating scale (1-5 scale for neck blast and per cent finger blast severity) for recording the neck and finger blast severity (Figs. 2 & 3). The use of this scale will facilitate estimation of exact damage caused by neck and finger blast phases of the disease. The given lesion dimensions in neck blast rating scale were not intended to be a rigid or fixed criterion upon which the rating scale are classified. Rather, they were given as supplemental information for describing and separating infection types and for general comparison by other researchers. We never intended, nor do we ever expect, any user of this scale to actually measure lesions for assessment of neck blast infection responses. It simply would not be practical to measure individual lesions for the routine evaluation of finger millet accessions for resistance to neck blast. Instead, one can use the color plates as a guide to quickly and easily estimate the relative size of lesions present on finger millet lines as needed for the classification of the neck blast. The newly developed rating scale for recording neck and finger blast adequately provide the actual disease severity on an individual plant and not merely the per cent disease incidence. We consider this as a significant step towards simplifying and improving the precision of disease scoring for identification and utilization of resistance sources.

The significant effect of year, as detected by Wald statistics that occurred in leaf, neck and finger blast infection levels between two years of experiment could be due to variable weather conditions. Such differences in weather conditions between two years could influence disease level is a known fact (Koutroubas et al. 2009). Environmental conditions, especially relative humidity and temperature could strongly affect the sporulation, release and germination of blast conidia (Ou 1985). In this study, a highly significant and strong positive correlation (r = 0.93) for neck and finger blast severity found between 2009 and 2010 suggest that the significant year effect didn't cause much impact on disease severity and reaction. Under favorable conditions, foliar blast occurred in a majority of accessions at the seedling stage, which did not correlate well with crop growth stages and maturity of the plants, probably because of the buildup of adult plant resistance. Hence, neck and finger blast reactions that are more destructive than leaf blast were considered important parameters for blast resistance (Takan 2004; Nagaraja et al. 2007; Nagaraja et al. 2010)

The negative correlations between blast severity and plant height indicates that tall and late maturing accessions might escape blast infection due to less favorable microclimatic conditions (Thakur et al. 2009). The role of plant height in rice blast resistance is well documented (Torres and Teng 1993; Koutroubas et al. 2009). A significant negative correlation was found between blast severity and days to flowering (DF) suggesting that early flowering accessions are more susceptible than the late ones (Pande et al. 1995; Mgonja et al. 2007). For instance, the accessions (IE 501, -3104, -4734, -5870 and -6082) that were earliest to flower (mean 54.2 days and range 48 to 62 days) recorded highest neck blast severity (score 3.5 to 4.9 on a 1 to 5 scale and mean 4.5) and 30 to 55% finger blast severity with a mean of 46% compared to highly resistant accessions with DF ranging from 60 to 92 (mean 72.5 days) (Table 2). In this study, positive association between spike type (top curved, incurved and long open) and blast severity was found. However, accessions with dark coloured seeds and compact heads have been reported to be more resistant compared to white seeded and open headed varieties (Takan et al. 2004).

Significant moderate correlations between leaf blast with neck and, finger blast suggests that a high level of leaf blast severity achieved by early inoculation may not result in severe neck or finger blast during the later stages of plant development. Poor correlation has been observed for leaf blast with neck blast (r = 0.04) and finger blast (r = 0.27) infection in finger millet (Somashekhara et al. 1991). It has been reported that seedlings of finger millet are more susceptible to leaf blast than mature plant (Rachie and Peters 1977). However, no relationship is known between the intensity of seedling infection and that of later neck and finger infection. Rather, prevailing weather conditions at a particular stage of crop development determine the intensity of blast infection (Esele 2002). Contrasting responses between the vegetative stage and reproductive stage often occur, indicating differential gene expression for resistance to leaf, neck and/or finger blast infection. The gene(s) responsible for

leaf blast resistance have not been found effective at reproductive stage in rice (Wu et al. 2004) indicate that researchers should not rely solely on the seedling reaction for assessing potential adult plant resistance. In contrast, a significant strong positive correlation found between neck and finger blast severity suggested that recording the blast severity using these two severity scales provided realistic data under field conditions at the right stage of the crop *i.e.* physiological maturity and also possible ability of the same gene(s) to induce resistance to both neck and finger blast. Significant positive correlation between neck and finger blast.

Developing improved blast resistant varieties is an important breeding goal in finger millet improvement (Mgonja et al. 2007). In this study, we refined the field screening technique for finger millet blast whereby germplasm can be effectively screened in the field and resistance confirmed through greenhouse screening. Large scale screening at the seedling stage for leaf blast resistance could be more economical and rapid in greenhouse than in the field. The inoculated plants in the seedlings stage were observed till the dough stage for neck and finger blast under greenhouse conditions. However, no neck and finger infection was observed in this experiment suggesting that prevailing weather conditions and availability of pathogen inoculum at a particular stage of crop development determine the intensity of blast infection (Esele 2002).

Among the resistant accessions, nine (IE 1055, -2821, -2872, -4121, -4491, -4570, -5066, -5091, and -5537) had desirable agronomic traits, such as early flowering (<65 days),

medium plant height (105-125 cm), semi-compact to compact inflorescence. These would be desirable sources of resistance for a finger millet breeding program. Of the resistant accessions, IE 3392 is a rich source of iron (Fe), IE 2957 of calcium (Ca) and IE 6537 of Ca and protein (Upadhyaya et al. 2011). Analysis and exploitation of such accessions would be an important and logical step towards breeding varieties with combined traits of high grain nutrient density, blast resistance and desirable agronomic traits. The information on clusters to which particular accessions with traits of interest belong will assist in looking extensively for more accessions with similar traits in the larger subsets, core collection and, eventually entire collection. For example, the mini-core accession IE 4709, only africana type finger millet found highly resistant to blast, agronomically desirable characters (Upadhyaya et al. 2007) and also been reported for rich source of grains nutrients such as Fe, Ca, Zinc (Zn) and protein contents (Upadhyaya et al. 2011). This type of accession could be involved in hybridization with agronomically superior accessions/breeding lines to combine blast resistance, grain nutrients and farmer, and consumer preferred traits. Similarly, IE 1012 was referred by Gowda et al. (1986) as an African cultivar exploited in India as a source of blast resistance. Introduction of micronutrient and protein dense blast resistant finger millet cultivars would help to reduce the losses caused by blast disease and more importantly malnutrition due to micronutrients and protein deficiency of resource-poor people, who consume finger milletbased diet in large quantities on a daily basis. It would be useful to evaluate the remaining africana type accessions from the core and entire collection that were not included in the minicore collection to identify additional sources of resistance to blast and grain nutrients, and test their resistance stability through multilocation testing in India and elsewhere. The disease scales illustrated in Figures 1, 2 and 3 may be useful criterion to evaluate a large number of finger millet germplasm for leaf, neck and finger blast. Identification of disease resistant accessions from the finger millet mini-core would permit use of diverse resistance sources for future breeding efforts and to ensure a better chance of success in finger millet improvement in developing cultivars with a broad genetic base.

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### **Figure captions**

Fig. 1. Disease rating scale for leaf blast of finger millet seedlings infected with *M. grisea*. Disease rating scales are based on the per cent leaf area covered with blast lesions observed on 10 days after inoculation under field and greenhouse conditions. The 9-class disease rating scale is classified into four general categories of resistant (1.0-3.0), moderately resistant (3.1-5.0), susceptible (5.1-7.0) and highly susceptible (7.1-9.0) responses. Class 1 is the immune response in which no lesions were observed, class 2 consists of typical lesions covering 1-5% of leaf area, class 3 consists 6-10%, class 4 consists of 11-20%, class 5 consists of 21-30% class 6 consists of 31-40%, class 7 consists of 41-50%, class 8 consists of 51-75% and class 9 consists of typical blast lesions covering >75% of leaf area or all the leaves dead. The leaves depicted in the plates are 1.0x their original sizes.



Fig. 2. A 1–5 rating scale based on lesion size on neck region for recording neck blast severity in finger millet infected with *M. grisea*. Neck blast rating scale based on relative size of lesions on the neck region under field conditions at the physiological maturity on 10 randomly selected individual plants of each accession. The 1-to-5 rating scale was classified into four infection types (*i.e.* 1.0-2.0 = resistant; 2.1-3.0 = moderately resistant; 3.1-4.0 = susceptible and 4.1-5.0 = highly susceptible). Class 1 = no lesions to pin head size of blast lesions on the neck region, class 2 = 0.1 to 2.0 cm size of typical blast lesion on the neck region, class 3 = 2.1 to 4.0 cm, class 4 = 4.1 to 6.0 cm and class 5 = >6.0 cm size of typical blast lesion on the neck region.



Fig. 3. The per cent finger blast severity for finger millet plants infected with *M. grisea*. (a). Out of 7 fingers, only half of the portion of finger is infected so, the per cent finger blast severity is 7% (b). Photograph showing more than 90% finger blast severity.



Fig. 4. Relationship between neck (NBS on 1 to 5 scale) and finger blast severity (FBS - %) of finger millet mini-core evaluated for blast resistance under field conditions during the rainy season 2009 & 2010 at ICRISAT, Patancheru, India.



Table 1. Estimates of variance due to genotype ( $\sigma^2 g$ ), genotype × environment ( $\sigma^2 g \times e$ ), and error ( $\sigma^2 e$ ) and their standard errors for leaf, neck and finger blast in the finger millet mini-core collection during 2009 and 2010 rainy seasons, ICRISAT, Patancheru, India.

Disease	Season	σ <sup>2</sup> g	SP	σ <sup>2</sup> σ×e	Se	σ <sup>2</sup> e	Se	Wald	Fnroh
Discuse	Scuson		5.00	o gre	5.0		5.00	(Season)	1 prov
Leaf	2009	0.094	0.031			0.161	0.026		
blast	2010	0.895	0.161			0.249	0.040		
	Pooled	0.034	0.067	0.526	0.101	0.203	0.023	11.07	0.002
Neck	2009	0.798	0.013			0.098	0.016		
blast	2010	0.987	0.161			0.101	0.016		
	Pooled	0.850	0.101	0.035	0.015	0.100	0.011	61.00	< 0.001
Finger	2009	180.354	28.827			7.591	1.240		
blast	2010	141.282	22.516			7.157	1.139		
	Pooled	155.371	24.790	4.593	1.372	7.339	0.834	14.99	< 0.001

IE No.			Leaf blast sev	-9 scale	e) <sup>ab</sup>	Neck bla	ty (1-5 scale) <sup>c</sup>	Finger	blast sev	verity (%) <sup>d</sup>	Agronomic traits <sup>e</sup>				
	Origin	Race	Greenhouse	2009	2010	Pooled	2009	2010	Pooled	scale) <sup>c</sup> Finger blast severity $(\%)^d$ ed20092010Pooled4.964.047.055.53.018.523.521.01.76.63.44.91.37.61.24.32.09.19.09.01.94.212.48.31.51.76.84.21.65.15.65.31.32.22.42.22.47.05.56.31.52.70.21.31.0-1.01.01.24.20.42.21.33.74.64.11.52.23.42.71.12.70.41.5	DF	Height (cm)	Spike type		
501	India	Vulgaris	3.2	1.5	2.0	1.8	4.8	5	4.9	64.0	47.0	55.5	48	81	TC
518	India	Vulgaris	3.6	2.0	2.5	2.3	2.8	3.2	3.0	18.5	23.5	21.0	60	110.8	TC
1055	Unknown	Vulgaris	2.1	1.5	1.6	1.5	2.1	1.3	1.7	6.6	3.4	4.9	62	121.2	TC
2034	India	Vulgaris	4.1	1.5	2.4	2.0	1.5	1.0	1.3	7.6	1.2	4.3	89	85	TC
2042	India	Vulgaris	3.5	1.8	2.0	2.0	2.1	1.9	2.0	9.1	9.0	9.0	60	105	TC
2217	India	Vulgaris	2.8	1.5	2.0	1.7	2.1	1.7	1.9	4.2	12.4	8.3	64	90	TC
2296	India	Vulgaris	2.9	1.5	1.6	1.5	1.9	1.1	1.5	1.7	6.8	4.2	72	93	TC
2312	India	Elongata	3.9	1.5	2.0	1.7	1.9	1.2	1.6	5.1	5.6	5.3	82	110	LO
2430	Kenya	Vulgaris	3.2	1.2	2.0	1.5	1.6	1.1	1.3	2.2	2.4	2.2	74	130	TC
2437	Kenya	Plana	2.0	1.0	2.0	1.5	2.6	2.2	2.4	7.0	5.5	6.3	75	123	TC
2457	Kenya	Compacta	1.1	1.5	2.0	1.7	2.0	1.1	1.5	2.7	0.2	1.3	78	115	TC
2572 <sup>f</sup>	Kenya	Plana	2.0	-	3.0	3.0	-	1.0	1.0	-	1.0	1.0	92	83	LO
2589	USA	Plana	2.0	1.2	2.0	1.5	1.4	1.0	1.2	4.2	0.4	2.2	71	137	TC
2606	Malawi	Vulgaris	3.9	1.5	2.0	1.7	1.6	1.0	1.3	3.7	4.6	4.1	80	95	TC
2619	Malawi	Vulgaris	2.7	1.2	2.0	1.5	1.7	1.3	1.5	2.2	3.4	2.7	82	105	TC
2710	Malawi	Plana	1.6	1.2	2.0	1.5	1.2	1.0	1.1	2.7	0.4	1.5	78	110	Ι
2790	Malawi	Elongata	2.0	1.2	1.6	1.3	1.8	1.0	1.4	5.1	3.8	4.4	79	70	LO
2821	Nepal	Compacta	2.1	1.2	2.4	1.7	1.1	1.0	1.0	2.2	3.6	2.8	62	113	Ι
2871	Zambia	Compacta	1.1	1.5	1.6	1.5	1.4	1.0	1.2	0.7	0.2	0.4	83	106	Ι
2872	Zambia	Vulgaris	1.0	1.2	1.6	1.3	1.5	1.0	1.3	3.7	1.4	2.5	60	103	TC
2911	Zambia	Vulgaris	1.0	1.5	1.1	1.3	1.1	1.0	1.0	1.2	0.2	0.6	83	129	TC
2957	Germany	Vulgaris	1.9	1.5	2.0	1.7	2.0	1.0	1.5	1.8	0.9	1.2	70	91	TC
3045	India	Vulgaris	2.1	1.5	2.0	1.7	1.8	1.6	1.7	7.2	7.5	7.3	62	114	LO

 Table 2. Leaf, neck and finger blast severity, origin, race type and variable agro-morphological characters of finger millet mini-core collection

3077	India	Vulgaris	3.2	1.2	2.0	1.5	1.3	1.2	1.2	5.2	5.3	5.2	62	104.2	TC
3104	India	Vulgaris	2.3	1.5	2.0	1.8	3.8	5.0	4.4	48.0	45.5	46.8	55	86	Ι
3317	Zimbabwe	Vulgaris	2.6	1.5	1.1	1.3	1.6	1.3	1.4	3.7	1.4	2.5	76	110	TC
3391	Zimbabwe	Vulgaris	3.2	1.5	1.6	1.5	1.2	1.0	1.1	2.7	0.4	1.5	79	122	TC
3392	Zimbabwe	Compacta	2.0	1.2	2.0	1.5	1.8	1.0	1.4	3.7	1.4	2.5	66	95	TC
3470	India	Vulgaris	3.7	1.5	1.6	1.5	1.4	1.3	1.3	6.2	6.0	6.0	62	109	TC
3475	India	Vulgaris	4.7	1.2	2.4	1.7	1.6	1.4	1.5	3.7	5.1	4.3	74	101	Ι
3614	Unknown	Plana	2.1	1.5	2.0	1.7	1.4	1.0	1.2	3.7	0.2	1.8	75	123	TC
3721	Uganda	Compacta	2.2	1.2	2.0	1.5	1.9	1.0	1.5	0.3	0.9	0.5	83	134	TC
3945	Uganda	Plana	2.0	1.2	2.0	1.5	1.8	1.0	1.4	1.8	2.1	1.8	79	123.2	TC
3952	Uganda	Plana	1.1	1.5	1.6	1.5	1.6	1.2	1.4	0.3	0.9	0.5	79	77	TC
3973	Uganda	Vulgaris	1.7	1.5	1.6	1.5	2.0	1.3	1.6	2.3	1.2	1.6	76	85	TC
4028	Uganda	Vulgaris	2.9	1.2	1.6	1.3	1.4	1.1	1.3	4.7	3.8	4.2	74	130	TC
4057	Uganda	Plana	3.2	1.5	2.4	2.0	1.0	1.1	1.0	1.3	1.4	1.2	70	105	TC
4073	Uganda	Elongata	1.5	1.7	1.6	1.8	1.1	1.0	1.1	0.6	0.2	0.3	79	96	TC
4121	Uganda	Plana	1.9	1.2	1.6	1.3	1.7	1.5	1.6	7.2	5.6	6.3	62	114	TC
4329	Zimbabwe	Vulgaris	3.2	1.5	1.6	1.5	1.2	1.1	1.1	2.7	0.7	1.6	74	114	TC
4491	Zimbabwe	Elongata	2.3	1.8	1.6	1.7	2.0	1.4	1.7	10.1	6.3	8.1	62	125	TC
4497	Zimbabwe	Vulgaris	2.5	1.2	1.6	1.3	1.8	1.4	1.6	5.7	5.8	5.7	70	110	TC
4545	Zimbabwe	Compacta	3.2	1.2	1.1	1.1	1.9	1.3	1.6	10.9	7.7	9.4	74	110	TC
4565	Zimbabwe	Elongata	2.2	1.7	2.0	2.0	1.3	1.0	1.2	4.5	0.7	2.6	73	125	TC
4570	Zimbabwe	Plana	1.5	1.2	1.1	1.1	1.2	1.2	1.1	2.1	2.1	2.1	62	118	TC
4622	Zimbabwe	Compacta	2.3	2.3	1.6	2.2	1.3	1.3	1.3	3.1	1.7	2.3	75	102	Ι
4646	Zimbabwe	Plana	3.6	1.4	1.1	1.3	1.7	1.2	1.4	4.0	5.1	4.5	79	93	TC
4671	India	Vulgaris	1.4	2.0	1.1	1.7	2.3	1.2	1.7	12.8	3.6	8.2	67	95	TC
4709	Burundi	Africana	1.2	1.9	1.9	2.2	1.3	1.6	1.4	0.2	0.4	0.1	45	85	LO
4734	India	Vulgaris	2.5	1.5	1.5	1.5	4.9	4.8	4.9	48.0	48.0	48.0	51	98	Ι
4757	India	Vulgaris	2.3	1.5	2.0	1.8	3.2	3.3	3.2	20.0	12.8	16.4	62	110.2	TC

4795	Zimbabwe	Vulgaris	2.2	1.7	1.6	1.7	2.1	1.1	1.6	9.9	8.0	9.0	73	105.2	TC
4797	Maldives	Vulgaris	6.6	1.5	6.5	4.0	2.8	1.9	2.3	31.0	21.8	26.4	74	90	TC
4816 <sup>f</sup>	India	Elongata	1.5	-	1.5	1.5	-	1.0	1.0	-	2.0	2.0	85	90	LO
5066	Senegal	Vulgaris	1.6	1.4	1.6	1.5	1.4	1.1	1.2	4.0	1.2	2.6	62	107	TC
5091	Zimbabwe	Vulgaris	2.0	1.2	2.0	1.5	1.9	1.4	1.7	8.9	2.9	5.9	62	110.2	TC
5106	Zimbabwe	Vulgaris	1.9	1.2	1.6	1.3	2.1	1.1	1.6	5.0	1.2	3.1	70	105	TC
5201	India	Vulgaris	1.4	1.4	2.0	1.7	1.7	1.0	1.4	1.6	7.7	4.7	80	109.8	LO
5306	Zimbabwe	Vulgaris	2.5	1.4	1.1	1.3	1.7	1.5	1.6	3.5	2.6	3.1	75	110	TC
5367	Kenya	Vulgaris	2.0	1.5	2.0	1.8	2.9	1.2	2.1	20.5	10.3	15.4	74	115	TC
5537	Nepal	Vulgaris	1.6	1.4	2.0	1.7	1.6	1.6	1.6	6.5	8.2	7.4	60	105	TC
5817	Nepal	Vulgaris	1.35	1.5	1.5	1.5	2.5	1.9	2.2	12.0	9.8	10.9	67	100	Ι
5870	Nepal	Vulgaris	3.1	1.5	4.0	2.8	3.8	3.4	3.6	32.0	28.5	30.3	55	100	Ι
6059	Nepal	Vulgaris	1.15	1.5	3.5	2.5	2.3	2.2	2.2	12.0	11.8	11.9	62	130	TC
6082	Nepal	Plana	5.2	1.5	6.5	4.0	4.9	5.0	5.0	60.0	43.0	51.5	62	105	Ι
6154	Nepal	Vulgaris	1.0	1.5	1.6	1.5	1.4	1.5	1.5	4.0	8.0	6.0	77	95.4	TC
6165	Nepal	Vulgaris	1.6	1.2	1.1	1.1	1.4	1.4	1.4	4.0	5.3	4.7	79	90	Ι
6221	Nepal	Vulgaris	1.0	1.2	1.6	1.3	1.6	1.3	1.5	4.5	4.1	4.3	74	100	Ι
6240	Zimbabwe	Vulgaris	1.3	1.2	2.0	1.5	1.3	1.2	1.3	2.6	3.6	3.1	71	115.2	TC
6294	Zimbabwe	Vulgaris	3.2	1.8	1.6	1.7	1.7	1.2	1.4	5.0	2.6	3.8	74	110	TC
6326	Zimbabwe	Vulgaris	3.0	1.2	1.1	1.1	1.6	1.2	1.4	5.5	0.2	2.8	75	95	TC
6337	Zimbabwe	Vulgaris	1.7	1.8	1.6	1.7	1.7	1.0	1.4	1.1	0.2	0.6	70	92	TC
6350	Zimbabwe	Vulgaris	1.9	1.8	1.6	1.7	1.4	1.0	1.2	4.5	1.2	2.8	79	95	TC
6421	Uganda	Vulgaris	1.0	1.2	2.0	1.5	2.0	1.1	1.5	9.4	4.6	7.0	70	110	TC
6473	Uganda	Plana	2.0	1.2	2.0	1.5	1.2	1.0	1.1	1.6	0.7	1.1	76	123	TC
6514	Zimbabwe	Vulgaris	2.1	1.5	2.0	1.7	1.8	1.0	1.4	10.9	4.6	7.8	76	115	TC
6537	Nigeria	Vulgaris	3.5	1.3	2.0	1.6	1.8	1.0	1.4	-0.2	0.2	0.2	78	95	TC
7018	Kenya	Vulgaris	1.4	1.8	2.0	2.0	1.8	1.0	1.4	6.5	0.2	3.3	70	121	TC
7079	Kenya	Vulgaris	3.5	1.8	2.0	2.0	1.4	1.2	1.3	5.5	3.1	4.3	62	115	TC

7320	Kenya	Vulgaris	4.5	1.2	1.6	1.3	1.2	1.1	1.1	4.0	3.1	3.6	62	128.2	TC
VR 708- Check	India	Vulgaris	8.0	1.2	4.7	2.8	4.7	4.4	4.6	70.1	83.6	76.8	55	90	TC
PR 202- Check	India	Vulgaris	3.0	1.8	2.0	2.0	1.7	1.3	1.5	8.4	5.6	7.0	62	110	TC
RAU 8- Check	India	Vulgaris	2.4	1.2	2.0	1.5	1.9	2.2	2.0	8.4	10.7	9.6	62	95	TC
VL 149- Check	India	Compacta	7.1	1.2	4.7	2.8	4.5	4.2	4.4	42.2	38.5	40.7	60	87.2	TC
LSD     (P<0.05)g	-	-	0.86	0.68	1.1	0.72	0.83	0.74	0.56	7.7	6.3	4.9	4.15	0.7	-

<sup>a</sup> Mean of two replications, 10 plants/replication at 40 DAS for leaf blast and at physiological maturity for neck and finger blast

<sup>b</sup> Leaf blast severity on 1 to 9 scale where 1 = no infection and 9 = >75% leaf area covered with lesions

<sup>c</sup> Neck blast severity on 1 to 5 scale where 1 = no infection and 5 = >6 cm lesions on the neck region

<sup>d</sup> Finger blast severity (%) across all panicles/all tillers in a row

<sup>e</sup> DF = days to flowering and Height = Plant height (cm), Spike type: TC = Top curved, I = Incurved, LO = Long open

<sup>f</sup> Two accessions could not get established in 2009

<sup>g</sup> Trial least significant difference.