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Pre-breeding for yield associated parameters and biotic stress resistance in finger millet (*Eleusine coracana* L.) under southern Chhattisgarh agroclimatic zone

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Abstract

The present experiment was conducted to portray the core collection of germplasm accessions; with special reference to trait specific classification, screening for blast disease and cross combination studies in finger millet (*Eleusine coracana* L.), during *Kharif* 2020-21 at Research cum Instructional Farm, S G College of Agriculture and Research Station, Jagdalpur, IGKV, Raipur, Chhattisgarh, India. Among the test accessions, characterization was done in respect to major yield related parameters and trait specific genotypes were short listed for future as donor parents for introgressive breeding for instance, genotypes GEC453, GEC53, GEC127I, C0477591, IC0477496, GEC5 andIC0477678 for high tillering capacity; GEC222, GEC322, GEC11, GEC92, IC0477556-X, GEC55, IC0476676, GEC79, IC0477304, IC0477620 for high numbers of finger per ear; GEC371, IC0476669-X, GEC223, GEC394, IC0477678 for finger length; and IC0477650, GEC69, IC0476495, GEC55, IC0476299, GEC254, GEC347, GEC470, IC0477507, GEC122 for test weight. In screening for all three phenotypic variants of blast disease GEC92, IC0477317, IC0477406, GEC147, GEC79 (for leaf blast) IC0477650, GEC41, GEC322 (For neck blast) and IC0476838, GEC132, GEC352, GEC106 (against finger blast) exhibited resistance, which need to verified in controlled environments. In cross compatibility studies with some specific parents F_1 had been harvested and selection will be made in segregating generations. Summarily, after achieving saturation in artificial selection and recombination breeding, research mode must be shifted towards widening of genetic base, traits specific breeding and development of variable need based cultivars with respect of quality and end produce to meet the growing population demands, raising insect pest pressure and changing climatic scenario.

Keywords: Finger millet, Prebreeding, Drought, Disease resistance, Adoption.

1. Introduction

Crop improvement is primarily focussed to develop genetically improved crop varieties for the economic benefit of farming community however, in present decade this is constantly challenged by population growth, diminishing arable land and global climate change,in terms of increasing the risk of crop production in terms of biotic and abiotic stress (Roos *et al.*, 2017; Frona *et al.*, 2019). Deployment of genetically improved crop cultivars

and better management practices are amongst the best strategies to increase food production and meet a projected doubling of food demand in the next 20 years (Miller *et al.*, 2010; Repinski *et al.*, 2011; Komarek and Masangi, 2019). Modern day crop improvement programme basically includes two activities, i.e., pre-breeding or germplasm enhancement and varietal development. These two steps are mutually interdependent and determining factors of the pace at which varieties are released timeously and constantly to farmers and equally determining factor for its sustainability against biotic and abiotic stresses (Sharma *et al.*, 2013). Prebreeding depends upon various factors namely breeding goals, genetics and agronomy of the crop, breeder's long-term objectives, availability of testing facilities and many others. Fleshing back to historical prospect, the Global Partnership Initiative for Plant Breeding Capacity Building (GIPB)/FAO and Biodiversity International use the term 'pre-breeding' to pronounce the various activities of plant breeding research that have to precede the stages involved in varietal development, testing and release (Biodiversity International and GIPB/FAO, 2008). Further, the Global Crop Diversity Trust defined pre-breeding as 'the art of identifying desired traits, and incorporation of these into modern breeding materials.' Pre-breeding is aimed to lower-down genetic uniformity among crop varieties by deploying the wider gene pool of available plant genetic resource while maintaining/enhancing the economic yield, resistance to pest and disease and other quality traits (Shimelis and Laing, 2012; Singh *et al.*, 2021).

Pre-breeding, the development of semi-finished products, provides a unique opportunity through introgression of desirable gene(s) from exotic germplasm into genetic backgrounds readily used by the breeders with minimum linkage drag (Sharma *et al.*, 2013b). The major activities of Prebreeding envisages characterization of land race populations, creation of new parent population, Introgression of new traits through recombination breeding and creation of novel traits (Upadhyaya *et al.*, 2014). Research carried out to date suggests that wild relatives have not only contributed genes for resistance to biotic stress, but also variations to yield and quality traits (Dwivedi *et al.*, 2008; Imai *et al.*, 2013). In finger millet (*Eleusine coracana* L) most of varietal improvement research were earlier restricted to artificial selection in local germplasm which were further progressed towards recombination and mutation breeding in current decade. The crop is gaining reliable picture due to suitability in water limited agriculture and also getting a permanent space in modern day diet chart in lieu of nutraceutical properties. But growing population demand, raising insect pest pressure and changing climatic scenario is asking the researchers to shift the momentum towards the widening of genetic base, traits specific breeding and development of variable need based cultivars with respect of quality and end produce.

2. Materials and methods

The present experiment was conducted to characterize the core collection of germplasm accessions, screening for blast disease and cross combination studies; for important yield related parameters; during *Kharif* 2020-21 at Research cum Instructional Farm, S G College of Agriculture and Research Station, Jagdalpur, IGKV, Raipur, Chhattisgarh, India.A set of 104germplasm were evaluated including four check varieties namely Indira Ragi 01, CG Ragi 02, GPU-28 and GPU-67 in Augmented Randomized Block Design. Each germplasm was represented by paired row plot of 3m length with inter and intra row spacing of 22.5cm and 10cm, respectively. The experiment was divided into 10 blocks each of which comprised of 10 test entries and four check varieties. Standard package of practice was followed to raise the healthy crop. The data was recorded as per DUS descriptors of PPV & FR guidelines for the traits namely days to 50 percent flowering (DAS), days to maturity (DAS), plant height (cm), numbers of tillers per plant (cm), finger length (cm), finger per ear, test weight (g), fodder weight per plant (g), grain weight per plant (g). The observations were recorded for each character's s per standard protocol of ICAR-AICRP Small Millets. The data collected for all quantitative characters were subjected to analysis of variance for augmented randomized block design. For crossing of male and female parents contact method was used by deploying hot water treatment at 40-45^{0}C. For evaluation of blast disease under field conditions protocols used as suggested byICAR-AICRP Small Millets.

3. Results and discussions

3.1 Characterization of plant genetic resource

Plant genetic resource or germplasm are often referred as 'cultivated native varieties' that are adapted to a specific

agro-ecological and farming system without a scientific form of selection. Germplasm is invariably heterogeneous and an excellent source of genetic variation for crop breeding programs (Sleper and Poehlman, 2006; Singh *et al*., 2021) andharbours useful genes such as genes for early maturity, yield potential, disease and pest resistance and other desired traits. Therefore, it is preliminary step to characterize this existing preliminary gene pool for various yield related traits. Based on DUS descriptors including farmers participatory selection genotypes were selected specific to trait (**Table 01**). In germplasm evaluation part average days to 50 percent flowering was found to be 79 DAS with a range of 50-93 DAS. Among the test accessions GEC411, IC0477325, IC0477890, IC0588007, GEC371 andGEC222 were selected for trait specific breeding as parent for improvement and breeding of mid durational varieties. Similarly for crop duration, genotypes IC0477043, IC0477650, GEC41, GEC453, GEC322, IC0477017, IC0477569, GEC11were short listed where the mean, maximum and minimum value was recorded to be 111 DAS, 126 DAS and 82 DAS respectively. Looking to significant correlation of canopy length towards grain and fodder yield and harvest index, ten genotypes were selected based on optimal range which can be used as parents in cultivar development phase for improving the trait. These were IC0588007, GEC11, GEC92, GEC55, IC0476299, IC0477602, IC0477047, GEC254, GEC79 andIC0477317. The length of plant canopy was ranged between 56-136cm while the mean length was recorded to be 82cm. Among another architectural trait i.e., tillers per plant exhibited wide variation among the existing plant genetic resource ranging from 1-5. Seven genotypes were selected which showed higher tillers count than mean values for future use namely GEC453, GEC53, GEC127I, C0477591, IC0477496, GEC5 andIC0477678. In relevance to ear three characters namely fingers count per plant, finger length and ear weight; were selected based on characters association and cause effect relationship reported by previous researchers. Among the germplasm accessions chosen for study an average number of five fingers were noted whereas minimum and maximum counts were three and ten respectively. Genotypes, GEC222, GEC322, GEC11, GEC92, IC0477556-X, GEC55, IC0476676, GEC79, IC0477304, IC0477620 were short listed for trait specific improvement in future breeding programmes. Finger length (cm) determines the number of seeds/grains harboured in a row, however larger finger with higher number of grains doesn't always leads to better yield due reduction in size and grain weight. Therefore, this trait is taken care in along with the test weight. In our studies the average length was recorded to be 6cm and genotype GEC371, IC0476669-X, GEC223, GEC394, IC0477678 were sorted out to improve the trait in other parental populations as prebreeding source. Ear weight, includes both unthreshed grain weight and base (on which the grains are placed/winded in circular manner) weight, exhibited a wide range of diversity and lied between 3.01 to 25.10g among test accessions. The arithmetic mean for the trait was 9.23g. The superior genotypes carefully chosen for these traits were GEC41, GEC322, GEC69, IC0476707, IC0476495, IC0477556-X. In due consideration of *per se* yield components, grain yield per plant (g), fodder yield per plant (g) and test weight (g) could be selected directly since these have been reported to possess high heritability coupled with higher genetic advance. For grain yield per plant (g) large amount of variation were recorded and when selection is to be made for the trait only, it is recommended to pick GEC371, IC0477650, GEC322, IC0476495, IC0476838, GEC132, GEC352, GEC137, IC0476864, GEC280, IC0477591 directly. Similarly, for fodder prospect genotypes GEC371, IC0477650, GEC41, GEC322, GEC274, IC0476676, GEC352, GEC106, GEC147, GEC79, IC0477317 can be chosen as parental line or entry in variety release programme. Test weight is one of the critical parameters in prebreeding of any crop species, since the bold seeded genotypes are always in preference for farming community due several reasons. Higher grain yield per unit area, lesser hulling and milling losses, better market price, reduced storage losses are some of the factors which makes the traits vary important. Here we obtained a series of genotypes having test weight right from 1.26g to 3.96g. Some accessions displayed appreciable performance regarding the parameters which were selected, so as to improve/incorporate in other cultivars, these were IC0477650, GEC69, IC0476495, GEC55, IC0476299, GEC254, GEC347, GEC470, IC0477507, GEC122. However, in most cases higher test weight is associated with reduction in number of seeds per unit area which levels the yield hike. Therefore, test weight must be taken together with grain yield when direct selection is planned and when deployed for prebreeding as donor view point can be considered individually.

Table 01. Traits specific cataloguing of germplasm accessions

S. No.	Traits	Mean	Range	Selected Germplasm Accessions
01.	Days to 50 percent flowering	79	50-93 days	GEC411, IC0477325, IC0477890, IC0588007, GEC371, GEC222
02.	Crop duration	111	82-126 days	IC0477043, IC0477650, GEC41, GEC453, GEC322, IC0477017, IC0477569, GEC11
03.	Plant canopy length	82	56-136cm	IC0588007, GEC11, GEC92, GEC55, IC0476299, IC0477602, IC0477047, GEC254, GEC79, IC0477317
04.	Tillers/plant	2	1-5	GEC453, GEC53, GEC127I, C0477591, IC0477496, GEC5, IC0477678
05.	Finger's count/plant	5	3-10 per ear	GEC222, GEC322, GEC11, GEC92, IC0477556-X, GEC55, IC0476676, GEC79, IC0477304, IC0477620
06.	Finger length	6cm	4-13cm	GEC371, IC0476669-X, GEC223, GEC394, IC0477678
07.	Ear weight	9.23g	3.01-25.10g	GEC41, GEC322, GEC69, IC0476707, IC0476495, IC0477556-X
08.	Grain yield per plant	11g	8-15 g	GEC371, IC0477650, GEC322, IC0476495, IC0476838, GEC132, GEC352, GEC137, IC0476864, GEC280, IC0477591
09.	Fodder yield per plant	32g	20-50g	GEC371, IC0477650, GEC41, GEC322, GEC274, IC0476676, GEC352, GEC106, GEC147, GEC79, IC0477317
10.	Test weight	2.23g	1.26-3.96g	IC0477650, GEC69, IC0476495, GEC55, IC0476299, GEC254, GEC347, GEC470, IC0477507, GEC122

3.2 Screening for blast disease resistance

A total of 100 germplasm accessions were screened for resistance against the major disease of finger millet. All three-stage specific phenotypic variant of the disease i.e., leaf blast, finger blast and neck blast were screen at respective stages with standardized protocol. The scale adopted was 0-5, where 0 represented no incidence and 5 represented more than 50 percent area of target plant part covered by the disease. Leaf blast is observed at seedling stage when crop is 35-40 days old and we could not find any genotype to be highly resistant and only five genotypes namely GEC92, IC0477317, IC0477406, GEC147 and GEC79exhibited resistance(**Table 02**). Additionally, 30 genotypes displayed moderate resistance, 35 genotypes moderately susceptible and other felled in susceptible category. However, we observed to re-establish most of genotypes at later growth stages which were heavily infested earlier. This might belong to recovering ability of plant metabolites to the concurrent and residual effect of fungal toxins. Later at the age of 70-80 days old plant observation were recorded for neck blast incidence.The disease incidence ranged from 17.8 to 66.0% among test populations. Three genotypes showed resistance against neck blast namely IC0477650, GEC41 and GEC322. 18 (Eighteen) genotypes were found to be moderately resistant, 12 genotypes Moderate susceptible, 50 genotypes susceptible and 20 genotypes highly susceptible.When plants began maturing, they were screened for finger blast disease where genotypes IC0476838, GEC132, GEC352, GEC106, GEC147, IC0476299, GEC254 were found to be resistant and the percentage of infection ranged from 19.7 % to 67.6 % compared to 98.8 % in check varieties (GPU 28, Indira Ragi-1, GPU 67, CG Ragi -2). Among the genotypes evaluated, moderate resistance was observed in case of seven genotypes *viz*., IC0476838, GEC132, GEC352, GEC106, GEC147, IC0476299andGEC254. Moderate susceptible 12 genotypes, 49 genotypes susceptible and 10 genotypes exhibited highly susceptible reaction against finger blast disease. In previous studies, Bal *et al.* (2020) screened eighteen genotypes under field conditions, out of which eight genotypes namely GPU 67, BR 14-3, L 352, KOPN 942, PR 202, VR 708, PR 10-35 and GPU 45 manifested similar reaction against finger blast and neck blast. Patro *et al.* (2018) also reported parallel score for all three kind of blast disease in his study with 25 genotypes. The objective of screening in available

breeding stock was to find out resistant source against the nationally important disease. The resistant genotypes so obtained can be deployed further in breeding programme and should be subject to revalidation under controlled conditions and/or molecular level (Babu *et al.*, 2013; Khadka *et al.,* 2013). Further susceptible genotype may be corrected by either back cross breeding of other approach, if high yielder and agronomically suitable.

Table 02. Field study against blast disease incidence

Information	Features
Activity Assigned	*Screening germplasm accession for leaf blast resistance*
Total lines screened	100
No of entries recorded resistance	05 (GEC92, IC0477317, IC0477406, GEC147, GEC79)
No of entries recorded moderate resistance	30
Final target of the breeder	Identification of resistant doner parents
Activity Assigned	*Screening germplasm accession for finger blast resistance*
Total lines screened	100
No of entries recorded resistance	03(IC0477650, GEC41, GEC322)
No of entries recorded moderate resistance	18
Final target of the breeder	Identification of resistant doner parents
Activity Assigned	*Screening germplasm accession for finger blast resistance*
Total lines screened	100
No of entries recorded resistance	07(IC0476838, GEC132, GEC352, GEC106, GEC147, IC0476299, GEC254)
No of entries recorded moderate resistance	12
Final target of the breeder	Identification of resistant doner parents

The resistant germplasm accessions will form a strong basis for future allele mining and full characterization of blast resistance in finger millet. Future studies will also need to characterize additional blast isolates in order to establish the rate of evolution of corresponding genes in the case of gene-for-gene interaction (Dida *et al.*, 2021). Similar studies have been undertaken in rice for various geographical regions (Fukuta *et al.*, 2019; Pagliaccia *et al.*, 2018; Zhang *et al.*, 2017) and will be important in finger millet to ensure relevant varieties are developed and deployed in strategic locations, which may have different blast pathotypes. In the current study, we used a blast isolate from the neck tissue but future studies will need to establish whether there are variations between blast isolates affecting various plant tissues. Differences in the aggressiveness of blast isolates from different tissues of the same plant have been reported (Ghatak*et al.*, 2013) in rice, and there is evidence suggesting tissueadapted fungal infection strategies (Marcel*et al.*, 2010). Similar studies in finger millet will aid in understanding any tissue specialization strategies by the pathogen.

3.3 Breeding potentials of selected crosses

Exploitation of genetic variability existing in the working germplasm is the first principle in the improvement of any crop (finger millet being no exception to this). Analysis and exploitation of existing genetic variability is a short-term strategy for developing improved cultivars for meeting immediate requirement of the farmers and the end-users(Laidig *et al.*, 2014; Smith *et al.*, 2014b). Exploitation of variability created by hybridization through recombination breeding is the major approach adopted in finger millet improvement programs (Byrne *et al.*, 2018). A total number of four crosses were attempted with an objective to incorporate some of the important traits of existing germplasm resource to the leading varieties. Parents GPU-28, GE-3678, BR-14-27, BR-31 were selected as female counterpart which are characterized by fair adoptability to local environmental conditions, earliness

in maturity and nonlodging type (**Table 03**). Similarly male parents were IR-01, BR-14-21, GE-10, BR-14 having features of High yielder, stuffy plant type and median type of maturity. By deploying the contact method of crossing with hot water treatment (temperature at 40-45^{0}C) crosses were made. Approximate 35-40 percent cross were failed might be due to nonsynchronous maturity of parental populations or variation in temperature. A total number of 130 populations have been selected and will be advanced in next generations. Crossing (hybridization) is purposefully employed in the breeding of domesticated plants to take advantage of transient hybrid vigor, move desirable variation among lineages, and generate novel phenotypes. The production of hybrid offspring generates the potential for gene flow between parent populations. If hybrids are fertile, they may backcross with either or both of the parents, resulting in introgression (Todesco *et al.*, 2016); however, introgression also may serve as an evolutionarily creative force by introducing new, possibly adaptive, genetic variation into a population (Goulet *et al.*, 2021; Singh *et al.*, 2021).

Table 03. Cross compatibility studies

Information		Features
Female Parents	Name of Female parents	GPU-28, GE-3678, BR-14-27, BR-31
	Features of female parents	Well adapted to local environment, earliness, Non-lodging
	Species	*Eleusine coracana* L
Male Parents	Name of Female parents	IR-01, BR-14-21, GE-10, BR-14
	Features of female parents	High yielder, stuffy plant type
	Species	*Eleusine coracana* L
No. of crosses attempted	04	
Success rate	60	
Generation advance, F_2, BC_1F_1, BC_2F_1 etc	130	
Final target of the breeder	High yielder recombinant, adopted to local agroecological conditions	

In current phase of crop breeding, there is a need for judicious use of plant genetic resources and conserve them for future need. Breeding dual purpose cereals and grain crops, breeding specifically for stress tolerance and degraded lands and finding new niches for fodder production are the keywords. Furthermore, these prebreeding strategies should be supplemented with appropriate breeding procedures, biochemical and physiological basis of improved production. The new techniques provided by biotechnology are relatively fast, resource efficient and highly specific. It may offer opportunities to increase sustainability and profitability visà-vis international competitiveness. Building up a strong genetic resource base of different forage crops will also require introduction, exploration and collection programmes from unexplored habitat.

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Author Contributions

PK, AK and BKD prepared the manuscript and PK, DS, VK and GV helped in preparing the final version of the manuscript and correspond to the journal.

Ethical Approval

This article does not contain any studies involving human or animal participants performed by any of the authors.

Conflicts of Interest:

The authors declare no conflict of interest.

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