

# Photosynthetic and yield traits identified through multivariate analysis in mungbean exhibiting tolerance to the combined stresses of low phosphorus and drought

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### Abstract

Experiments were conducted to study the genotypic variability for tolerance to combined stresses of low availability of phosphorus (P) and drought in 14 mungbean [Vigna radiata (L.) R. Wilczek] accessions. The accessions were evaluated under four conditions viz., control (sufficient P, irrigated), low P (without P, irrigated), drought (sufficient P, withholding irrigation) and combined stresses (low P, withholding irrigation). The relative stress tolerance was estimated for 22 agro-physiological traits. The principal component analysis (PCA) and relative stress indices (RSIs) of traits exhibited significant variation among the treatments and accessions. Based on RSIs, the PCA ranking analysis showed that the accessions IC 280489, PDM 139 and IC 76491 were highly ranked and tolerant to low P, drought and combined stresses. The relative increase in component traits such as photosynthetic parameters, relative water content, above-ground biomass, seed P content and number of pods plant<sup>-1</sup> were higher while canopy temperature and water use efficiency were reduced in tolerant accessions. In contrast, IPM 2-3 was found to be relatively sensitive to all three treatments. Tolerant accessions may be either included in the breeding program or used directly as cultivar that can be grown under low P and drought.

Key words: Drought, low phosphorus, mungbean, water use efficiency, photosynthesis, harvest index

### Introduction

Mungbean [*Vigna radiata* (L.) R. Wilczek] a legume crop, is primarily grown for grain and fodder by marginal and resource poor farmers mainly in the semi-arid and arid areas in the country. It contains 24% protein and

thus serves as major source of protein in vegetarian diet as well as improves the fertility of soil through biological nitrogen (N) fixation. Soil moisture stress limits crop productivity in the rainfed ecosystems worldwide. The deleterious effect of drought depends not only on its severity but also on the developmental stage of crop at which it occurs.

Generally, mungbean is cultivated under rainfed condition without or with minimal fertilizer. Mungbean faces moisture deficit episodes (erratic rainfall) during rainy as well as summer cultivation. It is cultivated as a short duration pulse crop and included in the rotation in several cropping systems under water scare environments. Although mungbean is considered as one of the drought sensitive crop among the grain legumes (Pandey et al. 1984; Daryanto et al. 2015) but its productivity is severely affected by water stress conditions, particularly in the spring and summer grown crops. The photosynthesis is inhibited by stomatal closure under drought stress that inhibits rubisco activity and increases respiration rates resulting in lower carbohydrate reserves and growth rate (Adams et al. 2009; Ghannoum, 2009). The ratio of photosynthesis to stomatal conductance given by  $WUE_{intr}$  (A/g<sub>s</sub>), is considered as a useful trait for selection of drought tolerant plants among C<sub>3</sub> species (Osmond et al. 1980). In mungbean, flowering is the most critical stage to water deficit stress leading to low grain yield (Morton et al. 1982; Daryanto et al. 2015). Occurrence of drought in mungbean during vegetative phase adversely affects the leaf expansion,

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biomass accumulation, number of pods and harvest index resulting in grain yield reduction (Robertson et al. 2004; Lalinia et al. 2004). However, Uprety and Bhatia (1989) observed that in mungbean, the grain yield and harvest index were the traits primarily influenced by terminal water deficit stress.

Mungbean has an innate capability to capture atmospheric N by symbiotic association with Rhizobium which is an energy intensive process. This makes phosphorus (P) nutrition a key factor in mungbean crop as P plays a major role in energy metabolism besides its involvement in membrane phospholipids, cellular signal transduction and regulation of several key enzymes. Under sufficient P, Rhizobium fixes ample amount of N which considerably increases protein content, biomass accumulation and seed yield of legume crops (Chaudhary and Fujita 1998). Deficiency of P has deleterious effects on plant growth, development and reproduction. Thus, optimum P supply stimulates biological processes such as nodulation, nitrogen fixation and nutrient uptake from soil and rhizosphere environment resulting in improved yield of legume crops. Constraint in plant growth and yield under P deficiency may be attributed mainly to its heterogeneous distribution and low mobility in soil (Bonser et al. 1996). The strong tendency of P-fixation in soils of dry and semi-dry regions is mostly because of the higher amounts of lime or oxides of aluminum and iron (Oertli 1991). Poor availability of P is the major challenge for pulses cultivation in India. Limited P supply to the chloroplast can restrict the photosynthetic process (Lawlor and Cornic 2002).

The movement of P in the soil is primarily through mass/bulk flow and diffusion which helps in contacting the root surface and root interception. So, it is expected that the acquisition of P by root would be reduced under moisture deficit condition (Pinkerton and Simpson, 1986). Thus, drought affects both the nutrient availability in soil, and growth and physiology of the plant. Higher P concentrations in plants increases WUE as it allows an inconsequential inhibition of photosynthesis per unit of water transpired there by improving the regulation of stomata (Jones et al. 2005; Waraich et al. 2011). Genotypic diversity in adaptive responses in mungbean (Pandey et al. 2014) and soybean (Vengavasi et al. 2016) to low P as well as to drought in mungbean have been reported (Raina et al. 2016, 2019). However, reports on combined stress tolerance to low P and drought in mungbean is meager. In this study, an attempt was

made to identify the efficient mungbean genotypes and key traits imparting low P stress tolerance under moisture deficit condition.

# Materials and methods

## Plant growth condition and observations recorded

Experiment was laid out in soil culture with 14 mungbean accessions at the Controlled Environment Facility, Division of Plant Physiology, ICAR-IARI, New Delhi. The pedigree information of accessions is presented in Table 1. Plants were grown in polyethene sleeves of size 14 cm diameter × 30 cm height filled

Table 1.Pedigree information of 14 mungbean<br/>genotypes

S. No.	Genotype name	Pedigree	Source
1	Pusa Ratna	VC 6368 x ML 267	Delhi
2	IC 280489	NKG 24	Odisha
3	PDM-139	ML 20/19 x ML 5	Uttar Pradesh
4	IC 76491	M 687	Delhi
5	IC 488875	PLM 376	Jammu & Kashmir
6	IC 119005	ML 9	Maharashtra
7	ML-818	5145/87 x ML 267	Punjab
8	IC 623705	Selection from NM 9	92 Delhi
9	EC 398886	VC 617B-13	Thailand
10	Asha	K 851 x L 24-2	Haryana
11	SML-668	Selection from NM 94	Punjab
12	IC 305250	Unknown	
13	IPM 2-3	IPM 99-125 x Pusa Bold-2	Uttar Pradesh
14	IC 398746	AKP-12/14	Bihar

with sieved soil low in available P (Olsen P, 7.80 mgP kg<sup>-1</sup> soil). The recommended fertilizer doses were uniformly mixed before filling except for single super phosphate (SSP). To create optimum P level, SSP (17.5 mg P kg<sup>-1</sup> soil) was applied to the soil. *Rhizobium leguminosarum* inoculated seeds were sown and after germination, plants were regularly irrigated. Drought treatment was applied by withholding irrigation for 10 days starting from 31 days after sowing until soil moisture was reduced to 10-11%. This moisture deficit in soil was retained for two days and subsequently, recovery was done by re-watering. The growth conditions were maintained as day/night temperature

at 30°C/27°C, relative humidity (RH) at 75-80% and natural light condition. The following observations were recorded:

A = Photosynthesis rate (imol  $CO_2 m^{-2} s^{-1}$ ); AGDW = above ground dry weight (g  $plant^{-1}$ ); Ci = Intercellular  $CO_2$  (µmol mol<sup>-1</sup>); CT = Canopy temperature (°C); E = Transpiration rate (mol m<sup>-2</sup>s<sup>-1</sup>); $g_{tc}$  = Total conductance to CO<sub>2</sub> (mol m<sup>-2</sup>s<sup>-1</sup>);  $g_{tw}$  = Total conductance to water (mol  $m^{-2}s^{-1}$ );  $g_{sw} =$ Stomatal conductance to water vapor (mol  $m^{-2} s^{-1}$ );  $GY = Grain yield (g plant^{-1}); HI = Harvest index (%);$ HCA = Hierarchical cluster analysis; ICE = Instantaneous carboxylation efficiency (A/Ci, %); IWUE = Instantaneous water use efficiency (A/E, µmol  $mol^{-1}$ ); NPP = Number of pods plant<sup>-1</sup>; NSP = Number of seeds  $pod^{-1}$ ; PCA = Principal component analysis; P = Phosphorus; PHI = Phosphorus harvest index (%); RWC = Relative water content (%); SI = Seed index (100 seed weight (g); SeedPc = Seed P concentration (mg  $g^{-1}$  dry weight); SeedPup = Seed P uptake (mg plant<sup>-1</sup>); ShootPc = Shoot P concentration (mg  $g^{-1}$  dry weight); ShootPup = Shoot P uptake (mg plant<sup>-1</sup>); WUE<sub>intr</sub> = Instantaneous intrinsic water use efficiency and  $A/g_{sw} = \mu mol mol^{-1}$ .

# Physiological observations

The RWC was estimated in fresh leaf discs of fully expended third trifoliate leaf from top. Leaf discs of 2 cm diameter were punched and fresh weight (FW) was recorded. Discs were floated in double distilled water for 24 h and the whole set up was kept in dark at 20°C for saturation. Turgor weight (TW) was taken after blotting excess water from leaf discs. Dry weight (DW) was taken after oven drying the leaf discs at 65°C for 48 h. The RWC was computed as per Barrs and Weatherley (1962):

$$RWC(\%) = \frac{FW - DW}{TW - DW} \times 100$$

The canopy temperature was recorded using an infrared thermal imaging camera (Testo-876, Testo India Pvt. Ltd., India). Images were taken at a height of 1.5 m from a tripod, covering six plants of each accession. Each observation consisted of three images of three replications (n = 9). Images were analysed by IRsoft 3.7 software and CT was estimated after avoiding soil background. For gas exchange measurements, fully opened third trifoliate leaves were used. Photosynthesis and related traits were recorded with the infrared gas analyzer (Li-6800, Li-COR Inc., NE, USA) under photosynthetic photon flux density

(PPFD) 1200 mmol m<sup>-2</sup> s<sup>-1</sup>, 400 µmol mol<sup>-1</sup> CO<sub>2</sub>, temperature of  $28\pm1^{\circ}$ C and RH 65  $\pm 2\%$  between 10.00-12.30 h. The IWUE (µmol mol<sup>-1</sup>) was computed from the ratio of A/E, WUE<sub>intr</sub> (µmol mol<sup>-1</sup>) as the ratio of A/g<sub>sw</sub> and ICE was calculated as P<sub>N</sub>/C<sub>i</sub>.

The pods were harvested in two pickings at maturity, that is, at 42 and 55 days after sowing. Seed yield and its attributes were determined by adding both pickings. GY, NPP, NSP 100-seed weight (g) were recorded.

# Estimation of phosphorus content in different tissue

Different plant tissues were dried separately for 48 hr at 80°C and ground into powder with a ball mill (MM 400, Retsch GmbH, Germany). Wet digestion was done with di-acid mixture composed of acids nitric:perchloric at 9:4 ratio and concentration of P was measured by inductively coupled plasma atomic emission spectrometry (ICP-OES; Model 5110, Agilent Technologies, Singapore). Mean were taken from the results of four replications and expressed as mg P  $g^{-1}$  dry weight. The P uptake in plant parts was obtained as P concentration multiplied by weight of seed or AGDW, and was expressed as mg P plant<sup>-1</sup>. The PHI was computed as P content in seed divided by total P content (seed P uptake + shoot P uptake).

# Stress tolerance evaluation

The stress tolerance of accessions under different treatments were evaluated by calculating the relative stress index (RSI) which was obtained by the formula, RSI = (Trait value under treated condition)/(Trait value under controlled condition) × 100. The principal component analysis (PCA) ranking values were used to assess stress tolerance of accessions under different treatments. For each mungbean accession, ranking value was determined by the formula (Zhu et al. 2014); PCA ranking value = (contribution of PC1 (%) × PC1) + (contribution of PC2 (%) × PC2) + (contribution of PC3 (%) × PC3) + (contribution of PC4 (%) × PC4).

#### Statistical analysis

The experiments were laid out with three factors in a completely randomized design with P levels, water regimes and accessions. The three-factor analysis of variance (ANOVA), PCA and hierarchical cluster analysis were performed by R ver 3.6.1 (R core team, 2019). PCA was used to identify the common trend of the multidimensional data sets. PCA analysis and

cluster analysis were carried out by FactoMineR (Sebastien et al. 2008) and cluster R package (Maechler et al. 2019), respectively.

# **Results and discussion**

# Response of mungbean accessions to low P, drought and combined stresses

The identification of phenotypic traits related to abiotic stress is the basic step for evaluating genotypic variation imparting tolerance. Data analysis and their interpretation collected from multiple stress conditions are still challenging. The agro-physiological traits of mungbean accessions were evaluated under different treatments. Gaseous exchange parameters (A, E, C<sub>i</sub>, g<sub>sw</sub>, g<sub>tc</sub>, g<sub>tw</sub>, ICE, IWUE and WUE<sub>intr</sub>), yield and its attributes (AGDW, GY, SI, NPp, NSp and HI), P related traits (SeedPc, SeedPup, ShootPc, ShootPup and PHI) and traits associated with plant water status (RWC and CT) were taken into consideration. The significance of sources of variability for genotypes (G), phosphorus levels (P), moisture levels (W) and interactions of 14 mungbean genotypes under two moisture environments (irrigated and moisture deficit) and two P levels (sufficient P 40.0 mg P kg<sup>-1</sup> dry soil and low P 7.8 mg kg<sup>-1</sup> soil) was computed. For most of the traits, influence of P, W and G, as well as their interaction effects (P×W, P×G and W×G) were significant (P  $\leq$  0.05). However, no significant effect of P level on E, C<sub>i</sub>, g<sub>sw</sub>, g<sub>tc</sub>, g<sub>tw</sub>, IWUE, WUE<sub>intr</sub>, NSp and HI parameters were observed.

To find out the key traits for evaluating low P, drought and combined stress tolerance, a heatmap was plotted with hierarchical clustering analysis considering all four treatments. The accessions clustered according to four treatments clearly demonstrated the trait variability for each accession (Fig. 1). Hierarchical cluster analysis divided the traits into three distinct groups, Cluster I included traits representing the relative response to drought stress (SI, IWUE, WUE<sub>intr</sub> and CT). Cluster II included P related traits such as ShootPc, ShootPup and SeedPc and ICE while photosynthesis and yield attributing traits were grouped into Cluster III in which most of accessions showed reduction in trait value under drought and combined stresses. PCA and HCA are considered as one of the best approaches for



Fig. 1. Heatmap and hierarchical clustering for yield and physiological parameters under control, low P, drought and combined stress conditions in mungbean accessions. Cluster analysis of mungbean accessions (left) showed two main groups where one group represents 14 accessions under the well-watered and low P condition; while another group represents those accessionsgrown under drought and combined stress

assessing the accessions under different environments (Nazari and Pakniyat 2010; Dehbalaei et al. 2013; Liu et al. 2015). Combined heat map with HCA is a data visualization method that can be useful in the exploration of complex relationships between multiple parameters under multiple conditions. However, the heat map could not differentiate clearly among the accessions under all treatment conditions.

To assess the contribution of each trait under all treatments, PCA was performed employing all 22 traits. Most of the traits were found to contribute significantly to the genotypic variation when grown under control and low P stress (Fig. 2). Hence, traits like A, RWC, AGDW, GY and SeedPup exhibited higher contribution in control while E,  $g_{sw}$ ,  $g_{tc}$ ,  $g_{tw}$  and  $C_i$  showed higher contribution under low P stress. However, among the 22 traits, IWUE, WUE<sub>intr</sub>, CT and SI were significantly positively related to drought and combined stress. ShootPc showed no contribution to grouping of accessions under any treatment. Higher shoot

biomass, HI, SPAD values, stomatal conductance and maintenance of CT are the drought tolerance indicators suggested in earlier reports (Krishnamurthy et al. 1999; Madhava et al. 2003; Jones et al. 2002; Sheshshayee et al. 2006; Rebetzke et al. 2013; Ramamoorthy et al. 2016). Many physiological traits such as P acquisition efficiency, SPAD values, organic root exudation, leaf P concentration and rate of transpiration are the potential targeted traits to improve low P tolerance (Peng et al. 1999; Pandey et al. 2014; Vengavasi et al. 2016; Pang et al. 2018).

# PCA biplot based on relative values of traits obtained under treated conditions

HCA and PCA revealed that all the recorded traits, except ShootPc, played a major role in categorizing the mungbean accessions under control, low P, drought and combined stresses. PCA based on RSI values of 21 traits revealed that the PC1 explained 53.1% of the genotypic variance while PC2, PC3 and PC4 explained an additional 12.4, 8.1 and 6.1% of the



Fig. 2. Biplot of various traits of 14 mungbean accessions under control (blue), low phosphorus (orange), drought (green) and combined stress (red)conditions. Arrows represent traits while its length is based on the contribution of each trait to separate the accessions

variance, respectively (Fig. 3). Most of RSI values showed higher variation under low P as compared to other treatment conditions. Photosynthesis related traits (A, E,  $g_{sw}$ ,  $g_{tw}$ ,  $g_{tc}$ ,  $C_i$  IWUE, and WUE<sub>intr</sub>), RWC and grain yield indicated higher variation under PC1 whereas ShootPup, PHI, and HI exhibited high variability under PC2. Least variation among the accessions were exhibited bytraits such as CT, SI and SeedPc. PDM139 (tolerant) showed higher value for photosynthetic traits (A, E,  $g_{sw}$ ,  $g_{tw}$  and  $g_{tc}$ ) and AGDW in comparison to IPM2-3 (sensitive) in all three treatment conditions. In earlier reports, PDM 139 was also found to be a P efficient genotype under hydroponic and soil culture conditions with low P availability (Pandey et al. 2013, 2014).

PCA-biplots showed the relative contributions of the estimated traits and clear distribution of all accessions under all conditions (Figs. 2 and 3). The RSIs for each trait were used to evaluate the relative tolerance to low P, drought and their combined

stresses in mungbean accessions. PCA results revealed that the tolerant accessions under all three treated conditions had higher RSIs as compared to the sensitive one. Similar indexes for PCA analysis under drought were used in switch grass (Liu et al. 2015), sesame (Dossa et al. 2017) and tomato (Aghaie et al. 2018). Liu et al. (2015) reported that relative stress indices of RWC, photosynthetic traits (g<sub>sw</sub>, A, E and C<sub>i</sub>) and electrolyte leakage were the most efficient traits to screen for drought tolerance in switch grass. Besides, the relative stress indices of biochemical traits such as antioxidative enzymes (SOD, APX, CAT), MDA, and proline were also found to be useful in selection of genotypes exposed to water deficit stress (Dossa et al. 2017; Aghaie et al. 2018). Based on relative <sup>14</sup>C exudation through roots and biomass, P efficient genotypes were identified in green gram (Pandey et al. 2013) and soybean (Vengavasi et al. 2016).

#### Stress tolerance ranking of mungbean accessions



Fig. 3. Biplot of the relative values of various physiological traits of mungbean accessions grown under low phosphorus, drought and combined stress conditions. Arrows represent agro-physiological traits while its length is based on the contribution of each trait to separate the accessions.

PC1 PC2 PC3 PC4 Ranking Numeric ranking Low P Drought Combined stresses PUSA RATNA<sup>p</sup> 446 15 -44 -190 223 1 IC 280489<sup>p</sup> 446 -25 -69 2 -186 216 PDM-139<sup>p</sup> 3 426 -32 -186 11 213 IC 76491<sup>p</sup> 357 27 -15 -150 183 4 IC 488875<sup>p</sup> 343 -35 1 -131 170 5 IC 119005<sup>p</sup> 307 4 -143 6 38 158 ML-818<sup>p</sup> 7 291 -20 50 -105 150 IC 623705<sup>p</sup> 287 -37 37 -120 143 8 EC 398886<sup>p</sup> 269 -54 26 -105 132 9 **ASHA**<sup>p</sup> -39 250 54 -93 126 10 SML-668<sup>p</sup> 207 -19 106 -80 111 11 EC 398886<sup>d</sup> 230 -40 16 -125 111 1 IC 305250<sup>p</sup> 207 -47 67 -57 106 12 IPM 2-3<sup>p</sup> 98 191 -31 60 -63 13 PDM-139<sup>d</sup> 147 -55 84 -50 75 2 IC 398746<sup>p</sup> -74 70 139 101 -44 14 IC 76491<sup>d</sup> 136 68 -73 69 3 -31 PUSA RATNA<sup>d</sup> 102 8 96 -75 58 4 ML-818<sup>d</sup> 105 -72 135 -77 53 5 IC 623705<sup>d</sup> 103 -64 6 112 -85 50 EC 398886b<sup>cb</sup> 116 -93 48 -94 48 1 IC 76491<sup>cb</sup> 101 -63 67 -76 47 2 IC 305250<sup>d</sup> 7 90 -57 101 -40 46 IC 305250<sup>cb</sup> -70 3 97 86 -59 46 PDM-139<sup>cb</sup> 91 -72 81 -32 44 4 SML-668<sup>d</sup> 85 -56 88 -82 40 8 IC 280489<sup>d</sup> 9 86 -86 138 -106 40 IC 623705<sup>cb</sup> 77 -83 93 -79 33 5 IC 398746<sup>d</sup> 65 -44 97 -88 32 10 PUSA RATNA<sup>cb</sup> -27 77 32 6 64 -86 IPM 2-3<sup>d</sup> 7 92 40 -56 26 11 IC 488875<sup>cb</sup> 54 7 -41 70 -74 25 IC 488875<sup>d</sup> 39 -25 104 22 12 -68 ASHA<sup>d</sup> -2 37 80 -65 22 13 ML-818<sup>cb</sup> 52 -134 113 -87 15 8 IC 280489<sup>cb</sup> 42 -121 105 -79 9 11  $\mathsf{ASHA}^{\mathsf{cb}}$ 15 -15 74 -77 7 10 IC 119005<sup>cb</sup> 10 0 73 -97 5 11 IC 119005<sup>d</sup> 4 -12 110 -83 4 14 SML-668<sup>cb</sup> -92 -3 11 102 -88 12 IC 398746<sup>cb</sup> -12 13 -65 101 -97 -12 IPM 2-3<sup>cb</sup> -41 -32 81 -66 -23 14

 Table 2.
 The four principal components (PC1, PC2, PC3 and PC4) and PCA ranking values of the physiological parameters of 14 mungbean accessions under low phosphorus (p), drought (d) and combined stresses (cb)

Superscript letters denote the stress condition

The PCA based on the RSI values of 21 traits showed that four components (PC1, PC2, PC3 and PC4) contributed 79.68% of total genetic variation in response to the treatments. The PCA ranking value was computed for each accession according to formula:

PCA ranking value = (53.1% of PC1 score) + (12.4% of PC2 score) + (8.1% of PC3 score) + (6.1% of PC4 score) (Table 2).

Accessions, IC 280489, PDM 139 and IC 76491 showed relatively higher ranking values under all treatment conditions suggesting that they were more tolerant to low P, drought and combined stress, whereas IPM2-3 showed relatively lower ranking values indicating more sensitive to P or drought stresses. Based on the PCA ranking values (Table 3), Pusa Ratna, IC 280489 and PDM139 and IC 76491 were

Table 3.Ranking summary of mungbean accessions<br/>exposed to low phosphorus, drought and<br/>combined stresses

Ranking	Low P	Drought	Combined stresses
1	PUSA RATNA	EC 398886	EC 398886
2	IC 280489	PDM-139	IC 76491
3	PDM-139	IC 76491	IC 305250
4	IC 76491	PUSA RATNA	PDM-139
5	IC 488875	ML-818	IC 623705
6	IC 119005	IC 623705	PUSA RATNA
7	ML-818	IC 305250	IC 488875
8	IC 623705	SML-668	ML-818
9	EC 398886	IC 280489	IC 280489
10	ASHA	IC 398746	ASHA
11	SML-668	IPM 2-3	IC 119005
12	IC 305250	IC 488875	SML-668
13	IPM 2-3	ASHA_d	IC 398746
14	IC 398746	IC 119005	IPM 2-3

identified as P efficient, whereas SML668, IC 305250, IPM 2-3, and IC 398746 were P inefficient under low P treatment. Drought tolerance was shown by accessions, EC 398886, PDM139, IC 76491 and Pusa Ratna while IPM 2-3, IC 488875, Asha and IC 119005 were drought sensitive. The accessions showing tolerance to combined stresses were EC 398886, IC 76491, IC 305250 and PDM-139, while accessions IC

119005, SML668, IC 398746 and IPM 2-3 were most sensitive to combined stresses. Similar PCA ranking values were used to identify the superior accessions in switch grass (Liu et al. 2016), onion (Zhu et al. 2014) and tomato (Aghaie et al. 2018).

The superior accessions exhibited higher relative values for photosynthetic traits (A, E, g<sub>sw</sub>, g<sub>tc</sub>, g<sub>tw</sub> and C<sub>i</sub>), RWC, AGDW, NPp and SeedPup, while it was less for CT and WUE in all three treatments. Generally, CT value increases under water deficit condition but accessions maintaining lower CT values are relatively stress tolerant. So, accessions IC 119005, SML668, IC 398746, IC 488875, Asha and IPM 2-3 possessing higher relative CT value showed susceptibility to drought and combined stresses. Further, higher E value was reported to be correlated with P concentration in leaf in chickpea (Pang et al. 2018). In present study, the higher relative values of E and gsw was recorded in P efficient genotype PDM139. According to Liu et al. (2015), higher relative stress indices for RWC, photosynthetic traits (g<sub>sw</sub>, A, E and C<sub>i</sub>), and lower electrolyte leakage were the most efficient traits to evaluate drought tolerance in switch grass.

In conclusion, the present study has identified a number of physiological markers that can be employed to analyse the diverse mungbean accessions for tolerance to drought and low P stress. Tolerant accessions (PDM139, IC 76491 and EC 398886) and traits such as photosynthetic traits, lower CT value, RWC, AGDW and SeedPup can be utilized in mungbean breeding programs to improve the tolerance to drought and low soil P availability. Further, the PCA ranking can be used as a robust tool to identify the tolerant accessions under multiple treatments based on changes in different traits.

# Authors' contribution

Conceptualization of research (RP); Designing of the experiments (SKM, RP); Contribution of experimental materials (RP, G, MP); Execution of field/lab experiments and data collection (SKM); Analysis of data and interpretation (SKM, RP); Preparation of manuscript (SKM, RP).

# Declaration

The authors declare no conflict of interest.

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