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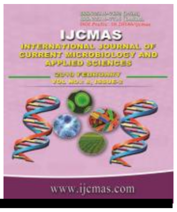

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# Identification of High Yielding Recombinant Inbred Lines (RIL) Derived from Two Bi-Parental Crosses in Dolichos Bean (*Lablab purpureus* L. Sweet)

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## ABSTRACT

**Keywords**

Dolichos bean, Multiple traits, Recombinant inbred lines (RILs), Quantitative traits, Trait-specific RILs

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The recombinant inbred lines (RILs) serve as useful source of pure-lines with desired combination of traits. Also, the chances of recovering high-yielding genotypes from RIL population are higher than those recoverable from $F_2$ populations handled through pedigree breeding. The 157 RILs derived from HACPI 3and 144 RILs derived from HACPI 6 and three check entries [HA 3, HA 4 and kadalavare] of dolichos bean were evaluated for six qualitative traits and nine quantitative traits at the University of Agricultural Sciences (UAS), Bengaluru, India, during 2014 and 2015 rainy seasons. Substantial variability among the RILs for quantitative traits and polymorphism for qualitative traits were documented. The pooled analysis of variance revealed highly significant mean squares attributable to RILs, checks and RILs vs. checks for all the traits in both the populations. The RILs, 3-26 and 3-35 were particularly superior to all the three checks, HA 3, HA 4 and kadalavare for fresh pods plant$^{-1}$, fresh pod yield plant$^{-1}$ and fresh seed yield plant$^{-1}$. HACPI 3-derived RILs such as, 3-3, 3-26, 3-30, 3-35, 3-40, 3-95, 3-134, 3-139, 3-141, 3-185 and those derived from HACPI 6,such as,6-262, 6-278, 6-310, 6-356, 6-364, 6-366 and 6-367 were promising for multiple quantitative traits and also for farmer and consumer-preferred qualitative traits. These RILs are suggested for preferential use in breeding high yielding dolichos bean varieties with desirable pod traits.

## Introduction

Dolichos bean (*Lablab purpureus* L. Sweet) is one of the ancient and important grain legume crops grown in India (Ayyangar and Nambiar, 1935; Vishwanath and Manjunath, 1971). It is predominantly a self-pollinated crop with 2n=2x=22 chromosomes (She and Jiang, 1986) with a genome size of 367 Mbp (Iwata *et al.*, 2013). In India, it is mainly grown as a rainfed crop for its fresh immature beans for use as a vegetable (Shivashankar and Kulkarni, 1989). Fresh pods containing immature beans are economic products in dolichos bean. It is considered as poor man's bean and is one of the major sources of dietary protein to the people who depend on vegetarian diet in southern parts of Karnataka, Tamil Nadu and Andhra Pradesh.

Dolichos bean has evolved as highly photoperiod sensitive short-day plants with indeterminate growth habit (Keerthi *et al.*, 2014). Most of the cultivars used for dolichos bean production are landraces which are photoperiod sensitive and exhibit indeterminate growth habit. Traditionally dolichos bean production is restricted to late rainy season characterized by short-days. However, of late, due to market economy there is demand for dolichos bean throughout the year. However, production of dolichos bean throughout the year is possible only using photoperiod insensitive determinate cultivars. Hence, development of high yielding photoperiod insensitive determinate pure-line varieties is the major breeding objective of dolichos bean (Keerthi *et al.*, 2016).

Pedigree selection of desirable genotypes derived from planned crosses is the most widely used breeding method to develop improved pure-line varieties in dolichos bean (Keerthi *et al.*, 2016). The recombinant inbred lines (RILs) derived from crossing two inbred parents contrasting for target traits are not only serve as an ideal population to develop DNA marker-based linkage map and identify markers linked to genomic regions controlling target traits for which their parents differ, but also a useful source of pure-lines with desired combination of traits. Besides this, chances of recovering high-yielding genotypes from RIL population are higher than those from $F_2$-populations handled from pedigree selection. This is because, in pedigree breeding method selection is initiated from $F_2$ which often results in rejecting the undesirable $F_2$plants which otherwise might result in RILs with desirable combination of traits in advanced generation. The objectives of the present investigation were to (1) phenotype and assess the variability of the two RIL populations derived from two bi-parental crosses for pod yield and its component traits and (2) to identify RILs with desired combination of traits.

## Materials and Methods

The material for the study comprised of 157 RILs derived from HA 4 x CPI 31113 (here after referred as HACPI 3) and 144 RILs derived from HA 4 x CPI 60125 (here after referred as HACPI 6) and three check entries [HA 3, HA 4 and kadalavare (KA)] maintained at All India Co-ordinated Research Project (AICRP) on pigeon pea, University of Agricultural Sciences (UAS), Bengaluru. The seeds of these RILs differed in thickness. Hence, seeds of RILs with thick seed coat were scarified by rubbing against hard surface without damaging the plumule, so that water can be easily imbibed to facilitate quick germination. The seedlings of all the RILs and the checks were raised in polythene covers and maintained for 15-20 days for proper rooting.

Subsequently, the seedlings of two RIL populations, along with three check entries were transplanted to field in an augmented design (Federer 1956) in eight compact blocks for each RIL population during 2014-2015 rainy seasons at the experimental plot of Zonal Agricultural Research Station (ZARS), UAS, Bengaluru. Each block consisted of 18-20 RILs, three checks and two border entries. The seedlings of each entry were transplanted in a single row of 2.5 m length, with a row spacing of 0.45 m. A basal dose of 25:50:25 Kg $ha^{-1}$ of NPK (nitrogen: phosphorous: potassium) was applied to the experimental plots. Recommended management practices were followed during the crop-growth period to raise a healthy crop.

## Sampling of plants and data collection

In HACPI 3-derived RILs, out of 157 planted only 136 individuals and in HACPI 6-derived RILs, out of 144 planted, only 119 individuals survived till the maturity. Data were collected only from survived individuals in both the RIL populations for two successive years. Data were recorded on five randomly tagged plants on six qualitative traits (growth habit, pod fragrance, pod constriction, pod orientation and seed coat colour) based on visual observation (Table 1). Although data on several other qualitative traits could be recorded, we considered only these six traits as they are most preferred by farmers and end-users (Vaijayanthi *et al.*, 2016). Data were also recorded on nine quantitative traits (days to 50% flowering, raceme bearing branches plant$^{-1}$, raceme length, racemes plant$^{-1}$, fresh pods plant$^{-1}$, fresh pod yield plant$^{-1}$, fresh seed yield plant$^{-1}$, dry seed yield plant$^{-1}$ and 100 seed weight) based on counting/measurement using appropriate scale (Table 2) depending on the trait in each RILs and check entries following the descriptors (Byregowda *et al.*, 2015).

## Statistical analysis

Variability among the individuals of two RIL populations for seven qualitative traits was quantified by computing percentage of RILs exhibiting different states of each of the qualitative traits. The quantitative trait means of each RILs and each check entriesfor all the nine characters were estimated. The components of variance were estimated using Residual Maximum Likelihood (REML) method following linear mixed model (Federer and Wolfinger, 1998). Pooled analysis of variance was carried out to detect the block×year, checks×year and RILs×year interactions by using REML linear mixed model approach (Patterson and Thompson 1971) implemented using PROC GLM in SAS 9.4 (SAS Institute Inc., Cary, NC, USA).Best linear unbiased predictors (BLUPs) (Schonfeld and Werner 1986) were obtained for all the quantitative traits for each RILs. Two-year pooled quantitative trait means were used for estimating descriptive statistics, such as trait range (R) = Max–Min, standardized range (SR) = (Min–Max)/X (where X = trait mean), and variance components attributable to genotypes ($\sigma^2_g$) = (MSSg – MSSe)/b (where, MSSg =Mean squares attributable to RILs, MSSe = Mean squares due to error, b= Number of blocks), Pooled phenotypic variance $\sigma_p^2 = \sigma_g^2 + (\sigma_{ge}^2)/y + (\sigma_e^2)/(y\times b)$(where,$\sigma_g^2$= Genotypic variance, $\sigma^2_{ge}$ = genotype x years, $\sigma_e^2$= Error variance, y= years and b = number of blocks). The phenotypic co-efficient of variability (PCV) and genotypic co-efficient of variability (GCV) for all the characters were estimated [2].

## Criteria to identify trait-specific RILs

Based on early flowering time (40–50 days from date of sowing) and traits expression of RILs significantly better than those of check entries, the trait-specific RILs and those desirable for combination of traits were identified.

## Results and Discussion

### Qualitative traits

The HACPI 3-derived RILs with determinate growth habit were higher in frequency followed by those with indeterminate and semi-determinate growth habits. On the other hand, HACPI 6-derived RILs with determinate and indeterminate growth habits are equally distributed followed by those with semi-determinate types (Figure 1a). The frequency of white flower-bearing RILs derived from both the crosses was higher than that of purple flower bearing RILs (Figure 1b).

**Table.1** Protocol followed to record data on six qualitative traits in RIL populations derived from two bi-parental crosses in dolichos bean

| Sl.no | Traits | Crop stage for recording data | Score | Classified as |
|---|---|---|---|---|
| **1** | Growth habit | At the time of flowering | 1 | Determinate |
| | | | 2 | Semi-determinate |
| | | | 3 | Indeterminate |
| **2** | Flower color | At the time of flower opening | 0 | White |
| | | | 1 | Purple |
| **3** | Pod fragrance | At the time of pod filling | 0 | Absent |
| | | | 1 | Low |
| | | | 2 | Medium |
| | | | 3 | High |
| **4** | Pod constriction | At the time of pod filling | 0 | No constriction |
| | | | 3 | Slightly constricted |
| | | | 5 | Constricted |
| **5** | Fresh seed coat color | At the time of harvesting | 1 | Green |
| | | | 2 | Cream |
| | | | 3 | Purple |
| | | | 4 | Brown |
| | | | 5 | Black |
| **6** | Dry seed coat color | At the time of harvesting | 1 | Green |
| | | | 2 | Cream |
| | | | 3 | Purple |
| | | | 4 | Brown |
| | | | 5 | Black |

**Table.2** Protocol followed to record data on nine quantitative traits in RIL populations derived from two bi-parental crosses in dolichos bean

| Sl.no | Traits | Procedure of measurement/counting |
|---|---|---|
| **1** | Days to 50% flowering | Number of days taken from sowing to 50% of the plants to produce flowers was counted. |
| **2** | Raceme bearing branches plant$^{-1}$ | Number of Raceme bearing branches plant$^{-1}$on 5 randomly selected plants were counted and averaged |
| **3** | Raceme length | Raceme length of 5 randomly selected plants were measured in centimeters and averaged |
| **4** | Racemes plant$^{-1}$ | Number of racemes from 5 randomly selected plants were counted and averaged |
| **5** | Fresh pods plant$^{-1}$ | Number of fresh pods from 5 randomly chosen mature plants were counted and averaged |
| **6** | Fresh pod yield plant$^{-1}$ (g) | Fresh pod yield of 5 randomly chosen plants were recorded in grams and averaged. |
| **7** | Fresh seed yield plant$^{-1}$ (g) | Fresh seeds of 5 randomly chosen plants were weighed, recorded in grams and averaged over 5 plants. |
| **8** | Dry seed yield plant$^{-1}$ (g) | Dry seeds of 5 randomly chosen plants were weighed, recorded in grams and averaged over 5 plants. |
| **9** | 100- seed weight (g) | 100 seeds from randomly selected pods were weighed and recorded in grams. |

**Table.3** Combined analyses of variance of RIL populations derived from two bi-parental crosses in dolichos bean for quantitative traits

| Source of Variation | df | | DFF | | RBP | | RL | | RP | | FP | | FPY | | FSY | | DSY | | TW | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | C-I | C-II | C-I | C-II | C-I | C-II | C-I | C-II | C-I | C-II | C-I | C-II | C-I | C-II | C-I | C-II | C-I | C-II | C-I | C-II |
| **Years** | 1 | 1 | 9.63 | 21 | 148.75** | 135.67** | 34.44** | 44.02** | 0.02 | 0.54 | 1191.01** | 1165.25** | 270.48* | 237.18** | 25.55 | 17.66 | 183.84** | 145.53** | 0 | 0 |
| **Block** | 7 | 7 | 30.96 | 22.87* | 0.96 | 0.73 | 1.39** | 1.39* | 0.64** | 0.89 | 42.19 | 38.94 | 142.91* | 124.44** | 36.67** | 27.26** | 3.47* | 3.16* | 1.41 | 2.11* |
| **Checks** | 2 | 2 | 1316.75** | 1640.09** | 207.53** | 340.96** | 98.18** | 122.52** | 0.64* | 1.99* | 304.48** | 359.77** | 490.19** | 963.24** | 193.23** | 263.25** | 295.79** | 398.43** | 28.25** | 21.41** |
| **RILs** | 135 | 118 | 109.80** | 239.66** | 27.04** | 21.29** | 11.14** | 8.02** | 3.24** | 3.13** | 188.30** | 87.92** | 814.77** | 215.13** | 166.54** | 46.52** | 42.25** | 33.28** | 23.10** | 14.86** |
| **Block*Years** | 7 | 7 | 3.44 | 3.52 | 0.11 | 0.08 | 0.15 | 0.15 | 0.07 | 0.1 | 4.69 | 4.33 | 15.88 | 13.83 | 4.08 | 3.03 | 0.39 | 0.35 | 0.16 | 0.24 |
| **Checks*Years** | 2 | 2 | 39.88 | 5.1 | 13.44** | 14.46** | 9.47** | 12.64** | 1.71** | 2.18* | 5.52 | 6.27 | 295.30** | 361.41** | 17.35* | 26.68** | 14.30** | 23.31** | 10.99** | 14.05** |
| **RILs*Years** | 135 | 118 | 106.63** | 127.89** | 11.86** | 8.96** | 3.59** | 3.77** | 0.83** | 0.89** | 34.32* | 37.18* | 66.92 | 79.27** | 16.29** | 19.02** | 6.80** | 33.57** | 3.29** | 1.91** |
| **Error** | 28 | 28 | 33.36 | 8.91 | 0.56 | 1.49 | 0.31 | 0.42 | 0.18 | 0.38 | 19.39 | 19.12 | 49.95 | 30.97 | 6.45 | 4.49 | 1.17 | 1.02 | 0.86 | 0.64 |

*= Significant at P=0.05 **= Significant at P=0.01

| | |
|---|---|
| **C-I - HACPI 3** | **FP- Fresh pods plant$^{-1}$** |
| **C-II - HACPI 6** | FPY- Fresh pod yield plant$^{-1}$ (g) |
| **DFF- Days to 50% flowering** | FSY Fresh seed yield plant$^{-1}$ (g) |
| **RBP- Raceme bearing branches plant$^{-1}$** | DSY Dry seed yield plant$^{-1}$ (g) |
| **RL-Raceme length (cm)** | TW- 100- seed weight (g) |
| **RP-Racemes plant$^{-1}$** | |

**Table.4** Descriptive statistics for quantitative traits in RIL populations derived from two bi-parental crosses in dolichos bean

| Traits | Mean ± SE | | Range | | | | Standardized Range | | PCV% | | GCV % | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | | | Min | Max | Min | Max | | | | | | |
| | HACPI 3 | HACPI 6 | HACPI 3 | | HACPI 6 | | HACPI 3 | HACPI 6 | HACPI 3 | HACPI 6 | HACPI 3 | HACPI 6 |
| **Days to 50% flowering** | 62.22±0.81 | 63.63±1.05 | 37.00 | 82.00 | 39.50 | 87.00 | 0.72 | 0.75 | 5.95 | 15.19 | 4.97 | 8.44 |
| **Raceme bearing branches plant$^{-1}$** | 12.28±0.35 | 12.48±0.31 | 6.70 | 28.80 | 6.50 | 24.90 | 1.80 | 1.47 | 14.97 | 21.28 | 14.81 | 12.61 |
| **Raceme length (cm)** | 12.01±0.21 | 12.31±0.19 | 6.01 | 20.38 | 6.58 | 21.40 | 1.20 | 1.20 | 9.83 | 13.75 | 9.69 | 7.92 |
| **Racemes plant$^{-1}$** | 8.39±0.12 | 8.77±0.12 | 5.10 | 12.00 | 5.50 | 12.60 | 0.82 | 0.81 | 7.60 | 10.30 | 7.38 | 6.69 |
| **Fresh pods plant$^{-1}$** | 28.30±0.86 | 27.88±0.62 | 13.50 | 66.50 | 10.50 | 46.50 | 1.87 | 1.29 | 17.14 | 19.11 | 16.23 | 10.52 |
| **Fresh pod yield plant$^{-1}$ (g)** | 46.07±1.86 | 39.40±0.97 | 12.20 | 140.40 | 14.58 | 86.85 | 2.78 | 1.83 | 21.91 | 20.40 | 21.22 | 12.18 |
| **Fresh seed yield plant$^{-1}$ (g)** | 22.20±0.83 | 18.43±0.45 | 8.73 | 65.65 | 5.96 | 34.22 | 2.56 | 1.53 | 20.55 | 21.05 | 20.15 | 12.44 |
| **Dry seed yield plant$^{-1}$ (g)** | 11.35±0.42 | 9.58±0.38 | 4.51 | 27.28 | 4.17 | 43.34 | 2.01 | 4.09 | 20.25 | 47.69 | 19.97 | 20.96 |
| **100- seed weight (g)** | 13.44±0.30 | 63.63±1.05 | 7.09 | 25.25 | 6.06 | 21.64 | 1.35 | 1.34 | 12.64 | 14.34 | 12.40 | 11.47 |

**Table.5** Promising trait-specific RILs derived from two bi-parental crosses in dolichos bean

| **Traits** | **Selection Criteria** | **RILs** | |
|---|---|---|---|
| | | HACPI 3 | HACPI 6 |
| **Days to 50% flowering** | Earliness (40-50 days after sowing) | 3-4, 3-7, 3-26, 3-27, 3- 35, 3-45, 3-69, 3- 71, 3-80, 3-89, 3-91, 3- 103, 3-119, 3-126, 3-162 and 3-182 | 6-190, 6-205, 6-209, 6-211, 6-214, 6-217, 6-238, 6-241, 6-248, 6-268, 6-285, 6-290, 6-302, 6-307, 6-308, 6-310, 6-311, 6-335, 6-349 and 6-350 |
| **Raceme bearing branches plant$^{-1}$** | High* | 3-31, 3-35, 3-40, 3-65, 3-76, 3-95, 3-108, 3-109, 3-120, 3-130, 3-132, 3-134, 3-139, 3-140, 3-141, 3-146 and 3- 184 | 6-198, 6-205, 6-208, 6-261, 6-262, 6-264, 6-266, 6-267, 6-269, 6-274, 6-276, 6-278, 6- 290, 6-296, 6-297, 6-303, 6-310, 6-343, 6-347, 6-351, 6-352, 6-355, 6-356, 6-365 and 6-366 |
| **Raceme length (cm)** | High* | 3-33, 3-36, 3-40, 3-53, 3- 68, 3- 91, 3- 134, 3-172 and 3-185 | 6-193, 6-209, 6-231, 6-253, 6- 261, 6-262, 6-278, 6-333, 6-356, 6-357 and 6- 363 |
| **Racemes plant$^{-1}$** | High* | 3-31, 3-36, 3-40, 3-73, 3-87, 3-88, 3-95, 3-129, 3-130, 3-134, 3-140, 3-141, 3- 142, 3-146, 3-164, 3-164 and 3-185 | 6-194, 6-262, 6-276, 6-278, 6-296 and 6-356 |
| **Fresh pod plant$^{-1}$** | High* | 3-3, 3-26, 3-30, 3-35, 3-125, 3-139, 3-141 and 3-182 | 6-198, 6-248, 6-364, 6-366 and 6-367 |
| **Fresh pod yield plant$^{-1}$(g)** | High* | 3-3, 3-26, 3-30, 3-35, 3-46, 3- 71, 3-95, 3-139, 3-141, 3-150 and 3-185 | 6-218, 6-364, 6-366 and 6-367 |
| **Fresh seed yield plant$^{-1}$(g)** | High* | 3-3, 3- 22, 3-26, 3- 30, 3- 35, 3-95, 3-124, 3- 139, 3-141, 3-163, 3- 176 and 3- 185 | 6-367 |
| **Dry seed yield plant$^{-1}$(g)** | High* | 3-26, 3-139 and 3-185 | 6-326 |
| **100- seed weight (g)** | High* | 3-60, 3-120, 3-124 and 3-185 | 6-264, 6-293, 6-310, 6-311, 6-333, 6-356 and 6-362 |

* - Significantly higher than the checks- HA 3, HA 4 and Kadalavare

**Table.6** Promising RILs identified for multiple traits in a HACPI 3 population

| Identity of RILs | Traits |
|---|---|
| **RIL 3-3** | Fresh pods plant$^{-1}$, fresh pod plant$^{-1}$ and fresh seed yield plant$^{-1}$. |
| **RIL 3-26** | Days to 50% flowering, fresh pod plant$^{-1}$ and fresh pod yield plant$^{-1}$, fresh seed yield plant$^{-1}$. |
| **RIL 3-30** | Fresh pod plant$^{-1}$, fresh pod yield plant$^{-1}$ and fresh seed yield plant$^{-1}$. |
| **RIL 3-35** | Days to 50% flowering, raceme bearing branches plant$^{-1}$, fresh pod plant$^{-1}$, fresh pod yield plant$^{-1}$ and fresh seed yield plant$^{-1}$. |
| **RIL 3-40** | Raceme bearing branches plant$^{-1}$, raceme length and racemes plant$^{-1}$. |
| **RIL 3-95** | Raceme bearing branches plant$^{-1}$, racemes plant$^{-1}$ fresh pod yield plant$^{-1}$ and fresh seed yield plant$^{-1}$. |
| **RIL 3-134** | Raceme bearing branches plant$^{-1}$, raceme length and racemes plant$^{-1}$. |
| **RIL 3-139** | Raceme bearing branches plant$^{-1}$ , fresh pod plant$^{-1}$, fresh pod yield plant$^{-1}$ fresh seed yield plant-$^{1}$ and dry seed yield plant$^{-1}$. |
| **RIL 3-141** | Raceme bearing branches plant$^{-1}$racemes plant$^{-1,}$ fresh pod plant$^{-1}$, fresh pod yield plant$^{-1}$ and fresh seed yield plant$^{-1}$. |
| **RIL 3-185** | Raceme length, racemes plant$^{-1}$, fresh pod yield plant$^{-1}$, fresh seed yield plant-$^{1}$ dry seed yield plant$^{-1}$ and 100-seed weight. |

**Table.7** Estimates of quantitative traits means of the HACPI 3-derived RILs promising for multiple traits

| Identity of RILs | DFF | RBP | RL | RP | FP | FPY | FSY | DSY | TW |
|---|---|---|---|---|---|---|---|---|---|
| **RIL 3-3** | 55.50 | 13.80 | 14.74 | 9.30 | 34.50 | 64.88 | 33.54 | 17.07 | 15.54 |
| **RIL 3-26** | 42.50 | 12.80 | 13.84 | 7.70 | 47.50 | 140.40 | 65.65 | 27.27 | 17.10 |
| **RIL 3-30** | 66.00 | 12.50 | 15.64 | 9.70 | 49.00 | 88.35 | 51.83 | 13.52 | 16.48 |
| **RIL 3-35** | 44.00 | 17.40 | 13.49 | 9.20 | 54.00 | 112.15 | 45.95 | 20.34 | 12.02 |
| **RIL 3-40** | 62.50 | 19.20 | 20.38 | 10.30 | 44.00 | 55.45 | 27.66 | 18.87 | 10.78 |
| **RIL 3-95** | 63.50 | 17.00 | 14.90 | 10.80 | 36.50 | 79.53 | 38.24 | 26.64 | 20.49 |
| **RIL 3-134** | 67.50 | 28.80 | 17.13 | 11.90 | 27.50 | 49.13 | 25.99 | 15.03 | 14.69 |
| **RIL 3-139** | 67.00 | 19.70 | 13.85 | 9.40 | 58.50 | 110.56 | 52.39 | 27.06 | 18.18 |
| **RIL 3-141** | 58.00 | 18.60 | 12.95 | 10.40 | 66.50 | 112.80 | 39.70 | 23.18 | 18.71 |
| **RIL 3-185** | 65.50 | 14.30 | 16.15 | 10.60 | 34.50 | 82.29 | 39.44 | 27.02 | 25.25 |
| **CHECKS** | | | | | | | | | |
| **HA 4** | 45.13 | 10.34 | 15.22 | 9.73 | 36.47 | 59.33 | 29.66 | 21.38 | 21.06 |
| **HA 3** | 47.88 | 9.25 | 11.20 | 9.33 | 34.13 | 57.89 | 27.77 | 18.52 | 18.81 |
| **Kadalavare** | 62.03 | 15.96 | 15.71 | 9.51 | 42.58 | 68.12 | 34.51 | 26.98 | 21.17 |
| **Sem±** | 2.04 | 0.26 | 0.20 | 0.15 | 1.56 | 2.50 | 0.90 | 0.38 | 0.33 |
| **CD @ P=0.05** | 5.90 | 0.76 | 0.57 | 0.43 | 4.50 | 7.22 | 2.59 | 1.10 | 0.95 |

| | |
|---|---|
| **DFF- Days to 50% flowering** | **FPY- Fresh pod yield plant$^{-1}$ (g)** |
| **RBP- Raceme bearing branches plant$^{-1}$** | FSY Fresh seed yield plant$^{-1}$ (g) |
| **RL-Raceme length (cm)** | DSY Dry seed yield plant$^{-1}$(g) |
| **RP-Racemes plant$^{-1}$** | TW- 100- seed weight (g) |
| **FP- Fresh pods plant$^{-1}$** | |

**Table.8** Promising RILs identified for multiple traits in a HACPI 6 population

| Identity of RILs | Traits |
|---|---|
| **RIL 6-262** | Raceme bearing branches plant$^{-1}$, raceme length and racemes plant$^{-1}$. |
| **RIL 6-278** | Raceme bearing branches plant$^{-1}$ , raceme length and racemes plant$^{-1}$ . |
| **RIL 6-310** | Days to 50% flowering, raceme bearing branches plant$^{-1}$, racemes plant$^{-1}$ and 100- seed weight. |
| **RIL 6-356** | Raceme bearing branches plant$^{-1}$,raceme length racemes plant$^{-1}$ and 100- seed weight. |
| **RIL 6-364** | Raceme bearing branches plant$^{-1}$, fresh pod plant$^{-1}$ and fresh pod yield. |
| **RIL 6-366** | Raceme bearing branches plant$^{-1}$, fresh pod plant$^{-1}$, and fresh pod yield plant$^{-1}$. |
| **RIL 6-367** | Fresh pod plant$^{-1}$, fresh pod yield plant$^{-1}$ and fresh seed yield plant$^{-1}$. |

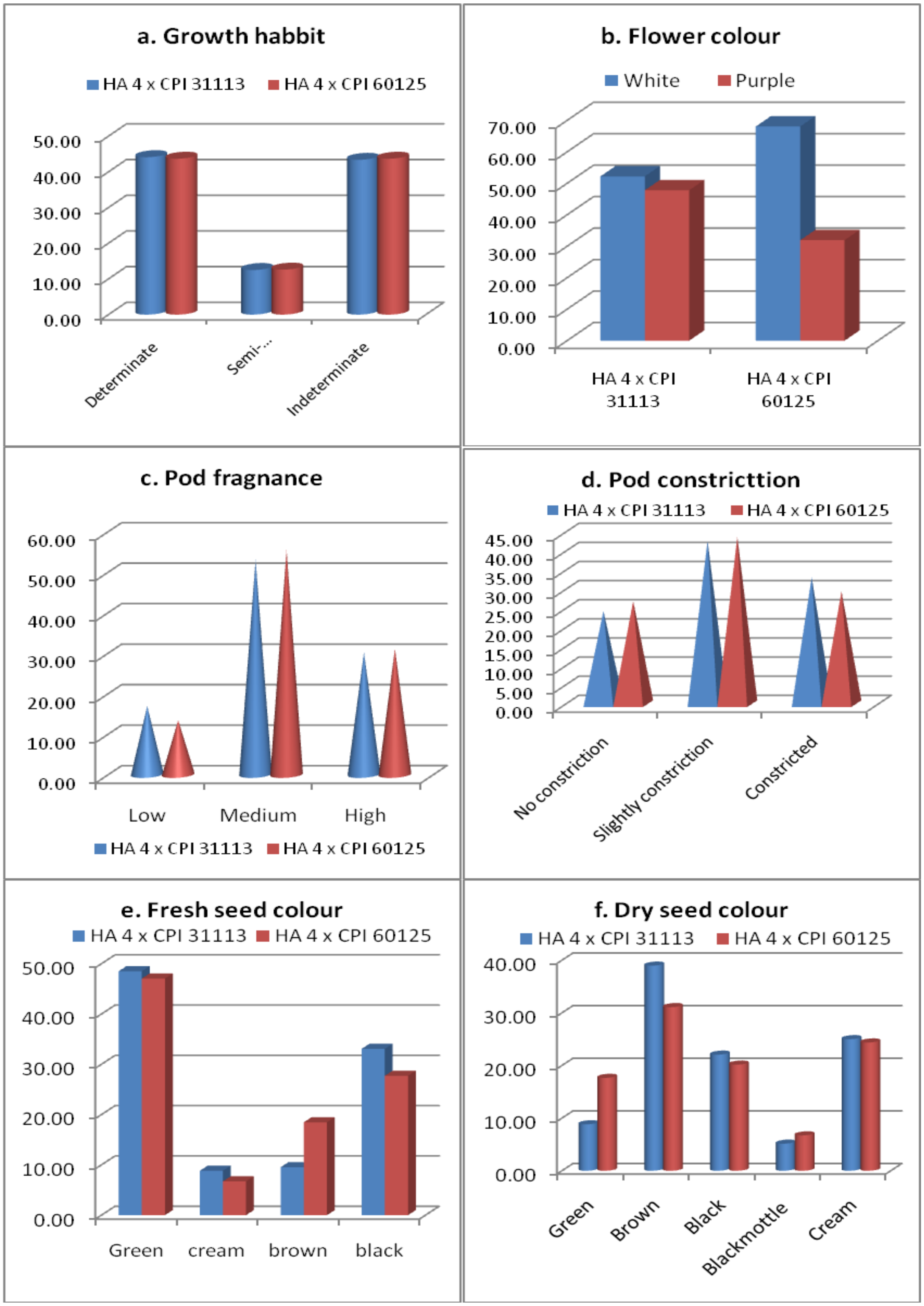
**Table.9** Estimates of quantitative traits means of the HACPI 6-derived RILs promising for multiple traits

| Identity of RILs | DFF | RBP | RL | RP | FP | FPY | FSY | DSY | TW |
|---|---|---|---|---|---|---|---|---|---|
| **RIL 6-262** | 65.50 | 18.04 | 17.26 | 15.36 | 19.30 | 16.78 | 10.23 | 7.19 | 11.96 |
| **RIL 6-278** | 71.94 | 17.95 | 18.39 | 12.95 | 24.41 | 41.09 | 19.76 | 12.35 | 10.65 |
| **RIL 6-310** | 41.78 | 23.22 | 12.73 | 10.54 | 33.89 | 57.71 | 23.95 | 14.66 | 22.90 |
| **RIL 6-356** | 69.65 | 19.31 | 16.67 | 12.60 | 20.03 | 51.98 | 28.49 | 14.19 | 25.62 |
| **RIL 6-364** | 92.82 | 13.81 | 16.30 | 9.34 | 52.85 | 71.40 | 28.97 | 10.90 | 14.59 |
| **RIL 6-366** | 85.07 | 31.27 | 10.90 | 8.30 | 51.39 | 112.35 | 34.48 | 11.96 | 15.01 |
| **RIL 6-367** | 73.01 | 12.14 | 12.58 | 9.68 | 55.03 | 74.05 | 41.07 | 20.45 | 16.36 |
| **CHECKS** | | | | | | | | | |
| **HA 4** | 45.16 | 9.95 | 15.41 | 9.73 | 35.53 | 56.05 | 29.13 | 20.01 | 20.56 |
| **HA 3** | 49.91 | 8.93 | 11.24 | 9.46 | 34.78 | 59.71 | 28.24 | 17.67 | 19.08 |
| **Kadalavare** | 64.58 | 17.39 | 16.47 | 10.16 | 43.34 | 70.94 | 35.67 | 27.25 | 21.35 |
| **Sem±** | 1.06 | 0.43 | 0.23 | 0.22 | 1.55 | 1.97 | 0.75 | 0.36 | 0.28 |
| **CD @ P=0.05** | 3.05 | 1.25 | 0.66 | 0.63 | 4.47 | 5.68 | 2.16 | 1.03 | 0.82 |

**Where,**

DFF- Days to 50% flowering
RBP- Raceme bearing branches plant$^{-1}$
RL-Raceme length (cm)
RP-Racemes plant$^{-1}$
FP- Fresh pods plant$^{-1}$
FPY- Fresh pod yield plant$^{-1}$ (g)
FSY Fresh seed yield plant$^{-1}$ (g)
DSY Dry seed yield plant$^{-1}$(g)
TW- 100- seed weight (g)

**Fig.1** Graph depicting frequencies of RILs with different states of growth habit, flower color, pod fragrance, pod constriction, fresh seed coat color, and dry seed coat color in two RIL populations derived from two bi-parental crosses in dolichos



The pods borne by majority of the RILs had medium fragrance (locally known as "Sogadu"), a highly preferred trait by farmers and consumers. The RILs (derived from both the crosses) with medium pod fragrance were more abundant than those with high and low fragrance (Figure 1c). Pod fragrance has been attributed to oily exudates that are reportedly composed of a mixture of fatty acids, of which trans-2-dodecenoic acid and tetra-dodecenoic acids are predominant (Fernandes and Nagendrappa, 1979; Udaykumar *et al.*, 2016).

Farmers prefer constricted pods, as they believe that cultivars that bear constricted pods have higher yielding ability than those that bear smooth pods, although there has been no documented experimental evidence about this belief (Vaijayanthi *et al.*, 2016). The consumer also prefers constricted pods with a belief that threshability of constricted pods is better than that of smooth pods. The RILs (derived from both the crosses) bearing slightly constricted pods were more frequent than those bearing constricted and absence of constricted pods (Figure 1d). (Vaijayanthi *et al.*, 2016) reported the abundance of germplasm accessions bearing slightly constricted pods than those bearing smooth pods in dolichos bean. RILs (derived from both the crosses) with green fresh seeds were more frequent than those with black, brown and cream fresh seeds (Figure 1e). The RILs bearing pods containing brown dry seeds were represented in higher frequency than those with cream, black and black mottled dry seeds (Figure 1f). Occurrence of higher frequency of green fresh seeds in the RIL populations could be useful as the farmer and end-users prefer cultivars bearing fresh pods with green seeds.

## Quantitative traits

REML analysis revealed highly significant mean squares attributable to "RILs", checks and "RILs vs. check varieties" for all traits in both the populations. These results suggested significant differences among the RILs and between RILs and checks, respectively. Mean squares attributable to checks vs. years were significant for all traits, except for days to 50% flowering, fresh pod plant$^{-1}$ in RILs (derived from both the crosses) and fresh seed yield plant$^{-1}$ in HACPI 3-derived RILs. The RILs derived from both the crosses interacted significantly with years for all the traits except for fresh pod yield plant$^{-1}$ in HACPI 3-derived RILs (Table 3). These results indicated differential performance of RILs and checks across years. The interactions of RILs with years implied that testing must be performed in multiple years for reliable results.

Genetic variability is a prerequisite for formulating appropriate selection strategies to develop improved dolichos bean varieties. The estimates of traitrange, one of the measures of trait variation provide clues about the occurrence of RILs with extreme expression. The standardized range of the RILs was higher for the raceme bearing branches plant$^{-1}$, raceme length, fresh pod plant$^{-1}$, fresh pod yield plant$^{-1}$, fresh seed yield plant$^{-1}$, dry seed yield plant$^{-1}$ and 100-seed weight compared to that for days to 50% flowering and raceme plant$^{-1}$which was amply reflected by the estimates of PCV in RILs derived from both the crosses (Table 4). The narrow differences in the estimates of PCV and GCV in the RILs derived from HACPI 3 and those derived from HACPI 6 suggested limited influence of environment in the expression of traits investigated (Table 4).

## Trait-specific RILs and those promising for multiple traits

Progress in crop genetic improvement and development of varieties with broad genetic base depends on the identification and use of new sources of genetic variation and superior genotypes for economically important traits. In the present study, some of the RILs were comparable to or superior to the checks HA 3, HA 4 and kadalavare with respect to nine traits, i.e., days to 50% flowering, raceme bearing branches plant$^{-1}$, racemes plant$^{-1}$, fresh pods plant$^{-1}$, fresh pod yield plant$^{-1}$, fresh seed yield plant$^{-1}$, dry seed yield plant$^{-1}$and 100-seed weight (Table 5). The RILs derived from HACPI 3, such as, 3-3, 3-26, 3-30, 3-35, 3-40, 3-95, 3-134, 3-139, 3-141 and 3-185 (Tables 6 and 7) were promising for multiple traits. The RILs derived from HACPI 6, such as, 6-262, 6-278, 6-310, 6-356, 6-364, 6-366 and 6-367, were promising for combinations of desirable traits (Tables 8 and 9). These RILs are also desirable for pod fragrance and constriction, the two most farmer/consumer-preferred qualitative traits. These RILs are suggested for preferential use in breeding high yielding dolichos bean varieties with desirable pod traits.

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