frontiers in Genetics

*Supplementary information*

# Genetic Architecture and Anthocyanin Profiling of Aromatic Rice from Manipur Reveals Divergence of *Chakhao* Landraces

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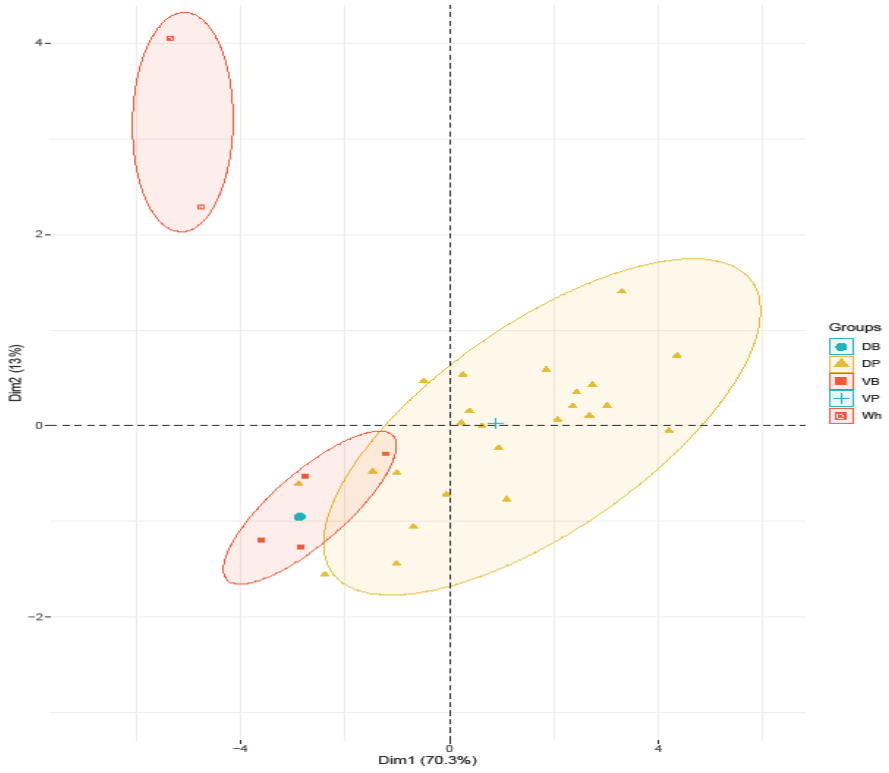


**Supplementary Figure S1.** Principal Component Analysis biplot of *Chakhao* rice genotypes from Manipur.

*Bhuvaneswari S, Gopala Krishnan S, Bollinedi H, Saha S, Ellur RK, Vinod KK, Singh IM, Prakash N, Bhowmick PK, Nagarajan M, Singh NK and Singh AK (2020) Genetic Architecture and Anthocyanin Profiling of Aromatic Rice From Manipur Reveals Divergence of Chakhao Landraces. Front. Genet. 11:570731.doi: 10.3389/fgene.2020.570731*

**Supplementary Table S1.** The aromatic rice germplasm from Manipur used in the present study

| Code | Accession No. | Local Name | District | Ecosystem | Pericarp colour | Aroma |
|---|---|---|---|---|---|---|
| MAR1 | IC0306333 | *Ching Chakhao Amubi* | Imphal east | Valley | Dark purple | SS |
| MAR2 | IC0461320 | *Chakhao* | NA | NA | White | MS |
| MAR3 | IC0461323 | *Chakhao Angenba* | NA | NA | White | MS |
| MAR4 | IC0461895 | *Chakhao Poireitol* | NA | NA | White | MS |
| MAR5 | IC0461922 | *Chakhao Poiretol* | NA | NA | White | MS |
| MAR6 | IC0464571 | *Chakhao Poireitol* | Thoubal | Valley | Dark purple | MS |
| MAR7 | IC0464574 | *Chakhao Poireitol* | Thoubal | Valley | Dark purple | SS |
| MAR8 | IC0464578 | *Chakhao* | Thoubal | Valley | White | MS |
| MAR9 | IC0464580 | *Chakhao* | Manipur Central | Valley | White | NS |
| MAR10 | IC0464583 | *Chakhao* | Manipur Central | Valley | Dark purple | SS |
| MAR11 | IC0464585 | *Chakhao* | Manipur Central | Valley | White | NS |
| MAR12 | IC0464590 | *Chakhao* | Thoubal | Valley | Light brown | SS |
| MAR15 | IC0465318 | *Chakhao* | Imphal west | Valley | Dark purple | SS |
| MAR16 | IC0465365 | *Chakhao Arongbi* | Thoubal | Valley | White | MS |
| MAR17 | IC0465376 | *Chakhao* | Manipur Central | Valley | Dark brown | MS |
| MAR18 | IC0467090 | *Kabo Chakhao* | Imphal East | Valley | Light brown | MS |
| MAR19 | IC0467095 | *Chakhao Angouba* | Imphal East | Valley | White | NS |
| MAR21 | IC0467110 | *Kabo Chakhao* | Manipur Central | Valley | White | MS |
| MAR22 | IC0467147 | *Chakhao Sempak* | Imphal East | Valley | Light brown | MS |
| MAR24 | IC0467155 | *Chakhao* | Manipur Central | Valley | Light brown | MS |
| MAR25 | IC0467157 | *Chakhao* | Manipur Central | Valley | Light brown | MS |
| MAR26 | IC0467169 | *Chakhao Poireitol* | Thoubal | Valley | White | MS |
| MAR27 | IC0467170 | *Chakhao* | Manipur Central | Valley | Light brown | MS |
| MAR28 | IC0467175 | *Chakhao* | Thoubal | Valley | White | SS |
| MAR30 | IC0596559 | *Chakhao Poireiton* | Imphal west | Valley | Dark purple | MS |
| MAR31 | IC0596560 | *Chakhao Angouba* | Imphal west | Valley | White | NS |
| MAR32 | IC0596564 | *Chakhao* | Senapati | Valley | White | NS |
| MAR33 | IC0596565 | *Chakhao Poireiton* | Thoubal | Valley | Dark purple | MS |
| MAR34 | IC0596566 | *Chakhao Amubi* | Thoubal | Valley | Dark purple | NS |
| MAR35 | IC0596567 | *Chakhao Poireiton* | Thoubal | Valley | Dark purple | MS |
| MAR36 | IC0596568 | *Chakhao Pungdol Angouba* | Thoubal | Valley | Dark purple | MS |
| MAR37 | IC0596569 | *Chakhao Poireiton* | Thoubal | Valley | White | SS |
| MAR38 | IC0596570 | *Chakhao Amubi* | Thoubal | Valley | White | NS |
| MAR39 | IC0596571 | *Chakhao Angouba* | Thoubal | Valley | White | NS |
| MAR40 | IC0596573 | *Chakhao Amubi* | Chandel | Hill | Variegated brown | SS |
| MAR41 | IC0596574 | *Chakhao Angouba* | Chandel | Hill | White | MS |
| MAR42 | IC0596585 | *Chakhao Phou* | Tamenglong | Hill | Light brown | MS |
| MAR43 | IC0596586 | *Chakhao Phou* | Tamenglong | Hill | White | - |
| MAR44 | IC0596587 | *Chakhao Poireiton* | Bishnupur | Valley | Dark purple | MS |
| MAR45 | IC0596588 | *Chakhao* | Bishnupur | Valley | Dark purple | NS |
| MAR46 | IC0596589 | *Chakhao Amubi* | Bishnupur | Valley | Variegated brown | MS |
| MAR47 | IC0596590 | *Chakhao Poireiton* | Bishnupur | Valley | White | NS |
| MAR48 | IC0350432 | *Buhman* | Churachandpur | Hill | White | MS |
| MAR49 | IC0350433 | *Buhman - A* | Churachandpur | Hill | Light brown | MS |
| MAR50 | IC0350434 | *Buhman Kungtum* | Churachandpur | Hill | Light brown | MS |
| MAR51 | IC0350436 | *Buhman Te* | Churachandpur | Hill | Light brown | NS |
| MAR52 | IC0350437 | *Buhman Tum* | Churachandpur | Hill | Light brown | MS |
| MAR53 | IC0350588 | *Sang Buhman* | Churachandpur | Hill | Light brown | MS |
| MAR54 | IC0591271 | *Buhman* | Churchandpur | Hill | Light brown | MS |
| MAR55 | IC0591272 | *Sang Buhman* | Churchandpur | Hill | White | NS |
| MAR56 | IC0596576 | *Buhman (Ch. Angouba)* | Churachandpur | Hill | White | NS |
| MAR57 | IC0596577 | *Buhman* | Churachandpur | Hill | Light brown | MS |
| MAR58 | IC0596578 | *Buhman* | Churachandpur | Hill | Light brown | MS |
| MAR59 | IC0596579 | *Buhman* | Churachandpur | Hill | White | MS |
| MAR60 | IC0596580 | *Buhman* | Churachandpur | Hill | Light brown | MS |
| MAR61 | IC0596581 | *Buhman/ Idaw* | Churachandpur | Hill | Light brown | MS |

| Code | Accession No. | Local Name | District | Ecosystem | Pericarp colour | Aroma |
|---|---|---|---|---|---|---|
| MAR62 | IC0596582 | *Buhman* | Churachandpur | Hill | White | NS |
| MAR63 | IC0596591 | *Maklei* | Ukhrul | Hill | Variegated brown | MS |
| MAR64 | IC0596592 | *Maklei* | Ukhrul | Hill | Dark purple | MS |
| MAR65 | IC0596593 | *Maklei* | Ukhrul | Hill | Light brown | MS |
| MAR66 | IC0596594 | *Maklei* | Ukhrul | Hill | Light brown | MS |
| MAR67 | IC0596583 | *Napnang Hangmei* | Tamenglong | Hill | White | MS |
| MAR68 | IC0596584 | *Napnang Hangmei* | Tamenglong | Hill | Light brown | SS |
| MAR69 | SB/PB 1 | *Chakhao Amubi* | Imphal East | Valley | Dark purple | MS |
| MAR70 | SB/SKS 1 | *Chakhao Poireiton* | Imphal East | Valley | Dark purple | SS |
| MAR75 | SB/LM 1 | *The Vumnu* | Chandel | Hill | Dark purple | SS |
| MAR76 | SB/LM 2 | *The Vumnu* | Chandel | Hill | Dark purple | MS |
| MAR77 | SB/LM 3 | *Ethe Buw* | Chandel | Hill | Variegated brown | MS |
| MAR78 | SB/SS 1 | Black rice | Imphal east | Valley | Variegated Purple | MS |
| MAR79 | SB/IMS 1 | *Buhman* | Churachandpur | Hill | Dark purple | MS |
| MAR80 | SB/IMS 2 | *Chakhao Kumbi* | NA | Valley | Light brown | MS |
| MAR81 | SB/IMS 3 | *Chakhao Poireiton* | NA | Valley | White | SS |
| MAR82 | SB/IMS 4 | *Chakhao Amubi* | NA | Valley | Dark purple | SS |
| MAR83 | SB/PB 2 | *Langphou Chakhao* | NA | Valley | Dark purple | MS |
| MAR84 | SB/IMS 5 | *Chakhao* | Tamenglong | Hill | Light brown | MS |
| MAR85 | SB/RKY 1 | *Chakhao* | Ukhrul | Hill | Dark purple | SS |
| MAR86 | SB/CT 1 | *Ching Chakhao* | Imphal east | Valley | Dark purple | SS |
| MAR87 | IC0352915 | *Chakhao Poireiton* | Thoubal | Valley | Variegated brown | MS |
| MAR88 | IC0467085 | *Chakhao* | Imphal East | Valley | White | MS |
| MAR89 | IC0467146 | *Chahou Poireiton* | Thoubal | Valley | White | MS |
| MAR91 | IC0521521 | *Chakhao* | Imphal | Valley | Dark purple | SS |
| MAR93 | IC0467102 | *Kabo Chakhao* | Manipur Central | Valley | White | MS |
| MAR94 | IC0350440 | *Chakhao Amubi* | NA | Valley | White | MS |
| MAR97 | IC0352872 | *Chakhao Amuba* | Imphal East | Valley | Dark purple | SS |
| MAR98 | IC0352914 | *Chakhao Angangbi* | Thoubal | Valley | White | MS |
| MAR99 | IC0352917 | *Chakhao Huikap* | Thoubal | Valley | Dark Purple | SS |
| MAR101 | IC0591277 | *Chakhao* | East Imphal | Valley | Dark Purple | MS |
| MAR102 | IC0591278 | *Chakhao* | East Imphal | Valley | White | SS |
| MAR103 | IC0306379 | *Chakhao Amubi* | Bishnupur | Valley | Variegated Purple | SS |
| MAR105 | IC0306382 | *Chakhao Poireiton* | Bishnupur | Valley | White | SS |
| MAR106 | IC0350441 | *Chahao Angouba* | NA | Valley | Light brown | MS |
| MAR108 | IC0297667 | *Chakhao* | Imphal East | Valley | Light Brown | MS |
| MAR109 | IC0306383 | *Chakhao Angouba* | Bishnupur | Valley | White | MS |

SS, strongly scented; MS, mild scented; NS, unscented; NA, data not available

**Supplementary Table S2.** Principal component analysis of agronomic and grain quality data a showing most significantly contributing traits (data shows first ten principal components)

| Statistics | PC1 | PC2 | PC3 | PC4 | PC5 | PC6 | PC7 | PC8 | PC9 | PC10 |
|---|---|---|---|---|---|---|---|---|---|---|
| Standard deviation | 2.31 | 1.58 | 1.32 | 1.12 | 1.07 | 0.99 | 0.94 | 0.80 | 0.73 | 0.68 |
| Proportion of Variance | 0.31 | 0.15 | 0.10 | 0.07 | 0.07 | 0.06 | 0.05 | 0.04 | 0.03 | 0.03 |
| Cumulative Proportion | **0.31** | **0.46** | **0.56** | **0.64** | **0.70** | 0.76 | 0.81 | 0.85 | 0.88 | 0.91 |
| Eigenvalues | **5.32** | **2.50** | **1.73** | **1.25** | **1.14** | 0.98 | 0.88 | 0.64 | 0.53 | 0.46 |
| Ligule colour (LG) | **-0.41** | 0.04 | 0.05 | -0.07 | -0.08 | 0.08 | -0.14 | 0.05 | -0.09 | 0.01 |
| Auricle colour (AU) | **-0.39** | 0.07 | 0.11 | -0.04 | -0.08 | 0.01 | -0.06 | -0.07 | 0.11 | 0.07 |
| Collar colour (CO) | **-0.39** | 0.04 | 0.05 | -0.08 | -0.03 | 0.00 | -0.12 | -0.17 | 0.10 | -0.03 |
| Basal leaf colour (BL) | **-0.33** | 0.07 | 0.06 | -0.12 | -0.13 | -0.03 | 0.09 | -0.08 | 0.71 | -0.07 |
| Grain colour (LP)* | **-0.38** | 0.08 | 0.03 | -0.02 | 0.07 | 0.05 | 0.06 | 0.06 | -0.31 | -0.09 |
| Pericarp colour (PC) | **-0.38** | 0.04 | 0.09 | -0.07 | -0.03 | 0.11 | -0.08 | 0.12 | -0.36 | -0.17 |
| Flowering days (DF) | -0.06 | -0.29 | 0.11 | -0.27 | -0.42 | -0.47 | 0.02 | 0.48 | -0.07 | 0.41 |
| Plant height (PH) | -0.19 | **-0.41** | 0.18 | 0.11 | 0.27 | 0.05 | 0.21 | -0.01 | -0.25 | -0.08 |
| Panicle number (PN) | -0.07 | -0.31 | -0.12 | -0.24 | 0.50 | -0.29 | -0.31 | -0.33 | 0.09 | 0.32 |
| Panicle length (PL) | -0.10 | -0.22 | 0.32 | 0.39 | 0.40 | 0.20 | 0.20 | 0.33 | 0.25 | 0.29 |
| Grain weight (GW) | 0.09 | -0.17 | 0.52 | 0.05 | -0.25 | -0.08 | -0.01 | -0.63 | -0.17 | 0.09 |
| Grain yield (GY) | 0.15 | -0.26 | 0.22 | -0.48 | 0.24 | -0.03 | -0.21 | 0.21 | 0.12 | -0.59 |
| Kernel length (KL) | 0.08 | -0.21 | 0.12 | 0.32 | -0.24 | 0.31 | -0.77 | 0.14 | 0.09 | 0.02 |
| Kernel width (KW) | 0.17 | 0.06 | 0.57 | -0.11 | -0.11 | 0.05 | 0.23 | 0.05 | 0.09 | -0.11 |
| Alkali spreading (AS) | -0.10 | -0.07 | -0.03 | 0.57 | -0.03 | -0.66 | -0.04 | 0.01 | 0.06 | -0.44 |
| Gel consistency (GC) | 0.07 | **0.44** | 0.27 | -0.04 | 0.28 | -0.25 | -0.24 | 0.15 | -0.18 | 0.14 |
| Amylose content (AC) | -0.01 | **-0.49** | -0.27 | -0.03 | -0.19 | 0.15 | 0.17 | -0.04 | -0.07 | -0.08 |

*Pigmentation of lemma and palea; The most contributing traits are indicated in red bold

**Supplementary Table S3.** Major allele frequency, allele number, gene diversity and polymorphic information content (PIC) of SSR markers used in the study

| SSR marker | Chr | Major allele frequency | Number of alleles | Gene diversity | PIC |
|---|---|---|---|---|---|
| RM495 | 1 | 0.557 | 2 | 0.494 | 0.372 |
| RM1 | 1 | 0.835 | 5 | 0.293 | 0.280 |
| RM283 | 1 | 0.602 | 3 | 0.502 | 0.403 |
| RM259 | 1 | 0.660 | 4 | 0.500 | 0.442 |
| RM312 | 1 | 0.888 | 2 | 0.199 | 0.179 |
| RM5 | 1 | 0.567 | 4 | 0.538 | 0.447 |
| RM237 | 1 | 0.786 | 3 | 0.360 | 0.331 |
| RM431 | 1 | 0.633 | 4 | 0.492 | 0.406 |
| RM154 | 2 | 0.367 | 4 | 0.671 | 0.600 |
| RM452 | 2 | 0.526 | 3 | 0.517 | 0.403 |
| RM489 | 3 | 0.856 | 3 | 0.250 | 0.223 |
| OSR13 | 3 | 0.704 | 3 | 0.433 | 0.362 |
| RM338 | 3 | 0.867 | 2 | 0.230 | 0.204 |
| RM55 | 3 | 0.786 | 3 | 0.355 | 0.322 |
| RM514 | 3 | 0.713 | 3 | 0.438 | 0.383 |
| RM307 | 4 | 0.408 | 6 | 0.733 | 0.694 |
| RM124 | 4 | 0.898 | 2 | 0.183 | 0.166 |
| RM507 | 5 | 0.831 | 3 | 0.284 | 0.249 |
| RM413 | 5 | 0.316 | 7 | 0.774 | 0.741 |
| RM161 | 5 | 0.889 | 2 | 0.198 | 0.178 |
| RM178 | 5 | 0.908 | 3 | 0.168 | 0.157 |
| RM334 | 5 | 0.446 | 6 | 0.646 | 0.577 |
| RM510 | 6 | 0.735 | 3 | 0.425 | 0.386 |
| RM162 | 6 | 0.786 | 3 | 0.341 | 0.289 |
| RM454 | 6 | 0.959 | 2 | 0.078 | 0.075 |
| RM125 | 7 | 0.939 | 2 | 0.115 | 0.108 |
| RM11 | 7 | 0.704 | 5 | 0.462 | 0.419 |
| RM455 | 7 | 0.867 | 2 | 0.230 | 0.204 |
| RM118 | 7 | 0.755 | 2 | 0.370 | 0.301 |
| RM408 | 8 | 0.418 | 4 | 0.661 | 0.591 |
| RM152 | 8 | 0.418 | 4 | 0.654 | 0.587 |
| RM25 | 8 | 0.629 | 5 | 0.558 | 0.520 |
| RM44 | 8 | 0.800 | 3 | 0.340 | 0.313 |
| RM284 | 8 | 0.885 | 2 | 0.203 | 0.182 |
| RM433 | 8 | 0.515 | 2 | 0.500 | 0.375 |
| RM447 | 8 | 0.755 | 4 | 0.400 | 0.365 |
| RM316 | 9 | 0.737 | 4 | 0.425 | 0.389 |
| RM105 | 9 | 0.469 | 3 | 0.587 | 0.498 |
| RM215 | 9 | 0.753 | 3 | 0.401 | 0.364 |
| RM474 | 10 | 0.560 | 6 | 0.624 | 0.583 |
| RM271 | 10 | 0.531 | 4 | 0.644 | 0.599 |
| RM171 | 10 | 0.804 | 3 | 0.322 | 0.282 |
| RM552 | 11 | 0.206 | 7 | 0.826 | 0.802 |
| RM536 | 11 | 0.857 | 3 | 0.252 | 0.233 |
| RM287 | 11 | 0.427 | 4 | 0.632 | 0.560 |
| RM144 | 11 | 0.306 | 7 | 0.785 | 0.753 |
| RM19 | 12 | 0.541 | 4 | 0.614 | 0.556 |
| RM277 | 12 | 0.495 | 3 | 0.555 | 0.455 |
| **Mean** | | **0.664** | **3.563** | **0.443** | **0.394** |

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Chr, chromosome; PIC, polymorphism information content

**Supplementary Table S4.** Retention time of different peaks found in the anthocyanin rich black rice extract (ABRE) of samples and known standards

| | **Peak 1** | **Peak 2** | **Peak 3** | **Peak 4** |
|---|---|---|---|---|
| Retention time (min) | 10.97 | 11.56 | 11.95 | 12.9 |
| Compounds | Unknown | Unknown | Cyanidin-3-O-glucoside | Peonidin-3-O-glucoside |