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# ASSESSMENT OF GENETIC DIVERSITY AMONG SUWANDAL RICE (*ORYZA SATIVA* L.) ACCESSIONS BASED ON MORPHOLOGICAL, MOLECULAR AND PHYSICOCHEMICAL CHARACTERISTICS

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## ABSTRACT

**An investigation was carried out to determine the extent of diversity and relationship among 14 Suwandal rice accessions collected from different parts of the country. Trial was established in the field in *yala* 2015 and *maha* 15/16. Forty three qualitative and quantitative morphological traits, six grain quality parameters, thirty SSR marker loci were used to analyze the diversity among these accessions. Considerable morphological diversity was observed in maturity, stem, leaf, panicle, floral and grain characters. These accessions grouped into four clusters according to the morphological analysis. Total number of alleles detected by 30 SSR markers was 82 and allele richness was ranged from 1-5. Out of 30 SSR loci 28 loci showed polymorphism across the tested accessions. Polymorphic Information Content varies from 0 to 0.71which underlined high genetic diversity among Suwandal accessions. Similar clustering pattern was observed in both morphological and molecular characterized data. AC11340, AC13300 & AC12827 grouped together both in morphological and molecular grouping and AC04197, AC04595 & AC004802 made another cluster. Significant variations were observed in gel consistency, antioxidant activity and gelatinization temperature among these**

**tested accessions. No duplicates were identified. Information generated through diversity analysis based on phenotypic genotypic data and physicochemical analysis could be efficiently used to identify the different of Suwandal accessions.**

**Keywords: Diversity, Molecular, Morphological, Physicochemical properties, Suwandal, Traditional rice**

## INTRODUCTION

Rice (*Oryza sativa* L.) is the world's single most important food crop and a primary food source for more than a third of the world's population (Khush,1997). It was originated and domesticated in tropical Asia and the oldest record dating 5000 years BC. According to the documentary evidences rice has a long history in Sri Lanka extending into pre-historic time (Rajapakshe, 1997). Cultivated rice is rich in genetic diversity therefore it has broad adaptation to a wide range of growing environments. Consequently thousands of rice varieties emerged all over the world.

Prior to green revolution rice cultivation in Sri Lanka was totally depend on traditional cultivars. However, with the increasing population and low productivity of traditional cultivars attention was paid to develop improved rice varieties. Nowadays with the extensive use of modern varieties, many traditional crop varieties have been replaced worldwide by a few improved high yielding varieties resulted narrowing down the crop diversity and it is leading to serious genetic erosion. As a precautionary measure, substantial crop diversity is being conserved in the gene banks around the world. At present, Gene bank of Plant Genetic Resources Centre (PGRC), Sri Lanka collected and conserves two thousand four hundred seven traditional rice accessions under the six hundred eighty four different cultivars names (gene bank information). Recently demand for traditional rice cultivars have been increased and consequently extend of cultivation also considerably increased. This is mainly due to the high consumer demand for quality rice. Different historical records revealed that traditional rice varieties were revered for their nutritional and medicinal properties. Therefore, according to the farmer preference and consumer demand seven traditional rice cultivars have been identified for cultivation. Those are *Suwandal, Kuruluthudawee, Pachchaperumal, Suduru samba, Kaluheenati,*

*Madathawalu* and *Pokkali*. However, due to poor quality seeds and large number of accessions coming under one cultivar name correct identification and quality seed paddy production with identical qualities are prerequisite.

*Suwandal* is one of the popular traditional rice cultivars with a special kind of aroma and taste. It's nutrient composition consists of 90% of starch, 7% of crude protein, 0.7% of crude fat, 0.1% of crude fiber, 0.9% of Ca, 2% of Fe and trace amount of vitamin B ( Liyanarachchi *et al.*, 2014). Rural Enterprise Network (2014) (http://www.rensrilanka.org/tradrice.html), reported that *suwandal* is capable of promoting fair and glowing skin, improving the function of the excretory system, improving vocal clarity and helps to control diabetes.

At present 14 rice accession under the name of *Suwandal* were explored from different parts of the country and conserved at PGRC genebank. However, systematic studies hence not conducted to study the diversity among there accessions and identify the repetitions. Therefore, present study was conducted to characterize these accessions using morphological, molecular and physicochemical characters to identify the available diversity, and identify *suwandal* accessions with identical characters for quality seed production and popularize among farmers.

## MATERIALS AND METHOD

**Morphological characterization-** Fourteen Suwandal rice accessions which conserved at seed gene bank of PGRC were used for the study (Table 1). Upland nurseries were established and twenty one days old seedlings were transplanted in the field at PGRC during *yala* 2015 and *maha* 2015/16. Plot size was 2 X 4 m. Spacing was 20 X 10 cm. Except fertilization crop management practices were done according to the recommendations of Department of Agriculture. Twenty six qualitative and seventeen quantitative morphological data were recorded at vegetative stage, flowering stage and at maturity according to the standard descriptor for rice (PGRC, 1995). These data comprise eight panicle characters, twelve grain characters, six culm characters, thirteen leaf characters and four growth characters. Internode colour, culm angle after flowering and culm strength were evaluated under stem characters. The panicle type at near maturity,

panicle exertion, panicle axis at maturity, panicle shattering, panicle threshability were the observed characters under panicle characters. Anther colour and stigma colour were evaluated under floral characters. Except grain characters, data were recorded from randomly selected 10 plants from each plot. Grain characters were recorded from the randomly selected seed lot from the bulk harvest of each accession.

**Table 1. Details of Suwandal rice accessions used for the study**

| Sample Name | Accession Number | Name | Collection district |
|---|---|---|---|
| 1 | AC 12827 | Suwandal | Unknown |
| 2 | AC 04197 | Suwandal | Unknown |
| 3 | AC 05420 | Suwandal | CRBS |
| 4 | AC 10646 | Suwandal | Anuradhapura |
| 5 | AC 12844 | Suwandal | Moneragala |
| 6 | AC 11088 | Suwandal | Kegalle |
| 7 | AC 04595 | Suwandal | Unknown |
| 8 | AC 10729 | Suwandal | Anuradhapura |
| 9 | AC 13136 | Suwandal | PGRC selection |
| 10 | AC 11340 | Suwandal | Matale |
| 11 | AC 04802 | Suwandal | Unknown |
| 12 | AC 04471 | Suwandal | Unknown |
| 13 | AC 04366 | Suwandal | Unknown |
| 14 | AC 13300 | Suwandal | Kalutara |

**Molecular characterization-** DNA was extracted from two weeks old immature leaves of 14 *Suwandal* accessions using CTAB method described by Murry and Thomson (1980) with minor modifications. A total of 30 microsatellite markers were used for genotyping (Table 2).

Primer amplification was done using Polymerase Chain Reaction (PCR). The PCR reaction mixture and PCR program was done according to the Wasala *et al*, (2014). After confirmation of PCR products in 0.8% agarose, products were resolved in 8 % (Acrilamide: Bis Acrilamide-29:1) non denaturing polyacrylamide gel electrophoresis followed by Ethidium Bromide (0.5 ug/ml) staining. Bands were visualized using gel documentation system (BIO RAD).

**Table 2. Data on Number of Alleles, Major Allele Frequencies, Genetic Diversity and Polymorphic**

Information Content (PIC) found among 14 Suwandel Rice Accessions

| Primer name | Major Allele Frequency | Genotype No | Allele richness | Gene Diversity | PIC* |
|---|---|---|---|---|---|
| RM237 | 0.71 | 2 | 2 | 0.41 | 0.32 |
| RM 25 | 0.52 | 5 | 3 | 0.61 | 0.54 |
| RM 259 | 0.71 | 2 | 2 | 0.41 | 0.32 |
| RM 536 | 0.62 | 3 | 2 | 0.47 | 0.36 |
| RM 201 | 0.58 | 2 | 2 | 0.49 | 0.37 |
| RM 219 | 0.57 | 2 | 2 | 0.49 | 0.37 |
| RM 208 | 0.61 | 3 | 2 | 0.48 | 0.36 |
| RM 224 | 0.82 | 3 | 2 | 0.29 | 0.25 |
| RM 241 | 0.64 | 2 | 2 | 0.46 | 0.35 |
| RM 480 | 0.53 | 4 | 3 | 0.57 | 0.48 |
| RM 412 | 0.92 | 2 | 2 | 0.14 | 0.13 |
| RM 228 | 1.00 | 1 | 1 | 0.00 | 0.00 |
| RM 220 | 0.69 | 4 | 4 | 0.49 | 0.45 |
| RM 207 | 0.62 | 2 | 2 | 0.47 | 0.36 |
| RM 216 | 0.86 | 2 | 2 | 0.24 | 0.21 |
| RM 236 | 0.82 | 3 | 2 | 0.29 | 0.25 |
| RM 571 | 0.50 | 2 | 2 | 0.50 | 0.38 |
| RM 255 | 1.00 | 1 | 1 | 0.00 | 0.00 |
| RM 20b | 0.79 | 2 | 2 | 0.34 | 0.28 |
| RM 217 | 0.42 | 4 | 4 | 0.68 | 0.62 |
| RM213 | 0.76 | 3 | 3 | 0.39 | 0.35 |
| RM202 | 0.33 | 5 | 5 | 0.75 | 0.71 |
| RM560 | 0.86 | 2 | 2 | 0.24 | 0.21 |
| RM84 | 0.47 | 3 | 3 | 0.58 | 0.49 |
| RM539 | 0.43 | 4 | 4 | 0.70 | 0.65 |
| RM515 | 0.63 | 3 | 3 | 0.53 | 0.48 |
| RM418 | 0.50 | 3 | 3 | 0.62 | 0.55 |
| RM440 | 0.32 | 7 | 4 | 0.74 | 0.69 |
| RM215 | 0.43 | 3 | 3 | 0.65 | 0.58 |
| RM518 | 0.93 | 2 | 2 | 0.13 | 0.12 |
| RM270 | 0.57 | 3 | 3 | 0.54 | 0.45 |
| Mean | 0.65 | 3 | 3 | 0.44 | 0.38 |

*PIC-Polymorphic Information Content

Physical and physicochemical quality characterization- This study was done at grain quality laboratory at Rice Research and Development Institute, Batalagoda. Brown rice % (BR), Total milling % (TM), Head grain % (HG), Amaylose content (AC), gel consistency (GC) and gelatinization temperature (GT) was evaluated according to the Cruz & Khush (2000) and Rebeira *et al* (2014). Antioxidant activity was measured based on free radical scavenging ability of rice grain extracts according to Sutharut & Sudarat (2012).

**Data analysis-** Cluster analysis were done to investigate the morphological diversity and group these accessions according to their similarities using qualitative and quantitative morphological data using MINITAB var. 17 software. Principle Component Analysis (PCA) was done for quantitative data using MINITAB var 17.

Molecular data-The banding pattern for each primer was scored using Quantity One 4.6. Data analysis was done using POWERMARKER V 3.25 software ( Liu *et al.*, 2005) to estimate the genetic diversity of each accession. Genetic distances were calculated according to the Nie's genetic distance (Nie & Chesser, 1983). These data were used to construct a phylogenetic tree based on Unweighted Paired Group Method with Arithmetic Average (UPGMA) algorithm.

Physical and physicochemical data- Analysis of variance was performed for amylose content, free radical scavenging activity and gel consistency using MINITAB var 17. Square root transformation was done for gel consistency data prior to analysis. Mean separation was done according to the Turkey's T test.

## RESULTS AND DISCUSSIONS

### Morphological characterization

**Qualitative characters-** Similar qualitative characters were observed in both seasons. Therefore, one season (*yala* 2015) were used to analyze the characteristics. Out of 44 different plant characters nine characters were uniform among the tested accessions. Those are variety group (indica), ligule colour (absent), collar colour (pale green), internode colour (green), panicle type (compact), secondary branching in panicle (heavy), lemma

and pelea pubescence (short hairs), sterile lemma colour (straw) and endosperm type (non-glutinous). Highly diverse morphological characters were summarized in Table 3.

**Stem characters-** Two different patterns of culm angles were observed. Twelve accessions were showed erect (the angle is less than 30° from the perpendicular) while accession numbers AC12827 and AC 04197 showed intermediate pattern which the angle is about 45°. Culm strength ie. lodging resistance varied among the accessions. AC13136 showed intermediate culm strength, AC05420, AC11340, AC04366 and AC 013300 showed moderately strong culm strength and rest of the accessions showed strong culm strength.

**Leaf characters-** Two accessions (AC05420, AC13136) showed glabrous pubescence and other 12 accessions showed intermediate pubescence on leaf blade. Four accessions (AC12827, AC12844, AC04595 & AC13300) displayed erect leaf angle and others were displayed intermediate angle. Shape of the ligule is an important character for variety identification. In these tested accessions thirteen accessions had an acute to acuminate shaped ligule and one accession (AC12844) had cleft ligule.

**Panicle Characters-** All the accessions displayed compact panicle type and heavy secondary branching pattern. Three accessions (AC11088, AC04366 and AC13136) displayed droopy panicle axis at maturity and others displayed straight axis.

**Floral characters-** Eight accessions were displayed purple colour stigma while 6 accessions displayed yellow colour stigma. Two basic anther colours were recorded as 9 accessions with white colour anthers and 5 accessions with light yellow colour anthers.

**Seed Characters-** This is one of the main characters to differentiate different rice accessions. One accession (AC12844) showed an awn after full heading. Two apiculus colours were observed, 5 accessions were brown in colour and other 9 accessions were straw in colour. Four distinct lemma and palea colours were observed. Those are brown, straw, brown furrows on straw and brown spots on straw were observed (Figure 1& Table 3).

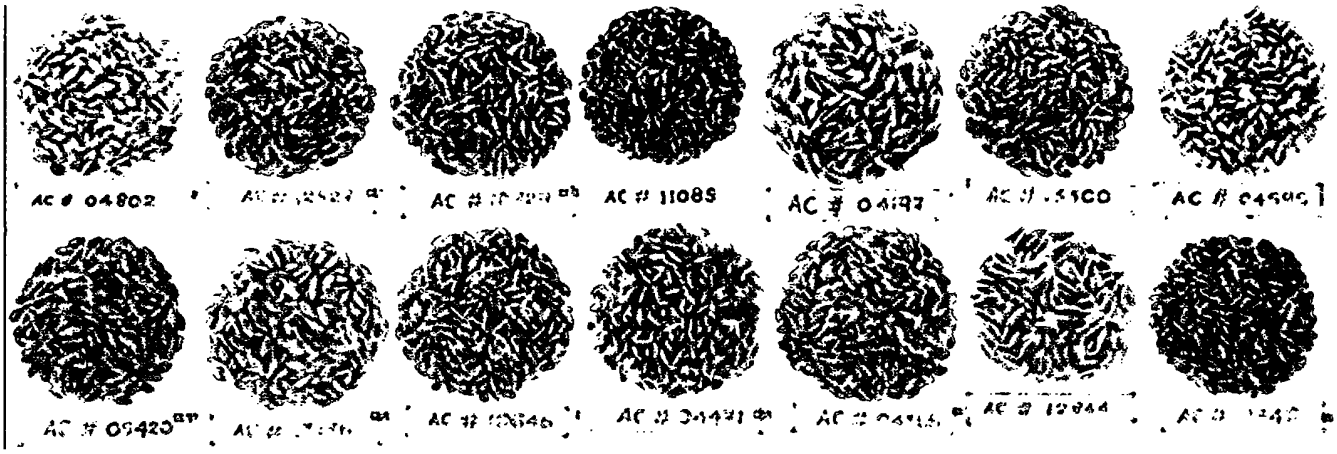

**Figure 1, Different types of lemma and palea colour at maturity of tested Suwandal accessions**

Considerable variation of seed coat colours were shown. Six accessions showed white colour (AC12827, AC10646, AC11088, AC 10729, AC04471, AC13300), five accessions showed brown colour ( AC04197, AC05420, AC 12844, AC11340, AC04366) and three accessions showed brown colour seed coat (AC04802, AC13136, AC04595).

**Quantitative characters** - Number of days to flowering ranged from 86 days to 119 days. Six accessions came to flowering after 86-87 days from planting. Four accessions came to flowering 94-95 days after planting. Rest of the accessions came to flowering 115-119 days after planting. In the same way six accessions matured within 114 to 118 days (3.5 months) from planting. Four accessions matured within 123 to 125 days (4-4.5 months) from planting and rest of the accessions came to maturity 151 to 155 days (5 months) from planting (Table 3). It revealed that these *Suwandal* accessions belong to the different age groups at maturity.

Ligule length of these accessions showed considerable variation from1.38 mm (AC 10729) to 16.4 mm (AC 12844). Distinct variations were observed in plant height of these tested accessions (Table 3). It ranged from 57.08 cm (AC12827) to 91.44 cm (AC04595). The mean culm number at full heading varied from 13 (AC4595) to 4 (AC10646). The 100 grain weight was the highest in AC12844 with 2.61 g and minimum in AC10729 with 1.21 g. Grain length varyed from 5.8 mm (AC12844) to 8.6 mm (AC04366). Grain width ranged from 2.43 mm (AC04802) to 3.39 mm (AC10646).

**Table 3 Morphological characteristic data of tested Suwandal accessions based on *yala* 2015 trial**

| Accession no. | AC 12827 | AC 4197 | AC 5420 | AC 10646 | AC 12844 | AC 11088 | AC 4595 | AC 10729 | AC 13136 | AC 11340 | AC 4802 | AC 4471 | AC 4366 | AC 13300 |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Leaf blade length (cm) | 35.4 | 17.67 | 29.56 | 24.75 | 47.38 | 30.08 | 42.76 | 30 | 26.48 | 33.5 | 37.36 | 34.3 | 30.14 | 32.2 |
| Leaf blade width (mm) | 7.5 | 4 | 6.8 | 7.75 | 14 | 7 | 11.4 | 8 | 6.8 | 7.4 | 5.4 | 7.2 | 7.8 | 5.6 |
| Ligule length | 13.6 | 11.6 | 7.8 | 8 | 16.4 | 7.2 | 11.8 | 1.38 | 4.4 | 12.2 | 1.74 | 6.02 | 9 | 8 |
| Panicle length ( cm) | 21.28 | 26.24 | 13.74 | 16.4 | 30.96 | 22.7 | 23.62 | 21.22 | 18.14 | 20.8 | 21.42 | 18.98 | 18.8 | 17.2 |
| Panicle wt per tiller/ (g) | 2 | 4.11 | 2.17 | 1.63 | 5.84 | 2.92 | 3.69 | 2.02 | 3.01 | 1.96 | 2.94 | 2.27 | 1.49 | 2.43 |
| 100 grain weight (g) | 1.29 | 2.16 | 1.64 | 1.26 | 2.61 | 1.36 | 1.52 | 1.21 | 2.29 | 1.32 | 1.63 | 1.32 | 1.37 | 1.36 |
| Grain length (mm) | 5.89 | 7.99 | 7.54 | 6.06 | 8.6 | 5.97 | 6.03 | 6.26 | 7.46 | 6.04 | 6.87 | 6.2 | 5.8 | 5.96 |
| Grain width (mm) | 2.57 | 3.04 | 2.69 | 2.43 | 2.93 | 2.62 | 2.92 | 2.44 | 3.14 | 2.65 | 3.39 | 2.65 | 2.5 | 2.61 |
| Plant height (cm) | 57.08 | 88.62 | 70.08 | 58.18 | 75.26 | 54.9 | 91.44 | 58.2 | 61.58 | 53.32 | 89.54 | 70.58 | 58.4 | 57.42 |
| Flag leaf angle | 1 | 1 | 1 | 1 | 3 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 3 | 1 |
| Ligule shape | 1 | 1 | 1 | 1 | 2 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 |
| Auricle colour | 0 | 1 | 1 | 0 | 1 | 0 | 0 | 1 | 1 | 0 | 1 | 0 | 0 | 0 |
| Culm strength | 1 | 1 | 3 | 1 | 1 | 1 | 1 | 1 | 5 | 3 | 1 | 1 | 3 | 3 |
| Apiculus colour | 3 | 2 | 3 | 2 | 2 | 3 | 2 | 2 | 2 | 3 | 2 | 2 | 2 | 3 |
| Stigma colour | 5 | 5 | 3 | 3 | 3 | 5 | 3 | 3 | 5 | 5 | 5 | 3 | 5 | 5 |
| Lemma and palea colour at maturity | 4 | 3 | 2 | 4 | 0 | 3 | 0 | 4 | 4 | 4 | 0 | 4 | 4 | 4 |
| Seed coat (bran) colour at maturity | 3 | 2 | 2 | 1 | 2 | 1 | 4 | 1 | 4 | 2 | 4 | 1 | 2 | 1 |
| Days to heading | 86 | 119 | 87 | 94 | 115 | 95 | 119 | 86 | 94 | 86 | 115 | 94 | 86 | 86 |
| Days to Maturity | 114 | 155 | 118 | 123 | 151 | 123 | 154 | 115 | 122 | 117 | 154 | 122 | 114 | 114 |

Flag leaf angle 1(erect),3 (intermediate), Ligule shape 1 (acute to acuminate) 2 (cleft), Auricle colour 0 (absent), 1(pale green), culm strength 1(strong), 3moderate strong), 5 (intermediate strong), apiculus colour 2 (straw), 3 (browm), stigma colour 3(yellow), 5 (purple), lemma and palea colour 0 (straw), 2 (brown spots on straw), 3 (brown furrow on straw, 4(brown), seed coat colour (1(white), 2(light brown), 3(speckled brown), 4(brown)

Principal component analysis (PCA) grouped the 11 quantitative traits into 11 principal components (data not shown). The first 4 PCs explained 91.5% of the total variability. The score plot diagram showing variation of tested Suwandal accessions based on first two PC values was depicted in Figure 2.

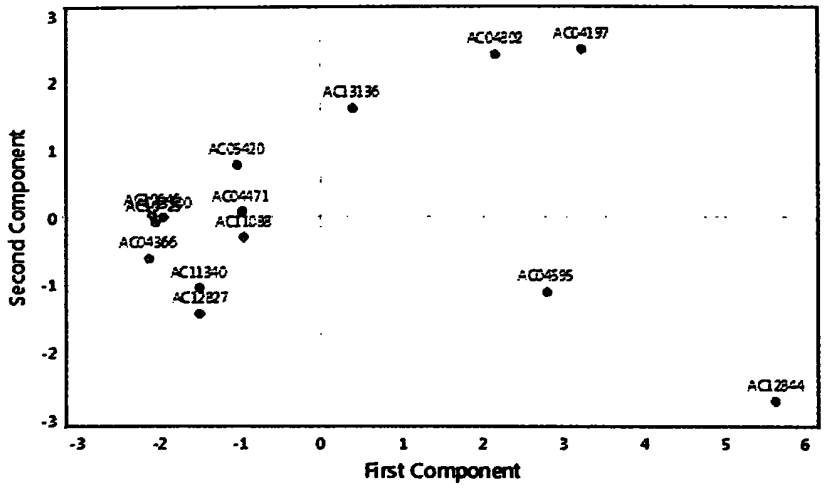


**Figure 2. Bi plot showing variation of Suwandal accessions based on the 1st and 2nd PC values using 11 quantitative traits**

Phylogenetic tree which developed based on qualitative and quantitative morphological data of *Suwandal* rice accessions is depicted in the Figure 3. The analysis grouped these accessions into three clusters. The AC12844 was very much different from other accessions and forming a separate individual cluster (cluster 1). AC04802, AC04595 and AC04197 formed a separate cluster (cluster 2). Rest of the 10 accessions grouped into cluster 3 with more than 50% similarity. Sub clusters were identified within cluster 3 which shows more than 70% similarity. AC13300, AC11340, AC12827, AC04366 & AC11088 grouped into sub cluster A and AC10729, AC04471 & AC10646 grouped into sub cluster B. AC05420 and AC13136 stay separately within the cluster 3. Based on these morphological data no duplicates were present within these 14 accessions. Days to maturity, panicle length, panicle weight, grain weight, grain size, presence of own, ligule shape are the main characters which involve with grouping of these 14 accessions.

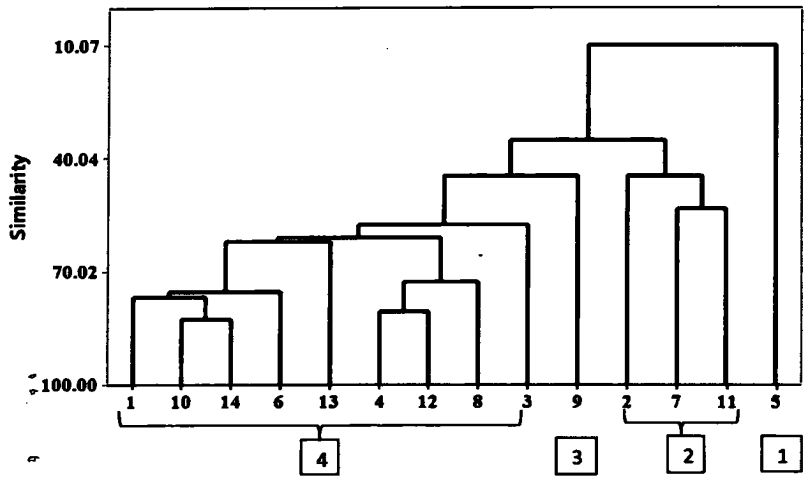


**Figure 3. Phylogenetic tree developed for different '*Suwandal*' accessions based on morphological parameters using Euclidean distance and average linkage method [1-AC12827, 2-AC04197, 3-AC05420, 4-AC10646, 5-AC12844, 6-AC11088, 7-AC04595, 8-AC10729, 9-AC13136, 10-AC11340, 11-AC04802, 12-AC04471, 13-AC04366, 14-AC13300]**

## Molecular characterization

**Allelic Diversity:** A total of 82 alleles were detected at 30 microsatellite markers across 14 *Suwandel* rice accessions. Out of these 30 SSR loci 28 loci showed polymorphism. Allele richness varied from 1 (RM 255 & RM 228) to 5 (RM 202) alleles across tested populations. The frequency of the most common allele at each locus ranged from 33% (RM 202) to 100% (RM 255 and RM 228) with an average of 65%. A high level of genetic diversity existed among 30 loci studied across 11 *Suwandel* rice accessions. It ranged from 0.00 to 0.75 with an average of 0.44 (Table 2).

SSR markers were highly informative and polymorphic as evident from its PIC values. It measures the polymorphism among populations for a marker locus. The PIC value of each marker could be evaluated basis of allele frequencies. It varied from 0.0 (RM255 and RM228) to 0.71(RM202) with the average of 0.38 (Table 2). The highest PIC value 0.71 was obtained for RM202 followed by RM 440 (0.69), RM539 (0.65), and RM217 (0.62). PIC values of the present study revealed that the locus RM202 would be the best in screening these 14 *Suwandel* rice accessions followed by RM440, RM539 and RM217.

The mean value of allele per locus and PIC value obtained in this study is considerably lower than the genetic study of diverse rice germplasm (Thomson *et al.*, (2006), Hossain *et al.*, (2007) and Pervaiz *et al.*, (2009). One of the reasons for low diversity in this study might be due to consideration of single traditional rice cultivar which collected from different areas under same name. The diversity exist among the accessions might be due to the genetic drift of the populations and miss identification of cultivars.

The allele frequencies of the most common allele at each locus ranged from 33 % -100 % (Mean 65 %). It was higher than the experimental results of Hossain *et al.*, (2007). Our results of allelic frequencies were mostly comparable to allelic frequency range reported by Pal *et al.*, (2003) who studied the genetic similarity of Basmati and Non Basmati rice varieties. High mean major allele frequency reflected that majority of tested alleles are common among the group of Suwandal accessions.

**Genetic relationship among Suwandal accessions-** To study the genetic relationship among tested accessions, pairwise genetic distances were computed using Nie's genetic distance which based on infinite allele model (Kimura and Crow, 1964). Since these tested accessions are coming under same cultivar name vast genetic distances were not identified. Maximum genetic distance was observed between AC12827 and AC4595 (0.68) and least genetic distance was observed between AC11340 and AC13300 (0.09). According to the phylogenetic tree developed based on molecular data these 14 *Suwandal* accessions separated into 3 clusters (Figure 4). Cluster 1 comprises four accessions with close genetic relationships (AC11340, AC13300, AC12827, AC011088 & AC13136). Cluster 2 consist three accessions (AC004197, AC004595 & AC004802). Cluster 3 comprises seven accessions. When compare two phlogenetic trees constructed based on morphological and molecular data similarities were identified. In both morphological and molecular grouping AC11340, AC13300 & AC12827 clustered together. Further, AC004197, AC004595 & AC004802 clustered together.

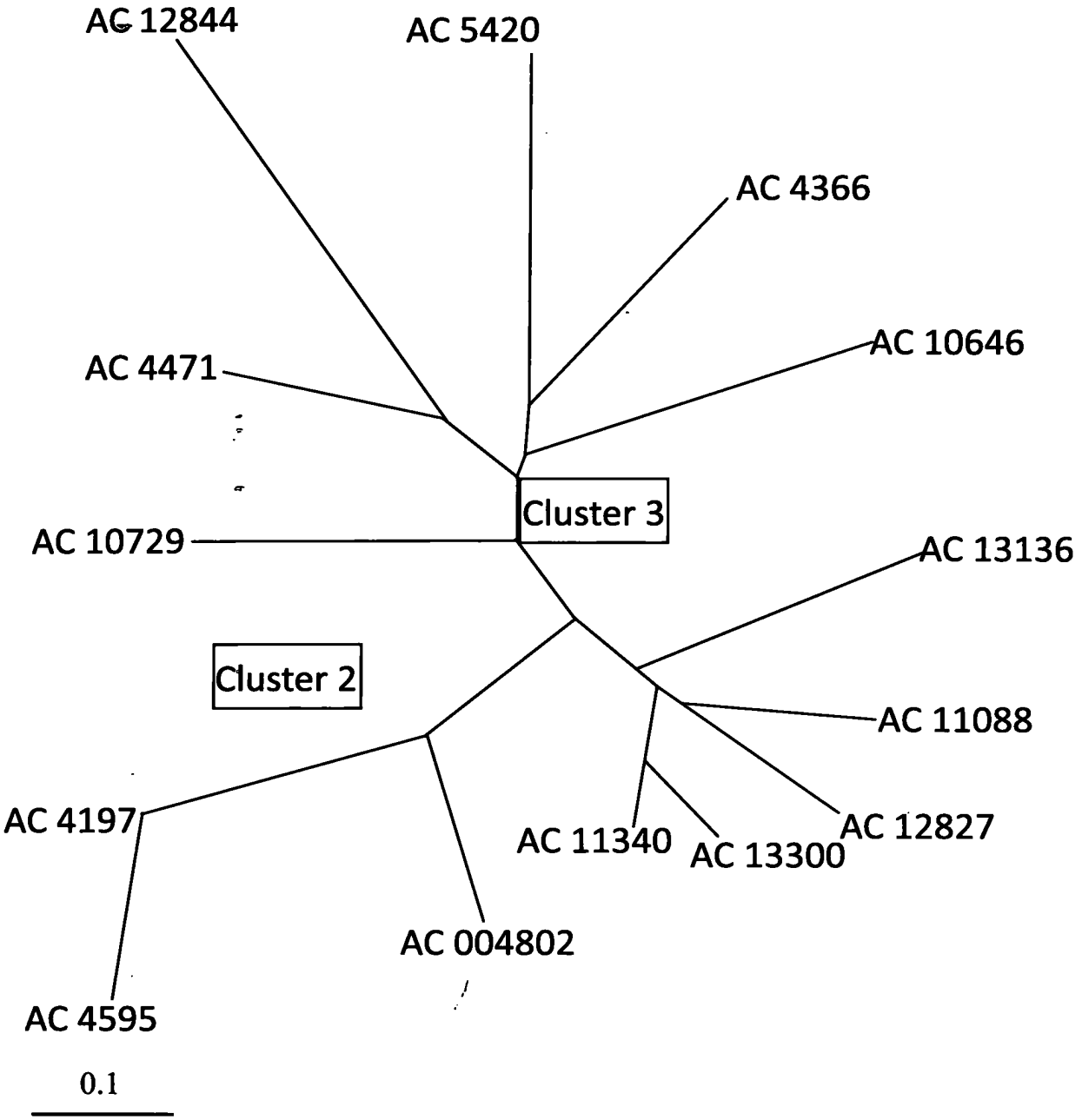


**Figure 4. Phylogenetic tree based on similarity index according to the UPGMA algorithm**

Based on this analysis high genetic diversity was observed among tested *Suwandal* accessions. Genetic distances showed considerable variation and none of the accession pair showed zero genetic distance. It concludes that even though accessions identified with the similar name there were no duplicates in genetic level. AC12844 showed distinctly different characters. It can be a miss identification of accession as *Suwandal*. However, AC13300 and AC11340 showed very close genetic relationship both under molecular

and morphological characterization. Similar study was done by Ahmed *et al.*, (2015) for different accessions of *Dhaliboro* rice cultivar at Bangladesh using agro morphological characters and identified some duplicates as well as genetically different accessions. In this study tested landraces were obtained from various agro climatic regions of the country and evolve through long process under different environmental conditions hence genetic divergence can be occurred. Similar study was done by Sulgotra *et al.*, (2015) and identified similar genetic diversity among Basmathi rice germplasm.

**Physical and physicochemical properties-** Eating and cooking qualities of rice are mainly controlled by the physicochemical properties which greatly influence the consumer's affinity (Rohilla *et al.*, 2000). Physicochemical properties of rice are determined based on amylose content (AC), gel consistency (GC) and gelatinization temperature (GT) and antioxidant activity. Physical and physicochemical properties of tested accessions were summarized in Table 4. In the present study AC % was ranged from 24.76 (AC05420) to 28.93 (AC12827). Except AC05420 rest of the tested accessions grouped into the high (>25%) amylose content. Gel consistency which indicates the texture of cooked rice of these tested accessions ranged from 3.4 to 5.1. Except AC04197 (5.1) rest of the accessions gave high gel consistency value (<4). Time required for cooking is determined by the gelatinization temperature. Except AC12844 rest of the accessions showed intermediate GT values. Antioxidant activity of these accessions tested based of free radical scavenging activity. Phenolic acids, α tocopherol, γ oryzanol and anthocyanins are mainly responsible for the antioxidant content and activity in rice bran (Laokuldilok *et al*, 2011). In these tested accessions % free radical scavenging activity value ranged from 34.6 (AC13300) to 87.5 (AC05420) which is significantly different among accessions ($p>0.001$) (Table 4). AC05420 and AC4595 showed over 87 % free radical scavenging activity.

**Table 4. Physical and physicochemical characteristics of tested Suwandal accessions**

| Accession no | BR % | TMR% | HG% | AC(%) * | GC (cm) | FR%* | GT | Seed coat colour |
|---|---|---|---|---|---|---|---|---|
| AC12827 | 76.7 | 71.2 | 42.5 | 28.9 | 3.8bc | 36.3f** | Intermediate | speckled brown |
| AC04802 | 71.9 | - | - | 25.1 | 3.9b | 45.7cde | Intermediate | Light brown |
| AC04366 | 77 | 62.1 | - | 25.6 | 3.9b | 58.9b | Intermediate | Light brown |
| AC11340 | 76.2 | 70.9 | 54 | 28.1 | 3.4cd | 37f | Intermediate | white |
| AC04595 | 69.6 | - | - | 25.5 | 3.4cd | 87.a | High/Inter | Light brown |
| AC13136 | 79.7 | - | - | 26.6 | 3.8bcd | 36.8f | Intermediate | White |
| AC11088 | 78 | 73.1 | 62.9 | 25.2 | 3.7bcd | 37.4ef | Intermediate | brown |
| AC13300 | 76.3 | 70.9 | 46.8 | 27.8 | 3.5bcd | 34.6f | Intermediate | white |
| AC10646 | 75 | 70.2 | 57.7 | 26.8 | 3.4cd | 41def | Intermediate | brown |
| AC10729 | - | - | - | 27.6 | 3.5bcd | 40.2def | Intermediate | Light brown |
| AC12844 | 74.2 | 68.6 | 63.5 | 28.6 | 3.3d | 46.2cd | Low | brown |
| AC04471 | 73.5 | 68.3 | 65.3 | 28.9 | 3.8bcd | 38.2def | Intermediate | white |
| AC04197 | 70.5 | | | 26.9 | 5.1a | 52.6bc | High/Inter | Light brown |
| AC05420 | 76.7 | 69.1 | 40 | 24.8 | 3.9b | 87.5a | Intermediate | white |

*Significant at 0.001 probability level, ** Mean with same letters are not significantly different at 0.05% probability level.

BR-Broken rice, TMR-Total milling rice, HG-Head grain, AC-Amylose content, GC-gel consistancy , FR-Free radical scavenging activity, GT-gelatinization temperature

## CONCLUSIONS

Based on morphological molecular and quality characterized data, no duplicates were identified among these conserved *Suwandal* accessions. AC12844 showed distinctly different characters and it can be a miss identified cultivar. AC13300, AC11340 and AC12827 showed close relationship both under morphological and molecular data which comes under 3.5 month age group. AC04197, AC04595 & AC04802 made another group with close relationship which comes under 5 month age group. Physicochemical activity of these tested accessions showed significant variation among the accessions and it is not reflected by the morphological and molecular diversity.

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