RESEARCH ARTICLE

Identification of aromatic rice from genetic landrace resource using molecular marker integrated with chemical assessment

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The price of aromatic rice is higher than the price of non-aromatic rice due to its aroma, which is the most integral characteristic of rice. The previous molecular analysis identified the deletion of an eight base-pairs (8-bp) region in exon 7 as the contributor to the aroma of most rice varieties, including *Basmati* and *Jasmine* rice. Therefore, the current study aimed to screen a set of Malaysian landrace rice with the use of a functional marker targeting the 8-bp deletion integrated with sensory assessment using KOH solution. Aromatic rice varieties exhibit a copy of the gene with the deletion of an eight base-pairs (8-bp), resulting in a frame shift mutation that hinders the enzyme activity of betaine aldehyde dehydrogenase (BADH2). As the aroma characteristic was expressed in a recessive manner, sensory assessment using KOH solution was performed on accessions with *fgr/fgr* allele only. The screening of total 186 rice accessions using functional markers revealed the following results: (1) 24 landrace rice accessions showed *fgr/fgr* (12.9%); (2) 149 landrace accessions showed *Fgr/Fgr* (80.1%); (3) 13 landrace rice accessions showed *Fgr/fgr* (7.0%). As for the chemical assessment using KOH solution, all 24 accessions with *fgr/fgr* allele expressed their aromatic sensory in the leaf aromatic test and grain aromatic test. However, the remaining accessions with *Fgr/Fgr* and *Fgr/fgr* may be aromatic due to the presence of other genes that control the aroma of rice. Hence, exploring the variability of the landrace was deemed significant in the identification of novel donor to be used or enhanced in the breeding program through marker-assisted breeding. Conclusively, this study successfully proved the feasibility of integrating molecular marker approach and sensory assessment for the mass screening of *fgr/fgr* allele in identifying aromatic rice from large germplasm collection and breeding program.

Keywords: aromatic rice; *fgr*; functional marker; landrace rice; sensory analysis.

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Introduction

The majority of the global population consumes rice (*Oryza sativa L.*), which subsequently establishes rice as the most essential cereal crop and food. Depending on the aroma, there are two sub-groups of rice, which are aromatic rice and non-aromatic rice [1]. Aromatic rice varieties are mainly low yielding resulting in poorer agronomic performance, highly susceptible to the environmental conditions, and produced in certain countries [2, 3]. Despite that, aromatic

rice varieties are more favorable than non-aromatic rice due to their aroma and grain quality [4]. The aroma, texture, palatability, and desirability of aromatic rice have contributed to its growing popularity locally and globally and subsequently, its significant role in the global rice trading [5-7].

There are different sensory approaches to identify aromatic rice. For examples, by chewing and boiling grains or plant parts with KOH solution, and by smelling [8]. However, the sensory approaches have certain limitations that affect their reliability, particularly when a large sample is involved. Nevertheless, the traditional approaches used to identify aromatic rice are even more challenging due to their negative environmental impact and low narrow sense heritability of aroma [7]. To date, genomic sequences-based functional markers serve as a more robust tool for genetic assessment. The use of aroma-linked genomic markers is generally more advantageous in terms of simplicity, affordability, reproducibility, and constant across different phases, times of year, settings, and agronomic methods [7].

Genetically, the presence of betaine aldehyde dehydrogenase (BADH2) gene (*fgr/badh2/ os2AP/osbadh2*, *LOC_Os08g0424500*), which undergoes mutation and expression on chromosome 8 under homozygous recessive conditions, regulates the aroma of rice [9, 10]. In an earlier study by Bradbury, *et al*. (2005), functional BADH2 enzyme was found to hinder the biosynthesis of the main element of aroma or specifically 2-acetyl-1-pyrroline (2AP) [9]. Unlike non-aromatic rice varieties that display a complete functional copy of the gene encoding BADH2, aromatic rice varieties exhibit a copy of the gene with the deletion of an eight base-pairs (8-bp), resulting in a frame shift mutation that hinders the enzyme activity of BADH2. Therefore, for this study, the identification of aromatic rice from a set of Malaysian landrace rice was genetically conducted by using a functional marker that targets the functional polymorphism of BADH/Fgr gene.

Accordingly, landraces play various integral functions in crop improvement and agricultural production throughout the global history of crops [11]. Crop improvement typically makes use of the diversity of landraces in order to produce new cultivars [12, 13]. A restricted gene pool of advanced cultivars or breeders' lines that can be simply used without consecutive backcrossing for the removal of unwanted characteristics is typically favored among breeders [14, 15]. However, landraces offer a distinctive source of specific characteristics with nutritional quality or tolerance against diseases, pests, and certain environmental conditions [13]. Hence, the objective of this study is to identify novel source of aromatic rice in Malaysian rice accessions using molecular marker approach integrated with chemical sensory evaluation. Hence, this study provided a significant value for future breeding program through marker-assisted breeding.

Materials and methods

A robust and rigorous approach to identify aromatic rice from a set of Malaysian landrace rice, which involved two screening phases, was applied in this study. In the first screening phase, the presence of *fgr/fgr* allele was examined through the molecular screening of all landrace accessions. As for the second screening phase, chemical evaluation using KOH solution was used to examine landrace accessions with *fgr*/*fgr* allele.

Plant materials

A set of 186 landrace accessions was acquired from Malaysian Agriculture Research and Development Institute (MARDI) Rice GenBank (Penang, Malaysia). These landrace seeds were germinated and only three uniform plants of each accession were maintained for the molecular evaluation and chemical evaluation. Two improved varieties, MRQ76 (a Malaysian commercial aromatic white rice) and MR303 (a Malaysian commercial white non-aromatic white

rice), were also included as positive and negative controls, respectively.

Molecular analysis

(1) Leaf DNA extraction:

The young leaves of three uniform plants of each accession were gathered and air dried. The genomic DNAs were extracted based on CTAB DNA extraction buffer [16]. Briefly, the small pieces of these dried leaves were first placed in 96 well DNA extraction plate with 3 mm stainless steel beads and frozen overnight at -80˚C. The samples were removed from the freezer the day after and immediately ground by using Tissue Lyser (Qiagen, Düsseldorf, Germany) prior to incubation with DNA extraction buffer (100 mM Tris-HCl (pH 8), 1.4 M NaCl, 20 mM EDTA, CTAB (3% w/v), β-mercaptoethanol) at 65˚C for 1 h. Following that, 0.8% agarose gel was used to measure the integrity of DNA whereas Fluoroskan Ascent (Thermo Fisher Scientific, Waltham, Massachusetts, USA) was used to measure the concentration of DNA.

(2) PCR primer preparation:

Two pairs of functional markers (Badex7-5 and FMbadh2-E7) flanking the 8-bp removal were used to screen the presence of *Fgr*/*fgr* in the selected landrace rice samples. The PCR primer sequences were (1) Badex7-5 primers (forward primer: TGT TTT CTG TTA GGT TGC ATT; reverse primer: ATC CAC AGA AAT TTG GAA AC) [5] and (2) FMbadh2-E7 primers (forward primer: GGT TGC ATT TAC TGG GAG TT; reverse primer: CAG TGA AAC AGG CTG TCA AG) [17]. According to the report from Schuelke [18], the PCR primers were prepared by ligating the primers (either forward or reverse) and fluorescent dye (FAM) with M13 primer sequence (TGT AAA ACG ACG GCC AGT). Both primers and fluorescent dye were ligated together to make them visible for observation by using capillary approach *via* ABI 3130xL Genetic Analyzer (Applied Biosystems, Waltham, Massachusetts, USA).

(3) PCR amplification:

A mixture for the PCR reaction was prepared to attain the target volume of 10 µL, which consisted of 1× PCR buffer (Invitrogen, Waltham, Massachusetts, USA), 10 µM of each forward and reverse primer; 5 µM fluorescence-labelled M13 adaptor, 2 µM of each dNTP, 0.1 µL of bovine serum albumin (BSA) as PCR enhancer, 1 U of Taq DNA polymerase (Invitrogen, Waltham, Massachusetts, USA). The reaction was performed by using GeneAmp® PCR System 9700 (Applied Biosystems, Waltham, Massachusetts, USA) with the program of pre-denaturation for 2 min at 94°C; followed by 35 cycles of 94°C for 30 s, 55°C for 45 s, 72°C for 45 s; and lastly, post-extension for 5 min at 72°C.

(4) PCR result analysis:

Subsequently, the PCR product was multiplexed and mixed with Hi-Di formamide. The GeneScan 500 LIZ (Thermo Fisher Scientific, Waltham, Massachusetts, USA) was used as standard molecular weight ladder. ABI 3130xL Genetic Analyzer (Applied Biosystems, Waltham, Massachusetts, USA) was employed to generate allelic based electropherogram. The size of allele was determined by using GeneMapper (version 5) (Thermo Fisher Scientific, Waltham, Massachusetts, USA). Referring to the method of Arif, *et al*. [19], the obtained electropherograms were scored accordingly to avoid misinterpretation of the allele. Allele were scored based on peak resolution and intensity.

Sensory analysis (leaf aromatic test and grain aromatic test)

The leaf aromatic test was conducted in this study. Briefly, 0.2 g of leaf samples from each genotype was acquired and were sliced into tiny pieces and placed in the glass petri-dishes. After the addition of 10 mL of 1.7% potassium hydroxide (KOH) solution to each petri-dish, the petri-dish was instantly covered and placed under the room temperature for 10 minutes. Following that, each petri-plate was opened one at a time for the leaf aromatic test. The content of each petri-dish was subsequently smelt and scored in terms of aroma. In addition, the grain aromatic test was performed by soaking 10 grains of each genotype in 10 mL of 1.7% KOH solution in a covered glass petri-dish at room temperature

for 1 h. Three panelists from MARDI who are familiar with sensory test were invited to complete the scoring of the aroma of each accession for both tests.

Results and discussion

By using Badex7-5 and FMbadh2-E7 molecular markers, the molecular characterizations of a set of 186 Malaysian landrace rice demonstrated the consistency of both markers in producing alleles. The screening using both markers revealed the following results: (1) 24 landrace rice accessions showed *fgr*/*fgr* (12.9%); (2) 149 landrace accessions showed *Fgr*/*Fgr* (80.1%); (3) 13 landrace rice accessions showed *Fgr*/*fgr* (7.0%). The details of allelic segregation of each landrace rice were summarized in Table 1. It showed that the aromatic rice (*fgr*/*fgr*) presented the allele size of 112/112 bp while the non-aromatic rice varieties showed the allele size of 120/120 bp (Figure 1). It was conformed the 8-bp deletion in the aromatic varieties.

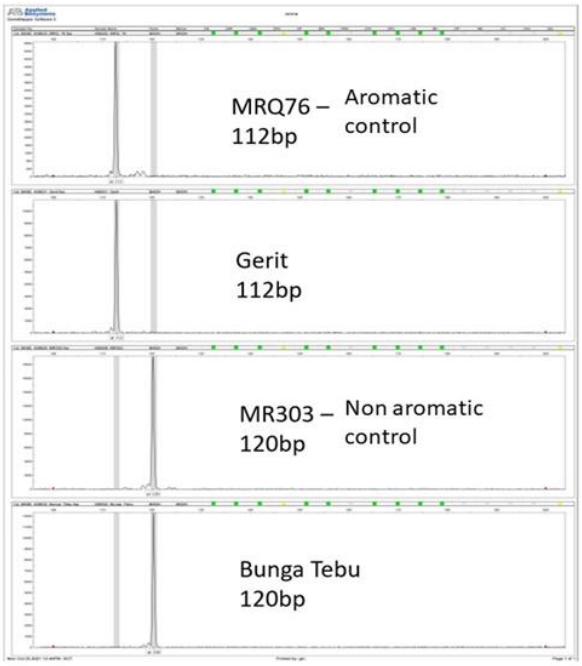

Figure 1. Allelic segregation of Badex7-5. MRQ76 was a control for aromatic rice variety while MR303 was a control for non-aromatic rice variety.

According to Bradbury, *et al*. [9], the deletion of 8-bp in exon 7 was found to play an integral role in the aroma characteristic of most rice varieties, including *Basmati* and *Jasmine* rice. As the aroma characteristic was expressed in a recessive manner, chemical assessment using KOH solution was considered to examine accessions with *fgr*/*fgr* allele only in this study. As a result, all 24 accessions with *fgr*/*fgr* allele were found to express their aromatic sensory in the leaf aromatic test and grain aromatic test. However, the remaining accessions with *Fgr*/*Fgr* and *Fgr*/*fgr* might be aromatic due to the presence of other gene that controls the aromatic trait of rice. Nevertheless, these accessions clearly did not harbor *fgr*/*fgr* allele that was responsible for the aroma of rice. Hence, exploring the variability of the landrace was deemed significant in the identification of novel donor that could be used or enhanced in the breeding program through marker-assisted breeding.

Overall, the consistent results of leaf aromatic test and grain aromatic test demonstrated the applicability of both markers in marker-assisted breeding for the production of aromatic rice varieties. The use of traditional breeding approaches to develop quality characteristics is very challenging due to the difficulties in assessing these characteristics during the breeding process itself as well as the considerable amount of time required to select loci from a large population. However, the use of marker-assisted selection (MAS) promotes the productivity, reliability, and consistency of quality rice breeding programs under different environmental conditions [20]. Furthermore, the application of marker-assisted breeding effectively facilitates the attainment of backcross breeding goals and gene pyramiding for specific characteristics as well as incorporates target quantitative trait loci (QTLs) into the breeding programs [21]. Moreover, numerous studies successfully demonstrated the effectiveness of MAS in enhancing the quality of aromatic rice varieties. The suitability of MAS to identify the presence of BADH allele in aromatic rice from large germplasm collection was also

Table 1. Summary of molecular and sensory analysis of 186 landrace rice.

Variety	GenBank accession number	Badex7-5	Fmbadh2-e7	Leaves sensory test	Grain sensory test
MRQ76	Breeder seed	*fgr/fgr*	*fgr/fgr*	Aromatic	Aromatic
MR303	Breeder seed	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Tega	MRGB04052	*Fgr/fgr*	*Fgr/fgr*	-	-
Rambut	MRGB03834	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Opurak	MRGB04214	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Bunga Tebu	MRGB06022	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Gerit	MRGB05975	*fgr/fgr*	*fgr/fgr*	Aromatic	Aromatic
Kurau	MRGB05970	*Fgr/fgr*	*Fgr/fgr*	-	-
Kinabalu	MRGB04838	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Kajang	MRGB00628	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Jambi	MRGB00559	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Radin Ebos 41	MRGB01356	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Majat D	MRGB00833	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Tapan A	MRGB03099	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Anak China	MRGB02920	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Ruan	MRGB03555	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Campa	MRGB03685	*Fgr/fgr*	*Fgr/fgr*	-	-
Buku Rotan 3	MRGB03933	*Fgr/fgr*	*Fgr/fgr*	-	-
Badak	MRGB06039	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Pulut Tempunai	MRGB05968	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Berteh	MRGB05965	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Pulut Hitam	MRGB05964	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Padi Gamen	MRGB07119	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Bulat	MRGB06003	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Uban	MRGB06038	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Geli 25	MRGB06025	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Kuku Belang	MRGB06001	*fgr/fgr*	*fgr/fgr*	Aromatic	Aromatic
Taring Pelanduk	MRGB06000	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Putih(Huma)	MRGB06049	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Pulut Manis	MRGB06053	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Gemalah	MRGB06044	*fgr/fgr*	*fgr/fgr*	Aromatic	Aromatic
Piya	MRGB06048	*fgr/fgr*	*fgr/fgr*	Aromatic	Aromatic
Jambok	MRGB06212	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Jabat	MRGB06060	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Bidor	MRGB06301	*fgr/fgr*	*fgr/fgr*	Aromatic	Aromatic
Langkatan A	MRGB03260	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Pulut Hitam Siam	MRGB06620	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Sat 4	MRGB06631	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Ranjam	MRGB06568	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Lebar 1	MRGB07116	*fgr/fgr*	*fgr/fgr*	Aromatic	Aromatic
Muar 1	MRGB06583	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Padi Gunong	MRGB07120	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Kuku Balam 1	MRGB07111	*fgr/fgr*	*fgr/fgr*	Aromatic	Aromatic
Padi Huma	MRGB07121	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Patih II	MRGB07122	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Ulat Kuning	MRGB07125	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Geli 25	*MRGB06025*	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Dari	MRGB07159	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Pulut Simanggang	MRGB09177	*Fgr/fgr*	*Fgr/fgr*	-	-
Wangi Puteh	MRGB03838	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Pulut Hitam Beras	MRGB03842	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Gerik	MRGB05976	*fgr/fgr*	*fgr/fgr*	Aromatic	Aromatic
Batu	MRGB05979	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Dara	MRGB06024	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Pulut Kurau	MRGB05967	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Selayang	MRGB05985	*fgr/fgr*	*fgr/fgr*	Aromatic	Aromatic
Sambung	MRGB05986	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Pulut Hitam (aroma)	MRGB06031	*Fgr/fgr*	*Fgr/fgr*	-	-
Pulut siaman	MRGB06033	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Kutip 2	MRGB06634	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Jenaga	MRGB06056	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Pulut Hitam Dukung	MRGB06074	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Kepak	MRGB06223	*Fgr/Fgr*	*Fgr/Fgr*	-	-

Pulut Coreng	MRGB06081	*fgr/fgr*	*fgr/fgr*	Aromatic	Aromatic
Pulut Kijang	MRGB06076	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Buih	MRGB06043	*fgr/fgr*	*fgr/fgr*	Aromatic	Aromatic
Serendah Kemboja	MRGB04994	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Pulut Merah 3	MRGB03843	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Rengan Wangi	MRGB03835	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Nangka Wangi 1	MRGB03978	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Langsat	MRGB03830	*Fgr/fgr*	*Fgr/fgr*	-	-
Buah rotan	MRGB03812	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Lenggong	MRGB03831	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Putih	MRGB03833	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Cepa	MRGB03813	*Fgr/fgr*	*Fgr/fgr*	-	-
Resid	MRGB03680	*fgr/fgr*	*fgr/fgr*	Aromatic	Aromatic
Radin	MRGB03683	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Kao pom	MRGB03686	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Merah wangi 1	MRGB03693	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Pulut pujoh 2	MRGB03705	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Pulut tupai 1	MRGB03707	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Rambut hitam	MRGB03710	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Bongkok	MRGB03811	*fgr/fgr*	*fgr/fgr*	Aromatic	Aromatic
Mansau	MRGB03369	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Engkabang	MRGB03441	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Pulut wan	MRGB03474	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Ubak Babui	MRGB03496	*Fgr/fgr*	*Fgr/fgr*	-	-
Rotan	MRGB03527	*Fgr/fgr*	*Fgr/fgr*	-	-
Pok	MRGB03570	*fgr/fgr*	*fgr/fgr*	Aromatic	Aromatic
Perempong	MRGB03601	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Pingkak	MRGB03602	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Raya	MRGB03662	*fgr/fgr*	*fgr/fgr*	Aromatic	Aromatic
Belau	MRGB03193	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Bisbang	MRGB03194	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Bawang	MRGB03213	*Fgr/fgr*	*Fgr/fgr*	-	-
Berangan	MRGB03214	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Langkatan A	MRGB03260	*fgr/fgr*	*fgr/fgr*	Aromatic	Aromatic
Lantit	MRGB03264	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Kasua	MRGB03300	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Kepapa	MRGB03321	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Mau	MRGB04472	*fgr/fgr*	*fgr/fgr*	Aromatic	Aromatic
Mama	MRGB03349	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Abong	MRGB03355	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Aboy	MRGB02936	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Sensay	MRGB02938	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Sedong	MRGB02964	*fgr/fgr*	*fgr/fgr*	Aromatic	Aromatic
Sia Langit	MRGB02968	*Fgr/fgr*	*Fgr/fgr*	-	-
Bidong A	MRGB02995	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Buntar A	MRGB03183	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Radin Kepek 130	MRGB03184	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Rambutan	MRGB01410	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Sadong	MRGB01444	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Sampong	MRGB01498	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Sebulu B	MRGB01513	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Seriap	MRGB01631	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Siam ER 72	MRGB01694	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Siamoi	MRGB01698	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Lilu	MRGB00802	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Tropik	MRGB01822	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Manik Siam	MRGB00842	*Fgr/Fgr*	*Fgr/Fgr*	-	-
manik 144	MRGB00845	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Mek Bujang Kelsom	MRGB00988	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Merjat	MRGB01009	*fgr/fgr*	*fgr/fgr*	Aromatic	Aromatic
Musang B	MRGB01043	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Pandan Gelap 149	MRGB01194	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Kolias	MRGB00709	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Kontol	MRGB00713	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Kontor	MRGB00715	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Kuatek	MRGB00719	*Fgr/Fgr*	*Fgr/Fgr*	-	-

Lawang Lawi 99	MRGB00764	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Lawang Lawi 168	MRGB00765	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Lawang Lawi	MRGB00761	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Lebai Yaakub	MRGB00769	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Lekon	MRGB00772	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Lekon 55	MRGB00773	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Jambak	MRGB00556	*Fgr/fgr*	*Fgr/fgr*	-	-
Jintan Putih	MRGB00620	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Jintan Merah	MRGB00610	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Bemban	MRGB00198	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Genan	MRGB00472	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Gambunt	MRGB00480	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Buntol	MRGB00257	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Buis	MRGB00238	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Kunyit	MRGB03828	*fgr/fgr*	*fgr/fgr*	Aromatic	Aromatic
Guabon H	MRGB04219	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Cheloring	MRGB03815	*Fgr/Fgr*	*Fgr/Fgr*	-	-
China	MRGB03817	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Huma Kuning	MRGB03824	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Huma Wangi	MRGB03825	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Tingkang	MRGB04433	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Ruabon	MRGB04228	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Ulat	MRGB04485	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Chali	MRGB03814	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Hitam	MRGB03819	*fgr/fgr*	*fgr/fgr*	Aromatic	Aromatic
Anak Puteh	MRGB00126	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Bodong	MRGB00223	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Bingka Petani	MRGB00211	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Biankok	MRGB00216	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Berayong	MRGB00203	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Gansayut	MRGB00459	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Belut Kuning	MRGB00194	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Cempaka 173	MRGB00334	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Anak Ru	MRGB00127	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Angkalias	MRGB00139	*fgr/fgr*	*fgr/fgr*	Aromatic	Aromatic
Gayo vato	MRGB00465	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Che Lawi 16	MRGB00325	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Kedah	MRGB00669	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Jalil 14	MRGB00552	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Gunong 88	MRGB00486	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Chatek Kuning	MRGB00318	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Kejang	MRGB00682	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Koja 2	MRGB00707	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Karok	MRGB00656	*fgr/fgr*	*fgr/fgr*	Aromatic	Aromatic
Katimun	MRGB00663	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Dongkong	MRGB04364	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Pulut Minyak	MRGB04999	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Poturu	MRGB05544	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Kadungkang	MRGB04502	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Tarat	MRGB04510	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Pulut Pelanduk	MRGB04515	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Pulut	MRGB05971	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Pulut Pisang	MRGB06018	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Huma Lenggong 2	MRGB03822	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Panji Putih	MRGB04186	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Panji Halus	MRGB04142	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Kemaman 7	MRGB04014	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Undus	MRGB04321	*Fgr/Fgr*	*Fgr/Fgr*	-	-
Rantai	MRGB05981	*fgr/fgr*	*fgr/fgr*	Aromatic	Aromatic

demonstrated in the current study, which highlights its potential for large-scale routine genotyping for flavor characteristics in breeding materials and germplasms.

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