Low-energy heavy-ion beam induced high-yield mutation breeding of Thai jasmine rice (*Oryza sativa* L. cv. KDML 105)

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Abstract. Thai jasmine rice was improved by low-energy heavy-ion beam induced mutation breeding for high yields. Seeds of Thai rice KDML105 and its ion-beam-induced primary mutant BKOS6 were bombarded by mixed atomic and molecular nitrogen ions accelerated by tens kV in a home-developed high-current ion implanter. Both phenotypes and genotypes of induced mutants were investigated. In M1 generation, more than a hundred plants with the photoperiod insensitivity potential were obtained. In the subsequent generations, tens of rice mutants with a broad spectrum of phenotypic variations dominantly supporting the high yield potential were selected and studied for the mutation stability till M5 generation. The mutants possessed not only high crop yields but also other properties improved or altered in the grains. DNA fingerprinting analysis revealed polymorphisms in the mutants distinguished from that of KDML105. The cDNA fingerprinting investigation indicated four additional fragments in the mutant profiles encoding proteins which could be involved in the high yield characteristics of the mutants.

1. Introduction

Thai jasmine rice KhaoDawkMaLi 105, or KDML 105 (*Oryza sativa* L. cv. KDML 105), is one of the two major high-quality and high-value fragrant rice varieties (KDML 105, basmati rice) in the world rice market. However, its mass production is limited because of a low crop yield (average 2.27 MT/ha) and photoperiod sensitivity. Therefore, development of new rice varieties improved from KDML 105 is an urgent national challenge to Thailand. Conventional breeding with marker-assisted selection has been applied for KDML 105 development [1‒3]. However, conventional breeding generally needs at least 10 generations to stabilize mutants, whereas mutation induction takes less time (5 generations). Gamma-ray radiation has been applied to induce mutation breeding of KDML 105 in Thailand. However, negative characteristics such as slender and delicate internodes, low grain yield, blast, and BPH susceptibility of the tall rice variety KDML 105 still remain in the mutants [4]. Thereafter, more effective techniques have been being investigated and developed. Ion beam biotechnology is a recently developed novel particle-irradiation biotechnology [5]. High-energy (> MeV) ion beams have been applied in rice development [6]. Nevertheless, low-energy (in an order of 10 keV) ion beam biotechnology has gained attention owing to a number of advantages, such as low damage rate, high mutation rate and broad mutation spectra [5, 7‒9]. Low-energy heavy-ion beam bombardment for plant mutation breeding has attracted increased interests from the developing world owing to its relatively low cost and simple
In Thailand, we have applied low-energy heavy-ion beams for development of KDML 105 mutants for more than a decade. Mutation of KDML 105 was induced by using low-energy N-ion bombardment [7,9]. A mutant named BKOS6 [7] had a broad spectrum of mutation phenotypes, including photoperiod-insensitivity and semi-dwarfism, but a lower yield. To effectively induce further mutation of KDML 105 by focusing on the high yield characteristics while maintaining already obtained positive characteristics, the present work continued applying low-energy heavy-ion beams to treat KDML 105 and also retreat BKOS6 seeds.

2. Materials and methods

The target materials were seeds of KDML 105 and its primary mutant BKOS6. The ion bombardments were conducted in off-season cultivation (dry season: March–July), using the 150-kV high-current non-mass-analyzing ion implanter [10]. Prior to the ion beam treatment, to break the rice dormancy, rough seeds were incubated at 49°C for 5 days. The incubated seeds were manually dehusked to expose the embryos and individually placed in holes of a special rice seed sample holder in such a way that the embryos were faced to the ion beam incident direction and then moved into the target chamber of the ion implanter. Ion bombardment was operated under a pressure of 10⁻³ Pa using mixed singly charged atomic and molecular nitrogen ion (N⁺, N₂⁺) beam at a beam current of about 2 mA in a beam interrupting mode. Totally, 22,800 KDML 105 seeds were bombarded by the ions accelerated by 60, 80 and 100 kV to ion fluences of 1, 2, 4, 8×10¹⁶ ions/cm², respectively, and 6,000 BKOS6 seeds were bombarded by the ions accelerated by 60 kV to ion fluences of 1 and 2×10¹⁶ ions/cm², respectively. After ion bombardment, the seeds were then planted in a culture field.

In the phenotype study, cultivation and selection of potential mutation of M1 plants were carried out in the off-season period (March–July). The ion-bombarded rice seeds were cultured on March 1 and then transplanted on April 1. Early-maturity rice mutants flowered in the beginning of May, while the middle-maturity ones showed panicles in the beginning of June. Consequently, the rice seeds of early- and middle-maturity rice mutants could be harvested in June and July, respectively. Selection of rice mutants in the M1–M9 generations was performed through a pedigree method. M1 plants bearing panicles were marked and one panicle of each M1 plant was collected for M2 cultivation. In M2–M5 generations, the rice plant of each mutant that had the high tillering capacity and number of seeds per panicle was selected and mutational stabilities in such as photoperiod-insensitivity and phenotypic variations were observed and recorded. In M6 generation, 23 KDML 105 and 6 BKOS6 rice mutants were emphatically planted in the in-season period (July–December) to study phenotypic and genomic variations. Phenotypic characteristics supporting the high yield potential of the mutants including culm length, tillering capacity, panicle length, seeds per panicle, and flag leaf length were measured in triplicate (n = 60). The plant height (semi-dwarf, intermediate, or tall) of the mutants in the maturing stage was classified according to the Standard Evaluation System for Rice of IRRI [11]. In M7–M9 generations, potentially high-yielding rice mutants were selected using the plant type characteristics of high yielding rice, such as semi-dwarfism or intermediate height with short or stiff culms; erect, short, narrow, thick, dark-green leaves; and high tillering capacity [12–16]. In M10 generation, crop yield and rice grain properties of the selected potentially high-yielding mutants were finally studied and compared with their wild type, KDML 105.

For the genotype study, leaves were collected from rice plants in M6 generation at the age of 15 days after sowing (vegetative stage). Total DNA was isolated from the leaf tissues of 23 KDML 105 mutants, 6 BKOS6 mutants, and controls, namely the wild type KDML 105. For cDNA fingerprint analysis, one of the mutants, HyKOS1, which had several characteristics corresponding to the plant-type concept of high yielding rice including semi-dwarf, photoperiod-insensitive, and compact canopy with high tillering capacity, clearly distinct from the wild type, as shown in table 1, was chosen for intensive study on gene expressions and compared with those of KDML 105, BKOS6, two semi-dwarf KDML 105 mutants (PKOS1 and PKOS3, previously obtained from ion beam induction), and a spindly KDML 105 mutant (TKOS4, previously obtained from ion beam induction). Total RNA from the leaves of HyKOS1, BKOS6, KDML 105, PKOS1, PKOS3, and TKOS4 at the ages of 65 days, 75 days, 85 days, and 95 days (reproductive stage) was extracted and reversely transcribed to cDNA by using M-MLV reverse
transcriptase (Invitrogen, Brazil) by following the manufacturer’s instructions. The fragments were then sequenced at First BASE Laboratories, Malaysia.

Five selected KDML 105 mutants (HyKOS3, HyKOS3-1, HyKOS7-1, HyKOS16, and HyKOS22) and one selected BKOS6 mutant (HyKOS21) were tested for the crop yield in the in-season period (wet season: July–December) and compared with their wild type (KDML 105) in M10 generation. The protein and the fat contents in brown rice and milled rice were determined (n = 9) by following the AOAC method (AOAC, 2000). Additionally, the amylose contents of brown rice and milled rice were analyzed (n = 9) according to the method of Juliano [17]. Furthermore, 2-AP analysis was performed (n = 9) in brown rice and milled rice using headspace-gas chromatography and nitrogen–phosphorus detector [18]. All the analyses were done in triplicate.

Means of each character of the rice mutants and their wild types were subjected to analysis of variance (ANOVA), and Duncan’s multiple range test (DMRT) was applied for the determination of the differences between the means of each property of the rice mutants and their wild type (P ≤ 0.05).

3. Results and discussion

Tables 1 and 2 show a broad spectrum of rice mutation phenotypes. Most of the KDML 105 mutants showed shorter and sturdier stature compared to their wild type, whereas BKOS6 rice mutants showed a significantly longer (P ≤ 0.05) culm length than their wild type. Generally, most of the KDML 105 and BKOS6 rice mutants showed higher numbers of tillers per plant and seeds in panicles and longer length in panicles and flag leaves than those of their wild types. Color variations in several tissues such as leaf sheath, pericarp, ligule, auricle, etc. were also found in some rice mutants. From the yield test, the mutants showed higher tillers and panicles per hill but shorter culm length than the wild type (figure 1). HyKOS22 achieved the significantly highest (P ≤ 0.05) crop yield among all mutants, whereas the wild type had the lowest yield. Rice grain properties are important in rice development since the properties directly affect consumer acceptance, especially the 2-AP content. Differences in the sizes of the rough seeds and the brown rice between the rice mutants and their wild type were clearly observed. The DNA fingerprints obtained revealed genomic variations in the rice mutants. Among ten primers used in the experiment, eight primers, OPAR14, OPAV02, OPAV05, OPAV08, OPAV15, OPAW11, OPAW12, and OPAW18, exhibited distinguished genomic DNA variations between KDML 105 and the mutants, represented by the additional bands in the mutant samples as shown in figure 2 (a). Figure 2 (b) shows the analysis of cDNA fingerprints, including their DNA sequences. Four sequences of different additional fragments showed similarity to interesting proteins which were potentially involved in phenotypic variations in HyKOS1 and might be involved in its high yield potential. The similarities of the amino acid sequences of encoded proteins from additional fragments and protein sequences in GenBank database are seen as follows: (1) Hy4-1 of 692 bp encoded partial protein similar to Spotted leaf protein 11 (ABG22055.1) of O. sativa subsp. Japonica with the identity of 100%, (2) Hy5-3 of 464 bp encoded partial protein similar to putative gibberellin action negative regulator SPY protein (OsSPY) (Q6YZI0.1) of O. sativa subsp. Japonica with the identity of 100%, (3) Hy9-5 of 345 bp encoded partial protein similar to Cytochrome P450 family protein (ABF97993.1) of O. sativa subsp. Japonica with the identity of 100%, and (4) Hy21-6 of 795 bp encoded partial protein similar to putative growth-regulating factor 1 (BAD67915.1) of O. sativa subsp. Japonica with the identity of 99%.

4. Conclusion

N-ion beams at tens-keV energy bombarded seeds of KDML 105 rice and re-bombarded its ion-beam-induced primary mutant for enhancing and multiplying effectiveness in mutation induction. Stabilized mutants having a broad spectrum of high-yield-based mutation phenotypes with multiple improved characteristics were obtained. Genomic variations were distinguished between KDML 105 and its mutants from analyzed DNA fingerprints. Expression of four genes, identified from cDNA-RAPD, encoded Spotted leaf protein11, Putative gibberellin action negative regulator SPY, Cytochrome P450 family protein, and Growth-regulating factor 1, could be involved in the phenotypic variations and high yield potential characteristics of the mutants.
Table 1. Ion bombardment conditions and phenotypic variations of KDML 105 and BKOS6 mutants in M6. The rice code names in short are used in DNA fingerprint analysis in figure 2. "+" or "−": the property is exhibited positively or null, respectively. W: white; P: purple; R: red.

| Rice code | Ion beam condition (KV/ions/cm²) | Plant height in maturing stage | Culm length (cm) | Tiller per plant | Flag leaf length (cm) | Panicle length (cm) | Seeds per panicle | Color of brown rice |
|-----------|---------------------------------|---------------------------------|------------------|-----------------|----------------------|-------------------|------------------|-------------------|
| KDML 105  | -                               | -                               | 115±2.54          | 29±4.3          | 25.4±2.56           | 27.4±2.15         | 113±23           | W                 |
| KDML 105 mutants: | | | | | | | | |
| HyKOS1    | 60/1×10⁶                          | -                               | 73.9±2.90         | 17±3β           | 31.7±2.42           | 25.6±1.91         | 133±23           | W                 |
| HyKOS3    | 60/1×10⁶                          | +                               | 74.7±4.91         | 21±3             | 26.6±1.95           | 21.9±1.89         | 117±20           | W                 |
| HyKOS3-1  | 60/1×10⁶                          | -                               | 101.0±3.20        | 17±3b            | 35.7±2.72           | 27.4±1.86         | 184±16           | W                 |
| HyKOS3-2  | 60/1×10⁶                          | -                               | 103.6±4.16        | 18±3b            | 37.5±2.46           | 29.2±1.97         | 166±25           | W                 |
| HyKOS3    | 60/1×10⁶                          | -                               | 104.6±4.26        | 18±3b            | 36.2±3.11           | 28.2±1.14         | 151±20           | W                 |
| HyKOS3    | 60/1×10⁶                          | -                               | 102.9±4.12        | 17±3b            | 36.5±3.25           | 29.6±1.58         | 164±26           | W                 |
| HyKOS4    | 60/8×10⁶                          | +                               | 71.4±4.16         | 24±4b            | 25.7±1.21           | 23.5±1.40         | 96±22            | P                 |
| HyKOS5-1  | 80/1×10⁶                          | -                               | 83.1±3.13         | 24±3             | 30.8±3.36           | 25.8±1.73         | 120±23           | W                 |
| HyKOS5-2  | 80/1×10⁶                          | +                               | 76.9±4.93         | 22±3             | 26.4±2.89           | 28.2±0.86         | 161±25           | W                 |
| HyKOS6    | 80/2×10⁶                          | -                               | 118.8±4.50        | 13±3             | 33.8±1.72           | 27.7±1.41         | 117±26           | W                 |
| HyKOS7    | 80/2×10⁶                          | +                               | 76.4±2.98         | 18±3b            | 30.3±3.19           | 28.3±2.50         | 133±29           | W                 |
| HyKOS7-1  | 80/1×10⁶                          | -                               | 91.2±3.92         | 27±4b            | 29.3±2.38           | 25.7±2.20         | 110±21           | W                 |
| HyKOS7-2  | 80/1×10⁶                          | +                               | 90.6±4.59         | 15±3             | 29.1±1.97           | 29.8±1.58         | 115±16           | W                 |
| HyKOS8    | 100/2×10⁶                         | -                               | 121.4±4.63        | 25±3             | 42.1±3.37           | 26.3±1.82         | 118±22           | W                 |
| HyKOS9    | 100/2×10⁶                         | +                               | 76.3±3.63         | 24±5             | 29.5±1.83           | 26.7±1.62         | 109±19           | R                 |
| HyKOS10   | 60/2×10⁶                          | -                               | 78.5±3.71         | 25±3             | 29.7±1.30           | 26.7±1.91         | 125±28           | W                 |
| HyKOS11   | 60/4×10⁶                          | +                               | 125.7±3.30        | 19±4b            | 41.3±3.39           | 28.3±1.16         | 176±28           | W                 |
| HyKOS12   | 80/8×10⁶                          | -                               | 84.3±3.20         | 23±4             | 32.2±2.06           | 25.9±1.61         | 112±24           | W                 |
| HyKOS13   | 100/1×10⁶                         | +                               | 79.3±3.84         | 24±3             | 41.0±3.19           | 27.6±1.40         | 135±29           | W                 |
| HyKOS14   | 100/4×10⁶                         | -                               | 80.5±4.54         | 25±3             | 35.5±2.00           | 26.1±1.59         | 154±21           | W                 |
| HyKOS15   | 60/2×10⁶                          | +                               | 75.5±3.89         | 29±4             | 40.8±3.77           | 27.3±0.85         | 134±19           | W                 |
| HyKOS16   | 80/2×10⁶                          | -                               | 76.5±2.02         | 25±4b            | 35.2±3.13           | 24.0±1.46         | 175±19           | W                 |
| HyKOS22   | 60/2×10⁶                          | -                               | 100.6±3.85        | 22±4             | 36.5±3.30           | 28.3±1.35         | 195±12           | W                 |
| Average of the mutants’ phenotypes | 89.0±6.16 | 23.8±4 | 33.6±4.98 | 26.8±7.19 | 139±28 |
| BKOS6     | 60/2×10⁶                          | +                               | 67.4±4.21         | 18±3             | 20.5±3.14           | 19.2±1.45         | 60±15            | P                 |
| BKOS6 mutants: | | | | | | | | |
| HyKOS17   | 60/2×10⁶                          | -                               | 121.8±3.99        | 18±3             | 20.2±3.51           | 29.4±1.50         | 109±24           | W                 |
| HyKOS17   | 60/2×10⁶                          | -                               | 125.6±4.78        | 17±2             | 22.2±3.15           | 30.1±1.89         | 121±21           | W                 |
| HyKOS18   | 80/1×10⁶                          | +                               | 84.1±2.94         | 26±3             | 25.7±3.50           | 23.4±1.69         | 103±26           | P                 |
| HyKOS19   | 60/1×10⁶                          | -                               | 114.7±2.42        | 15±2             | 36.5±3.20           | 29.0±0.91         | 113±28           | P                 |
| HyKOS19   | 60/1×10⁶                          | -                               | 117.3±3.58        | 14±3             | 34.6±2.87           | 30.6±1.12         | 115±22           | W                 |
| HyKOS21   | 60/2×10⁶                          | +                               | 78.2±3.13         | 26±4             | 36.7±3.18           | 21.5±2.04         | 114±17           | P                 |
| Average of the mutants’ phenotypes | 107.0±20 | 19±5 | 29.3±7.49 | 27.5±3.95 | 113±6 |

Figure 1. Appearance of six selected rice mutants possessing the characteristic of high-yielding potential compared with their wild type (KDML105) in the M10 generation (planted in the same period and condition).
Table 2. Crop yield and rice grain properties of rice mutants.

| Property                | Wild type | HyKOS1 | HyKOS3-1 | HyKOS7-1 | HyKOS16 | HyKOS21 | HyKOS22 |
|-------------------------|-----------|--------|----------|----------|---------|---------|---------|
| Crop yield (MT/ha)      | 3.5±0.78a | 7.0±0.80b | 7.8±1.00b | 9.1±1.26b | 8.8±1.36b | 6.2±0.70b | 10.8±0.53b |
| Flowering time (day)    | 87        | 60     | 88       | 91       | 84      | 67      | 82      |
| Calm length (cm)        | 114.7±2.10a | 69.9±1.57c | 93.3±4.56c | 110.3±4.43b | 92.6±0.70d | 80.1±3.46d | 108±2.91b |
| Tiller per hill         | 14±1d     | 21±1e  | 17±1f    | 16±2d    | 25±3g   | 26±3h   | 22±1i   |
| Panicles per hill       | 8±1i      | 11±1j  | 12±1k    | 13±1l    | 14±2m   | 12±1n   | 13±1p   |

Size of rough seed

| Width (mm)              | 2.8±0.05c | 2.7±0.13bc | 2.6±0.03c | 2.8±0.05c | 2.9±0.06c | 2.8±0.09c | 2.7±0.08c |
| Length (mm)             | 11.0±0.17a | 11.1±0.10c | 10.4±0.14b | 10.9±0.07b | 11.1±0.08a | 10.3±0.06c | 9.9±0.14a |
| Thickness (mm)          | 2.8±0.05c | 2.1±0.05c | 2.2±0.05c | 2.2±0.07c | 2.2±0.03c | 2.0±0.07c | 2.2±0.02c |
| 1,000 seed weight (g)   | 23.9±0.81c | 27.3±0.65c | 26.4±0.69bc | 25.7±0.86c | 26.7±0.85c | 23.4±0.52c | 27.3±0.76c |

Size of brown rice

| Width (mm)              | 2.5±0.04c | 2.3±0.02c | 2.3±0.04c | 2.4±0.01c | 2.4±0.01c | 2.4±0.07c | 2.5±0.04c |
| Length (mm)             | 7.7±0.16a | 7.6±0.09bc | 7.8±0.17bc | 7.5±0.11bc | 7.7±0.23bc | 7.0±0.15bc | 7.1±0.03c |
| Thickness (mm)          | 2.0±0.02c | 2.0±0.01bc | 2.0±0.06bc | 2.0±0.02bc | 2.0±0.05bc | 1.8±0.08bc | 2.1±0.03c |

Amylose content

| Brown rice (g/100 g)    | 12.9±0.95a | 10.2±0.73a | 18.9±0.40a | 11.8±0.63a | 11.6±0.58a | 13.0±0.62b | 18.5±0.80a |
| Milled rice (g/100 g)   | 16.1±1.28a | 13.0±0.83a | 19.2±2.44a | 13.5±0.61a | 12.9±0.57a | 13.4±0.46a | 21.0±0.26a |

Protein content

| Brown rice (g/100 g)    | 9.6±0.74a | 10.8±0.36a | 7.9±1.19a | 7.1±1.31a | 8.3±1.11bc | 10.1±1.09bc | 8.3±0.68bc |
| Milled rice (g/100 g)   | 4.2±0.40a | 5.2±0.36a | 7.7±0.59a | 6.7±0.58a | 7.0±0.32a | 8.4±0.76a | 8.1±0.71a |

Fat content

| Brown rice (g/100 g)    | 2.2±0.23a | 1.7±0.24a | 1.8±0.24bc | 1.6±0.22a | 2.0±0.39bc | 2.6±0.24a | 1.8±0.16a |
| Milled rice (g/100 g)   | 0.3±0.12a | 0.3±0.06a | 0.4±0.13b | 0.8±0.20a | 0.7±0.16a | 0.6±0.25b | 0.4±0.17a |

2-AP content

| Brown rice (mg/kg)      | 9.8±0.51a | 6.4±0.43c | 1.1±0.22e | 9.2±0.63a | 7.8±0.62a | Not detected | Not detected |
| Milled rice (mg/kg)     | 10.2±0.38a | 6.9±0.74a | 1.5±0.52a | 10.5±0.34a | 8.0±0.37a | Not detected | Not detected |

Figure 2. Genotypic analysis. (a) DNA fingerprinting. HAT-RAPD amplification patterns obtained from the arbitrary primers (OPAV05, OPAW11, OPAW12, OPAW18) in the rice mutant samples. Arrows indicate the polymorphic bands among the rice varieties. M: DNA marker (λ/PstI). (b) cDNA fingerprints of KDML 105 and 5 KDML 105 mutants (BKOS6, TKOS4, HyKOS1, PKOS1 and PKOS3). (1) and (2) sample materials (leaves) collected at 75 and 85 days old, respectively. Arrows indicate the additional bands found in HyKOS1 mutants compared to KDML 105. M: DNA marker (λ/PstI).
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References
[1] Jairin J, Teangdeerith S, Leelagud P, Kothcharerk J, Sansen K, Yi M, Vanavichit A and Toojinda T 2009 Field Crop Res. 110 263–71
[2] Jantaboon J, Siangliw M, Im-mark S, Jamboonsri W, Vanavichit A and Toojinda T 2011 Field Crop. Res. 123 206–13
[3] Win K M et al 2012 Field Crop. Res. 137 186–94
[4] Bhattacharya K R 2011 Speciality rices Rice Quality ed K R Bhattacharya (Cambridge: Woodhead Publishing) chapter 10 pp 337–76
[5] Yu Z L (Chinese original), Yu L D, Vilaithong T and Brown I (English translators) 2006 Introduction to Ion Beam Biotechnology (New York: Springer)
[6] Ishikawa S, Ishimaru Y, Igura M, Kuramata M, Abe T, Senoura T, Hase Y, Arao T, Nishizawa N K and Nakanishi H 2012 P. Natl. Acad. Sci. USA. 109 19166–71
[7] Phanchaisri B, Chandet R, Yu L D, Vilaithong T, Jamjod S and Anuntalabhochai S 2007 Surf. Coat. Technol. 201 8024–8
[8] Phanchaisri B, Samsang N, Yu L D, Singkarat S and Anuntalabhochai S 2012 Mutat. Res. Fundam. Mol. Mech. Mutagen. 734 56–61
[9] Singkarat S et al 2015 Nucl. Instrum. Methods Phys. Res. B 365 414–8
[10] Davydov S, Yu L D, Yotsombat B, Intarasiri S, Thongleurm C, A-no V, Vilaithong T and Rhodes M W 2000 Surf. Coat. Technol. 131 558–62
[11] IRRI 2002 Standard Evaluation System for Rice (SES) (Manila: International Rice Research Institute) p 7
[12] Tsunoda S 1959 Jpn. J. Breed. 9(4) 161–8, 237–44; 1960 Jpn. J. Breed. 10(2) 107–11; 1962 Jpn. J. Breed. 12(1) 49–55
[13] Jennings P R 1964 Crop Sci. 4 13–5
[14] Ishizuka Y 1971 Adv. Agron. 23 231–315
[15] Yoshida S and Forno D A 1972 Exp. Agric. 8 203–12
[16] Yoshida S 1981 Fundamentals of Rice Crop Science (Manila: International Rice Research Institute)
[17] Juliano B O 1971 Cereal Sci. Today 16 334–40
[18] Wongpornchai S, Dumri K, Jongkaewwattana S and Siri B 2004 Food Chem. 87 407–14