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Yield and quality of traditional and improved Lao varieties of rice

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ABSTRACT: Improvements in its political and tourism sectors have encouraged Lao PDR to explore export options for its traditional waxy rice varieties. As such, farmers and agriculturists are exploring ways to enhance yield and capture high-quality traits into improved varieties such as Thasano1 (TSN1) and Thadokkham1 (TDK1). In this study, the effects of nitrogen (N) on yield and quality traits were investigated in two traditional varieties, Hom Nang Nouane (HNN) and Kai Noy Leuang (KNL), and in two popular improved varieties, TDK1 and TSN1. The plants were cultivated during the 2006 wet season at the Rice and Cash Crop Research Centre, NAFRI, Lao PDR, and were subjected to four N rates. Results showed that while increased N rates enhanced yields and yield components in TDK1 and TSN1, the two traditional varieties, HNN and KNL, were not as responsive. In contrast, the quality traits assessed in this study of the four varieties were not affected by N. Yield and textural attributes further suggest that TSN1 is a better candidate than TDK1 for breeding programs aimed at combining quality traits from HNN and KNL into improved varieties.

KEYWORDS: 2-acetyl-1-pyrroline, gelatinisation temperature, rapid visco-analyser, Thadokkham1, Thasano1, Hom Nang Nouane, Kai Noy Leuang

INTRODUCTION

Rice, the staple crop for people in Lao PDR, is mostly cultivated in the rainfed lowlands of Lao PDR[1] and it is almost all waxy (otherwise called glutinous). Waxy rice contains no amylose due to a mutation in the *Waxy* gene[2]. The Green Revolution, which changed rice-growing in so many countries, had little or no impact in Lao PDR because the germplasm of the Green Revolution was non-waxy, so it was not adopted by Lao rice farmers or consumers because they prefer glutinous rice[3]. For many years, Lao farmers have preserved and cultivated a diverse suite of traditional waxy varieties. Recently, 13 192 traditional varieties were systematically collected from Lao farmers in order to conserve them and preserve their diversity; 85.5% of these are waxy[4]. These figures illustrate (i) the importance of waxy rice to people of Lao PDR, and (ii) that enormous diversity potentially exists in the quality of waxy rices.

Traditional varieties are generally low-yielding, so for many farming families, there is often insufficient rice to last from one crop to the next. Before 1990, traditional varieties were cultivated on more than 95% of the rainfed lowland areas of Lao PDR[1]. In 1991, the Swiss Government commissioned the 15-year Lao-IRRI project to improve yield and food security in Lao PDR, particularly in the rainfed lowlands. The rainfed lowland still produces 72% of the Lao rice crop, but the varieties grown have changed significantly as a result of that project. During the Lao-IRRI project, 18 varieties were released for rainfed lowland production[1], and now two of them, Thadokkham1 (TDK1) and Thasano1 (TSN1), account for most of Lao's rainfed lowland production, and most of the rainfed area that is sown with improved varieties[1]. These improved varieties have had an enormous impact on food security in Lao PDR, and the traditional varieties that they replaced have entered gene banks for conservation[4]. However, two of the traditional varieties, Hom Nang Nouane (HNN) and Kai Noy Leuang (KNL), have continued to be cultivated. HNN is popular in central and southern areas while KNL is popular in central and northern regions of the country[5]. These two varieties are cultivated for their quality, principally their fragrance, and the softness of their grains after cooking[6,7]. In contrast, TDK1 and TSN1 are not fragrant.

The political situation in Lao PDR has changed over the years. At the same time, communication technologies and transportation networks (such as roads and vehicles) have undergone rapid development. As a consequence of the former, Lao PDR has opened its doors to tourism, and aided by the latter, the tourist industry has burgeoned[8]. Thus Lao PDR has been able to introduce visitors from all over the world to the high quality and uniqueness of the traditional waxy, aromatic rices of the country. This has prompted Lao PDR to explore export opportunities for these rices, and has led to a growing awareness among Lao agriculturists that these traditional varieties must be investigated for ways to improve their yield, understand their special quality traits, and include these traits into improved varieties, such as the popular TDK1 or TSN1, which are high-yielding and resistant to pests and diseases[9].

One of the first recommendations to farmers to increase yield is to increase the nitrogen (N) they apply to the crop, which can alter the quality of non-waxy rices[10]. Although quality evaluation programmes for rice have mostly focused on developing methods for non-waxy varieties, a number of analyses commonly used for non-waxy rices could indicate the effect of N fertilizer on the eating and cooking quality of waxy rices. For example, the texture of cooked grains is important to Lao rice consumers[6], and N affects the flavour[10] and texture[11] of the cooked rice. Viscosity traits can predict some sensory properties of rice[12] and viscosity profiles are affected by N[13]. Also, gelatinization temperature relates to cooking time of the rice[14] but has been shown to be unaffected by N[15,16]. The objective of this study is to investigate the effect of N on the yield and quality traits of the two traditional varieties, HNN and KNL, and the two popular improved varieties, TDK1 and TSN1, in order to determine strategies for improving the yield as well as the quality of Lao varieties.

MATERIALS AND METHODS

Plant growth and grain processing

Seeds of TDK1, TSN1, HNN, and KNL were sown in the wet season of 2006 in Lao PDR. One month later, seedlings were transplanted in a split plot design of sub-plots within main plots, in three replications at the Agriculture Research Centre, Vientiane, Lao PDR. Different amounts of N fertilizer were applied to each of the main plots and each main plot contained all the varieties, in three replications, in sub-plots. Each sub-plot was 2 m × 5 m, and 10 rows of 25 plants were transplanted at a spacing of 20 cm between plants and between rows. N was applied at four levels to each main plot, and the same amount of phosphorous and the same amount of potassium were applied for all plots as described in Table 1.

Table 1 Fertilizer and timing regime for the four N treatments.

Main plot	N–P–K (kg/ha)	Amount and time of application
N1	0–30–30	All amount applied as basal fertilizer (the same time transplanting)
N2	30–30–30	0–30–30 kg/ha applied as basal fertilizer and 2 top dressings: each of 15 kg N/ha (25 days and 45 days after transplanting)
N3	60–30–30	30–30–30 kg/ha applied as basal fertilizer and 2 top dressings: each of 15 kg N/ha (25 days and 45 days after transplanting)
N4	90–30–30	30–30–30 kg/ha applied as basal fertilizer and 2 top dressings: each of 30 kg N/ha (25 days and 45 days after transplanting)

The date of flowering was recorded and the number of panicles per plant was counted. Just before harvest, plant height was measured and the proportion of filled grains per panicle counted. At maturity, 21 plants in the middle six rows of each sub-plot were harvested and yield and yield components were recorded.

The harvested grain was then sent to the International Rice Research Institute in the Philippines for analysis of quality traits. Paddy from each sample was dehulled (Satake Rice Machine, Tokyo), milled (Grainman 60-230-60-2AT, Grain Machinery Mfg. Corp., Miami, FL), and a sub-sample ground to flour (Udy Cyclone Sample Mill 3010–030, Fort Collins, CO) to pass through a 0.5 mm sieve. Reagent-grade chemicals were used. Reverse osmosis water, filtered through a 0.22 µm Millipore (Billerica, MA) filter, was used throughout the study.

Protein

Protein content of flour from each treatment and variety was measured by near-infrared transmission. Flour was placed in 4.5/3 mm (uncompressed/compressed path length) sample cups and scanned in transmission mode on a near-infrared scanning monochromator (Infratec 1241 Grain Analyser, Foss, Sweden). Scans were obtained from 570 to 1100 nm. The protein contents of the samples were determined based on the manufacturer's application model for polished rice using the flour module. Each sample was scanned in triplicate.

Gelatinization temperature

Gelatinization temperature was measured by differential scanning calorimetry (Q100 TA Instruments, New Castle, DE). Flour (4 mg) was mixed with water (8 µl) in an aluminium hermetic pan which was then hermetically sealed. The temperature was raised from 25 °C to 100 °C at 10 °C/min. Thermal transitions were recorded and analysed using UNIVERSAL ANALYSIS 2000 software. The gelatinization temperature is reported as the peak of the gelatinization endotherm.

Aroma

The aromatic compound, 2-acetyl-1-pyrroline (2AP) was measured by gas chromatography (Agilent 6890N, Santa Clara, CA, USA), equipped with a mass spectrometer, exactly as described in Ref. 17. Chemically synthesised 2AP was provided by T. Yoshihashi (Japan International Research Centre for Agricultural Sciences, Ibaraki, Japan) and was used to quantify 2AP in the samples. 2AP was only measured and quantified using KNL and HNN grains because the two improved varieties are not aromatic.

Texture

Polished grains of each variety (25 grains) were cooked in the same way as is done in Lao PDR. Polished grains were soaked for 2 h in water, then the soaked grains were steamed for 30 min. For each N treatment, three grains were analysed for hardness and stickiness using a Ta.XT-Plus Texture analyser equipped with a cylindrical probe (35 mm diameter, Stable Micro Systems Ltd., Surrey, UK) immediately after cooking. Strain was set at 90% and the test speed was 0.5 mm/s. Measurements were conducted in triplicate. Not all samples of TDK1 could be analysed for texture because not enough whole grains remained after polishing.

Viscosity

Flour of each sample (3 g) was mixed with water (25 g) in a rapid visco-analyser (RVA) canister. Viscosity was measured by RVA (Newport Scientific model 4D) using the approved method 61-02[18].

STATISTICAL ANALYSIS

Balanced ANOVA, which is conducted when there are equal numbers of observations, was performed using R statistic software (version 2.11.0) for yield, yield components, and protein content. For gelatinization temperature, hardness, and stickiness, unbalanced ANOVA was conducted using SAS (version 9.1) because there were missing observations in some of the replications. Pair-wise comparison of means was done using least significant difference (LSD) at 5% level of significance. For each variety and N treatment, all three replicates were included for each analysis. The correlation matrix among RVA parameters, hardness, and stickiness was generated also using R statistic software, pooling values for all the replicates and all the varieties.

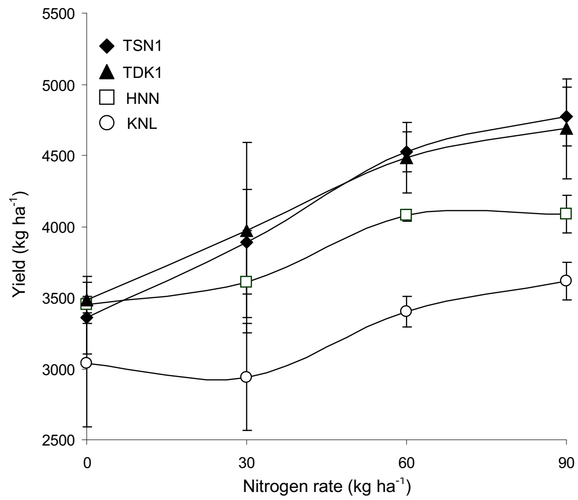

Fig. 1 Yield of improved varieties responded much more to N fertilizer than traditional varieties, and improved varieties were significantly higher yielding at N treatments greater than 30 kg N/ha ($LSD_{0.05}$=366 kg/ha).

RESULTS

Yield

Fig. 1 shows the increase in yield associated with N fertilizer rate of the traditional and the improved varieties. Yields were not significantly different between 0 and 30 kg N/ha, but became significantly different between 30 and 60 kg N/ha. The yield at 90 kg N/ha was similar to that at 60 kg N/ha. The interaction between N treatment and variety was not significant (data not shown). The two traditional varieties were less responsive to N than the two improved varieties (Fig. 1): the yields of TDK1 and TSN1 increased by 40%. In contrast, HNN and KNL showed a yield increase of only 18% between the lowest and the highest N treatments. Moreover, TSN1 always showed a higher yield than TDK1 (Fig. 1).

Table 2 Yield and yield components of the four varieties (V) across four N treatments.

(a)	Filled grains per panicle (%)				
V	N1	N2	N3	N4	Mean
HNN	91.7	90.3	90.9	90.7	90.9^{a}
KNL	82.7	86.9	81.9	84.9	84.1^{b}
TDK1	77.9	84.2	83.0	80.7	81.4^{b}
TSN1	66.4	81.0	77.1	80.7	76.3^{c}
Mean	79.7^{B}	85.6^{A}	83.2^{AB}	84.2^{A}	

Comparison ($LSD_{0.05}$) 2 V means: 4.3 2 N means: 3.6

(b)	Number of grains per panicle				
V	N1	N2	N3	N4	Mean
HNN	106.0^{bA}	126.0^{aA}	124.3^{bcA}	118.3^{bA}	118.7
KNL	117.3^{bB}	124.7^{aAB}	137.7^{abAB}	150.7^{aA}	132.6
TDK1	116.0^{bA}	111.0^{aA}	115.7^{cA}	116.0^{bA}	114.7
TSN1	168.0^{aA}	112.3^{aC}	151.7^{aAB}	141.0^{aB}	143.3
Mean	126.8	118.5	132.3	131.5	

Comparison ($LSD_{0.05}$) 2 V means at the same N level: 20.6
2 N means at the same N level: 26.3

(c)	1000-grain weight (g)				
V	N1	N2	N3	N4	Mean
HNN	31.4	31.9	31.8	31.8	31.7^{a}
KNL	26.6	26.0	25.6	26.3	26.1^{c}
TDK1	30.6	30.0	31.3	32.5	31.1^{a}
TSN1	27.1	28.3	28.4	29.6	28.3^{b}
Mean	28.9	29.0	29.3	30.0	

Comparison ($LSD_{0.05}$) 2 V means: 0.9 2 N means: NA

(d)	Number of panicles per plant				
V	N1	N2	N3	N4	Mean
HNN	6.0	5.4	6.3	6.1	6.0^{b}
KNL	5.6	5.3	6.3	6.0	5.8^{b}
TDK1	7.4	6.7	7.4	9.4	7.7^{a}
TSN1	5.5	5.5	6.3	6.9	6.0^{b}
Mean	6.1	5.7	6.6	7.1	

Comparison ($LSD_{0.05}$) 2 V means: 0.7 2 N means: NA

In this and the next table, in a column (row), means followed by the same lower (upper) case letter are not significantly different at 5% level of LSD.
NA: effects of nitrogen are not significant ($p > 0.05$).

Yield components, particularly 1000-grain weight and number of panicles per plant, were not significantly responsive to N treatment (Table 2c,d). On the other hand, the proportion of filled grains per panicle significantly increased between N rates of 0 and 30 kg/ha (Table 2a). Interactions between N treatment and variety were not significant for these three yield components. The only yield component with significant interaction between N rate and variety was the number of grains per panicle (Table 2b). Number of grains per panicle in HNN and TDK1 did not increase with N treatment. In KNL, an increase in this yield component was observed between 0 and 90 kg N/ha. In TSN1, the number of grains per panicle increased between 30 and 60 kg N/ha. N fertilizer increased plant height significantly for all varieties, but did not alter the days to flowering (data not shown).

Quality

The amount of protein in each variety differed significantly (Table 3b). The N treatments did not affect the amount of protein in the grains until the N rate was 90 kg/ha (Table 3). Gelatinization temperature of the four varieties was not significantly different, and the rate of N fertilizer did not alter it (Table 3a). The rate of N fertilizer did not affect hardness or stickiness of cooked rice (Table 3c,d). The only other significant differences in traits of texture were that cooked grains of HNN were harder than those of KNL and TSN1 and KNL was less sticky than HNN and TSN1. Possibly the texture of the cooked grains of TDK1 is similar to that of HNN, but we were unable to obtain sufficient whole grains of TDK1 after milling to carry out enough replication at each N level to determine statistical significance.

Fig. 2 shows the 2AP content of KNL and HNN

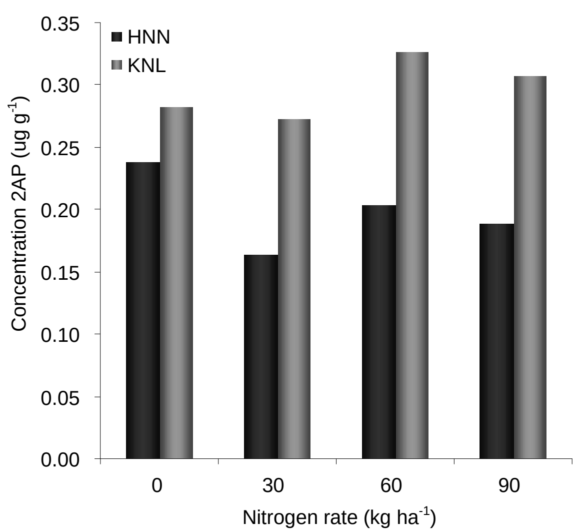

Fig. 2 Concentration of 2AP in the grains of the two aromatic traditional varieties at each N treatment. Differences between varieties and between N treatments are not significant ($LSD_{0.05}$=0.144).

Table 3 Effect of variety (V) and nitrogen treatment (N) on gelatinization temperature, protein content, hardness, and stickiness of freshly cooked rice grains.

(a)	Gelatinization temperature (°C)				
V	N1	N2	N3	N4	Mean
HNN	69.0^{cB}	70.0^{bA}	69.0^{cB}	70.2^{bA}	69.6
KNL	69.9^{bB}	70.9^{aA}	71.5^{aA}	71.4^{aA}	70.9
TDK1	70.9^{aA}	70.7^{abA}	70.6^{abA}	70.1^{bA}	70.6
TSN1	69.8^{bA}	70.5^{abA}	69.8^{bA}	70.1^{bA}	70.1
Mean	69.9	70.5	70.2	70.4	

Comparison ($LSD_{0.05}$) 2 V means at the same N level: 0.8
2 N means at the same V level: 0.9

(b)	Protein content (%)				
V	N1	N2	N3	N4	Mean
HNN	7.6	7.9	7.8	7.9	7.8^{c}
KNL	7.3	7.5	8.6	8.8	8.1^{bc}
TDK1	8.5	8.1	8.3	8.5	8.3^{b}
TSN1	8.6	8.5	8.6	9.3	8.8^{a}
Mean	8.0^{B}	8.0^{B}	8.3^{AB}	8.6^{A}	

Comparison ($LSD_{0.05}$) 2 V means: 0.3 2 N means: 0.4

(c)	Hardness (g)				
V	N1	N2	N3	N4	Mean
HNN	1910.1	1841.3	1874.6	1944.3	1892.6^{a}
KNL	1738.1	1677.9	1746.2	1713.0	1718.8^{b}
TDK1	NA	1855.5	1990.1	1962.4	NA
TSN1	1546.5	1720.2	1856.6	1916.9	1760.1^{b}
Mean	NA	1773.7	1866.9	1884.1	

Comparison ($LSD_{0.05}$) 2 V means: 92.8 2 N means: NA

(d)	Stickiness (g)				
V	N1	N2	N3	N4	Mean
HNN	−1047.9	−1014.7	−968.0	−983.6	-1003.6^{a}
KNL	−898.6	−900.0	−935.6	−854.0	-897.1^{b}
TDK1	NA	−1157.8	−1193.7	−1464.7	NA
TSN1	−995.8	−1088.8	−1124.3	−1046.5	-1063.9^{a}
Mean	NA	1040.3	1055.4	1087.2	

Comparison ($LSD_{0.05}$) 2 V means: 72.2 2 N means: NA

grains at each N treatment. The amount of 2AP in each variety was not significantly different at each N treatment (0 to 90 kg/ha), but the amount of 2AP from KNL grains was significantly greater than that from HNN grains at the three N treatments above zero (Fig. 2).

The RVA traces in Fig. 3 show significant differences ($LSD_{0.05}$) between varieties for some parameters. The viscosity parameters (and values derived from these parameters) of KNL were significantly

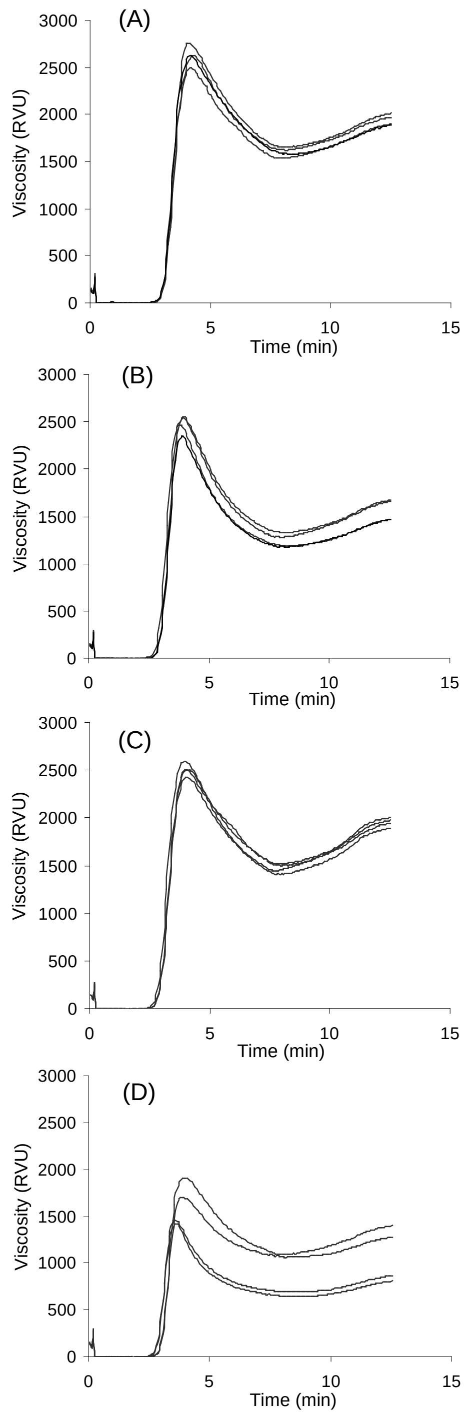

Fig. 3 RVA traces of (A) TDK1, (B) TSN1, (C) HNN, and (D) KNL at each of the four N treatments. Each curve is the average of 3 biological replicates. For KNL, the two lower curves are the lowest N treatments and the two higher curves are the higher N treatments.

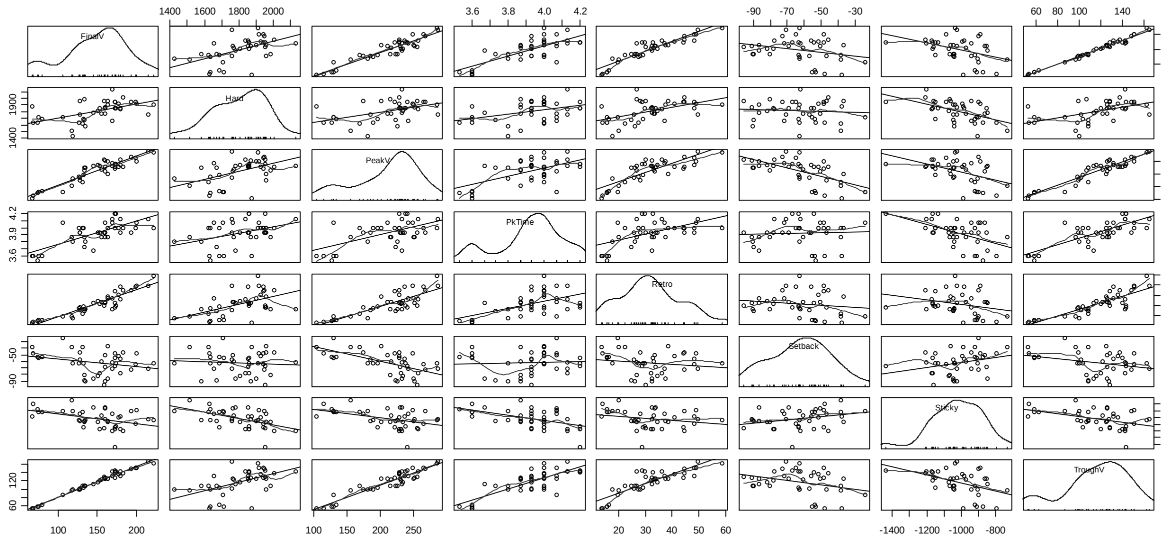

Fig. 4 Correlation matrix of RVA parameters, hardness, and stickiness for the four waxy varieties.

lower than those of the three other varieties. Also, peak, trough, and final viscosities and the derived value retrogradation (the difference between final and trough viscosities) were all significantly higher in TDK1 than in TSN1. The trough viscosity and the retrogradation of HNN were significantly different from those of TDK1 and TSN1. However, there were no significant differences among TDK1, TSN1, and HNN for setback and breakdown.

In contrast, the RVA traces in Fig. 3 show no significant difference due to N treatment for any of the viscosity parameters in TDK1, TSN1, and most parameters for HNN. For KNL, on the other hand, the peak, trough, and final viscosities of grains grown at 0 and at 30 kg N/ha were lower than of those grown the higher rates of N (Fig. 3).

Correlations among viscosity parameters, hardness, and stickiness were obtained for the four waxy varieties (Fig. 4). The direct values of peak, trough, and final viscosity values correlated very well with each other, as well as with the derived values of setback, retrogradation, and breakdown. Hardness correlates weakly with the RVA parameters of peak, trough, and final viscosities, and with retrogradation, but stickiness correlates slightly with hardness (Fig. 4).

DISCUSSION

Yield

The two improved varieties were more responsive to N treatment than the two traditional varieties in terms of yield. The number of panicles per plant was not significantly different among the N treatments (Table 2d). However, it has previously been reported to be one of the most important N responsive yield components [19]. Consistent with this, the yield increase of TDK1 and TSN1 with N might be attributed to an increased number of panicles per plant, as well as a minor effect of an increase in thousand-grain weight (Table 2c), suggesting greater translocation of substrates to the grain during grain-filling. Both of these varieties offer potential as parents in quality improvement programs. However, the enhanced responsiveness of TSN1 to N treatment in terms of yield makes it a more promising candidate than TDK1.

Yield of the two traditional varieties increased by 18% over the N treatments. This difference was significant (Fig. 1), but none of the measured yield components were significant (Table 2). Nevertheless, the main difference in yield components of KNL between the highest and the lowest N treatment was the number of grains per panicle (Table 2b), which explains the yield increase for this variety. The data suggest that KNL cannot increase the number of panicles initiated in response to N, but is able to increase the size of the panicles in response to N, and maintain the proportion of filled grain in the larger panicles.

KNL showed the lowest yield of the four varieties (Fig. 1), and relative to HNN and the improved varieties, the data in Table 2 indicates that this is due to the lower grain weight of KNL. The lower yield of HNN relative to the improved varieties is most likely to be due to the lower number of grains per panicle (Table 2b).

Quality

The main quality trait of the traditional varieties used in the present study that is prized by Lao consumers is fragrance. Although over 100 volatile compounds have been detected from rice, the compound that contributes the most to fragrance is 2AP[20]. It has previously been reported that the amount of 2AP in KNL is higher than in HNN[21]. This was also found in the present study at N levels of 30 kg/ha and greater (Fig. 2). The major gene for aroma is *betaine aldehyde dehydrogenase* (*BADH2*)[22]. At least 10 alleles of the *BADH2* gene are known to lead to different amounts of 2AP[17]. HNN has the most common allele for *BADH2*, but the genetic basis of aroma in KNL has yet to be identified[17,21]. Perhaps the gene for aroma in KNL is more active than in HNN, leading to higher amounts of 2AP.

The amount of 2AP that accumulates in grains is affected by environmental and management conditions[23]. Fig. 2 shows that the concentration of 2AP was not affected by N treatment for either variety, so either these two varieties are not affected by N or the accumulation of aroma is not affected by 2AP. The *BADH2* gene is expressed in the grain[24], so differences in panicle number or grains per panicle are not expected to dilute the amount of 2AP produced in grains of HNN. Although the genetic basis of aroma in KNL is unknown, the increased panicle size of KNL in the different N treatments did not have any significant affect on the high level of 2AP in the grains of KNL (Fig. 2), suggesting that the gene for aroma in KNL is also expressed in the endosperm.

Instrumental measurements of hardness and stickiness indicate that the samples used in this study did not respond to N treatment in terms of hardness and stickiness. Varietal differences, however, were observed. The hardness of TSN1 was similar to that of KNL (Table 3c) and its stickiness was similar to that of HNN (Table 3d). Comparisons with TDK1 could not be conducted in this study because the amount of milled whole grains was too low in the 0 kg N/ha trial. Nevertheless, hardness and stickiness of grains from N treatments with sufficient samples suggest that the texture of TDK1 is similar to that of HNN.

Rapid viscosity analysis (RVA) is generally used to predict a number of traits of eating quality of non-waxy rice[12]. The development of the viscosity profile has been described previously[25]. In brief, viscosity begins as starch granules absorb water and proteins swell. As temperature increases, the starch granules swell rapidly and amylose leaches from the granules, resulting in the initial increase in viscosity. Peak viscosity is reached when the rate at which the starch granules swell is about the same as the rate at which they burst. After this, disrupted granules start to align under constant shear and temperature, leading to a decrease in viscosity. The lowest point of this decrease is the trough viscosity. As the temperature decreases, viscosity begins to increase as the starch granules retrograde. At this stage, the leached amylose and other molecules form networks. The last point in the viscosity curve is the final viscosity.

It has previously been shown using a number of non-waxy varieties that N fertilizer decreases the peak and final viscosity of the curve disproportionately[13] suggesting that there may be grain components in non-waxy varieties that could be responsive to N. However, waxy varieties show quite different curves from non-waxy varieties[26]. In particular, the viscosity profiles of KNL grown in the lower N treatments were lower than those of grains grown in the higher N treatments (Fig. 3d). Moreover, curves from KNL had the most positive setback. These observations show that the relationship between N fertilizer and viscosity in non-waxy varieties was not observed in most of the waxy varieties used in the present study, which suggests that the usual correlations between RVA and quality traits might not apply to waxy rice.

The difference in viscosity profiles between KNL grown in high and low N treatments is perhaps due to the significantly lower protein content of the grains from the two lower rates of N compared with the grains from the two higher rates (Table 3b). Proteins absorb a large amount of water as they swell during heating[27,28], and the more protein in the grain, the more water required for cooking[29]. When water content is not increased to account for protein, as is usually the case when using the RVA, higher levels of protein cause the paste to become thicker, leading to higher viscosity curves[13]. The three other varieties did not show significant differences in protein content due to N rate, and perhaps consequently, did not show significant differences in the thickness of the paste formed during the RVA.

Small differences are found in some of the derived viscosity parameters between the varieties (Fig. 3), in particular, breakdown, setback, and retrogradation. The values for KNL are lower than for the other varieties, and perhaps this is because the whole viscosity curve is lower (Fig. 3), suggesting a more dilute paste requiring less force to stir. The cooked rice grains of KNL are slightly softer than those of the other varieties (Table 3c), but softness is more usually correlated with setback than with any other parameter of the curve[12,30–34]. N rate did not significantly affect the derived parameters tested within a variety, suggesting that the lower curves of the low N KNL grains are actually the same shape as the curves of the higher N grains, but made from a weaker paste.

Fig. 3c shows the viscosity traces of HNN and shows that the final viscosity rises from the trough significantly more than it does for the other varieties, leading to a higher final viscosity. This rise from the trough to the final viscosity is usually associated with firmness of the cooked rice [12], which is usually associated with amylose content [25,34–36]. Waxy rice does not contain amylose due to a mutation in the *Waxy* gene [2], but Fig. 4 shows that there is an association between hardness of cooked grains and the rise from the trough to the final viscosity. This must be due to structures in the grain other than amylose. HNN grains have lower average protein than the other three varieties (Table 3b). Examination of the data reported in Ref. 13 indicates that the lower the protein content of the grain, the higher is the difference between the final and trough values. However, removal of proteins from waxy varieties of rice can have very different effects, depending on the variety [25,37].

By only using waxy varieties to construct Fig. 4, interesting information about viscosity curves emerges. Strong correlations were found between all the direct parameters of peak, trough, and final viscosities and the derived parameters of breakdown, retrogradation, and setback (Fig. 4). These correlations indicate that the initial swelling of starch granules to reach a balance between shear and swelling, the peak, determines the degree to which the molecules undergo shear-thinning to the trough, and then this determines how the molecules of protein and amylopectin interact to rise to the final viscosity. Therefore, using RVA curves to predict the quality of waxy rice requires an understanding of what, in a waxy matrix, causes the differences in the ability of the granules from different varieties to swell and resist shear. Hardness of the cooked grains correlates well with peak and trough viscosity and quite well with other parameters. Thus understanding how the peak forms will then provide a screening tool to differentiate between the hardnesses of cooked waxy rices.

CONCLUSIONS

In the present study, it was demonstrated that N treatment affected the yield of the four varieties used, but an effect on quality was less apparent. The improved varieties used in the present paper, TDK1 and TSN1, were more responsive to N treatment than the traditional varieties HNN and KNL. The increase in yield among the varieties arose through different responses in yield components, such as grain weight and number of grains per panicle. The yield increase of TSN1 in response to N treatment makes it a more viable candidate in improving grain quality in Lao PDR. Quality parameters measured in this study were mostly affected by varietal effects rather than by N treatment. Aroma, textural attributes (hardness and stickiness), gelatinization temperature, and viscosity parameters were mostly non-responsive to N treatment. However, the viscosity response to N in KNL is attributed to changes in protein content associated with differences in N treatment. Taken together, the data indicate that further improvements to food security in Lao PDR could be achieved by combining the quality of KNL or HNN with the yield of TSN1. Moreover, a breeding programme attempting to capture the quality traits of a waxy variety into a high-yielding variety requires that tools to evaluate quality be developed and current tools customized to screen for the desired quality traits in waxy rices to enable breeders to use quality as a selection tool.

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