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# Correlation Analysis of Mineral Element Contents and Quality Traits in Milled Rice (*Oryza stavia* L.)

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The relationships among potassium (K), calcium (Ca), sodium (Na), magnesium (Mg), iron (Fe), zinc (Zn), copper (Cu), and manganese (Mn) contents in milled rice (Oryza stavia L.) of 274 genotypes and the relationships between these mineral element contents and other rice quality traits including 3 cooking quality traits, 17 amino acid contents, and protein content were investigated. The results showed that there were significant correlations among most of mineral element contents. Mg, Fe, and Mn contents were significantly correlated with most of the other mineral element contents, while Cu content had significantly negative associations with the K and Mg contents of rice. The relationships between mineral element contents and cooking quality traits showed that gel consistency (GC) was significantly correlated with K, Cu, and Mn contents of rice. Amylose content (AC) was significantly associated with K, Na, Mg, Cu, and Mn contents. The alkali spreading value (ASV) had closely positive relationships with Ca, Mg, and Mn contents. In addition, 8 mineral element contents had obvious correlations with different amino acid contents. Mg, Ca, and Zn contents were significantly correlated with most of the 17 amino acid contents, but Na content did not correlate with amino acid contents except aspartic acid of rice. Furthermore, significant associations were found between protein content and Na, Mg, Zn, Cu, or Mn content. Six principal components were extracted to explain 84.50% of the total variances and contained the information provided by the original 29 variables according to the principal component analysis.

KEYWORDS: Milled rice; mineral element; grain quality; correlation; principal component analysis

### INTRODUCTION

Rice is a key food for human nutrition because it supplies starch, protein, and the majority of micronutrients to humans, particularly in Asia (1, 2). In recent decades, genetic improvement of rice grain quality is important for rice breeding, and considerable progress has been made in quality breeding (3). However, rice scientists have long recognized its micronutrient deficiencies, which are the basis of numerous human health problems worldwide. Mineral nutrient deficiencies have egregious societal costs including learning disabilities among children, increased morbidity and mortality rates, lower worker productivity, and added high health care costs, all of which diminish human potential, felicity, and national economic development (4). In recent years, attention has turned to strategies for improving human vitamin and mineral nutrition, especially Fe, Zn, selenium (Se), and iodine (I) (5, 6).

Since 1992, researchers at IRRI have been evaluating the genetic variability of Fe and Zn content in rice grain (7, 8).

There were obvious differences in Fe and Zn contents among the genotypes tested, suggesting a genetic potential to increase contents of these micronutrients in rice grain (9). Furthermore, a significantly positive correlation was found between Fe and Zn content (10, 11). Researches also indicated a slight linkage between aroma and the high-Fe trait (on chromosomes 7 and 8) (6) and showed that higher phytic acid content inhibits mineral absorption, especially for Fe and Zn, in cereals and legume seeds (12, 13). Zhou et al. (2003) revealed positive correlations between protein content and Cu or Zn content and between amino acid contents and Cu or Zn content (14). Zeng et al. (2005) reported that mineral element contents of 653 Yunnan rice accessions were closely related to most quality traits, including significant correlations between amylose content and K, between protein content and Mg, Cu, or Mn content, between gel consistency and Fe or Zn content, and between gelatinization temperature and Mn content (15). Zhang et al. (2004) found that indirect selection of grain characteristics may be one of the breeding methods to select for Fe, Zn, and Mn content in black pericarp indica rice (16). Thus, it is important to improve understanding of the relationships between mineral nutrients and rice quality and select rice genotypes appropriate for breeding program.

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The objective of this study was to analyze the relationships among mineral nutrient contents of K, Ca, Na, Mg, Fe, Zn, Cu, and Mn in milled rice (*Oryza stavia* L.) of 274 genotypes as well as the relationships between these mineral element contents and other rice quality traits including 3 cooking quality traits (GC, AC, and ASV), 17 amino acid contents (aspartic acid, threonine, serine, glutaminic acid, glycine, alanine, cysteine, valine, methionine, isoleucine, leucine, tyrosine, phenylalanine, lysine, histidine, arginine, and proline), and protein content.

#### MATERIALS AND METHODS

**Materials.** The 274 rice genotypes were collected and planted on the experimental farm of Zhejiang University, Hangzhou, China. Fertilizer and water management in paddy fields were similar with the conventional practice of rice. The rice grains were dehusked in an electrical dehusker (Model B-76; Zhejiang Taizhou Food Instrument Factory, China) and milled by a sample miller (Model JB-20; Zhejiang Taizhou Food Instrument Factory, China). The milled rice of samples was ground to 100 mesh with a model 3010-019 cyclone grinder (Udy Crop., Fort Collins, CO), respectively.

Sample Treatment and Mineral Determination. Approximately 0.5 g of rice powder was weighed and placed into a crucible and then carbonized at 250 °C on an electrothermal plate until the sample was black but not smoking completely. The crucibles with samples were dry-ashed by heating in a muffle furnace at 550 °C (about 10–12 h) after sample incineration until a white residue was obtained. The residue was carefully transferred into a 50 mL volumetric flask after it was dissolved with 5 mL of 6 M HCl and then diluted to 50 mL with water. The diluted solutions were subjected to analysis for K, Ca, Na, Mg, Fe, Mn, Cu, and Zn by ICP-MS (inductively coupled plasma mass spectroscopy) (Agilent 7500A; Agilent Technologies, Inc.).

Determination of Grain Quality in Milled Rice. The moisture contents of all sample flours were balanced to about 12%. All of the sample flours were scanned by near-infrared reflectance spectroscopy (NIRS; model 5000 monochromator; NIRSystem, Silver Spring, MD). The calibration models for rice cooking qualities, including amylose content (AC, %), gel consistency (GC, mm), and alkaline spread value (ASV, grade), were well developed with standard errors of calibration (SEC) and calibration coefficient (RSQ) of 1.18% and 0.96, 8.32 mm and 0.81, and 0.74 grade and 0.85, respectively (17). Wu et al. (2002) have developed calibration models to evaluate the content of 15 amino acids: asparagine, threonine, serine, glutamine, glycine, alanine, cysteine, valine, isoleucine, leucine, tyrosine, phenylalanine, lysine, arginine, and proline (18). The results showed a higher RSQ (0.78-0.98) and lower standard errors of cross-validation (SECV) with 3 g samples of milled rice flour. With samples of high amino acid diversity, the calibration models for histidine and methionine were successfully developed with RSQ and SECV of 0.73 and 0.32 (mg  $g^{-1}$ ) and 0.71 and 0.55 (mg  $g^{-1}$ ), respectively. The calibration models for protein content (mg  $g^{-1}$ ) were also well developed with a high RSQ of 0.95 and a low SEC of 4.81 mg  $g^{-1}$ . In terms of the above calibration models, 21 different rice quality traits for each sample were determined by NIRS on platform WinISI II, version 1.04 (Infrasoft International Inc., Port Matila, PA).

**Statistical Analyses.** Data analyses were conducted using SPSS (version 11.0; Chicago, IL). We performed initial descriptive statistics, including mean, standard deviation (SD), and minimum and maximum values. Pearson correlation analyses were carried out among the contents of 8 mineral elements and between 8 mineral element contents and 21 rice grain quality traits. *P* values were two-tailed, and two significant levels were using P = 0.05 and 0.01. Principal component analysis was performed on the basis of the correlation matrix and the calculated eigenvalues and eigenvectors. The principal components were extracted with eigenvalues >1.0.

#### RESULTS

Variation in Mineral Contents in Milled Rice of Different Genotypes. The mean, standard deviation (SD), and content

 Table 1. Variation in Mineral Contents of Milled Rice among Rice
 Genotypes

mineral elements	no. of samples	mean (µg/g)	standard deviation (µg/g)	range (µg/g)
K	274	804.83	275.00	302.71-1832.75
Ca	274	119.50	47.41	42.27-341.70
Na	274	20.78	12.58	4.10-79.87
Mg	274	194.79	76.34	63.31-539.35
Fe	274	5.40	2.88	0.98-26.78
Zn	274	25.97	5.73	13.32-43.65
Cu	274	9.96	4.72	3.16-24.58
Mn	274	10.73	3.15	4.89-25.97

Table 2. Correlations among Eight Mineral Element Contents of Milled Rice

element	К	Ca	Na	Mg	Fe	Zn	Cu
Ca	0.049						
Na	0.060	0.329 <sup>b</sup>					
Mg	0.381 <sup>b</sup>	0.381 <sup>b</sup>	0.237 <sup>b</sup>				
Fe	0.121 <sup>a</sup>	0.170 <sup>a</sup>	0.196 <sup>b</sup>	0.127 <sup>a</sup>			
Zn	0.051	0.226 <sup>b</sup>	0.090	0.416 <sup>b</sup>	0.126 <sup>a</sup>		
Cu	$-0.338^{b}$	0.094	-0.079	-0.193 <sup>b</sup>	0.061	0.123 <sup>a</sup>	
Mn	0.338 <sup>b</sup>	0.249 <sup>b</sup>	-0.006	0.281 <sup>b</sup>	0.242 <sup>b</sup>	0.277 <sup>b</sup>	0.121 <sup>a</sup>

<sup>a</sup> Significant at 0.05 probability level. <sup>b</sup> Significant at 0.01 probability level.

range of K, Ca, Na, Mg, Fe, Zn, Cu, and Mn in milled rice (*O. stavia* L.) of 274 genotypes were summarized in **Table 1**. The mean content values in milled rice were as follows: K > Mg > Ca > Zn > Na > Mn > Cu > Fe. The content means of K, Ca, Na, and Mg were 804.83, 119.50, 20.78, and 194.79 µg/g, respectively, which were macronutrients for the human body. The elements of Fe, Zn, Cu, and Mn were micronutrients for the human body with mean values of 5.40, 25.97, 9.96, and 10.73 µg/g in milled rice, respectively.

Visible differences in mineral element contents were found among the genotypes studied. Among the macronutrients, Na content showed the largest relative difference and presented as the ratio of the maximum/minimum content with 19.48-fold. The contents of Mg, Ca, and K varied less among genotypes than Na, with ratios of the maximum/minimum content of 8.52-, 8.08-, and 6.05-fold, respectively. According to the selection criteria (mean  $\pm$  standard deviation) (1), G7/Zhefu 01 F<sub>8</sub>, G99-242/Zhenong 996 F<sub>9</sub>, and Jin97-47/Zhefu 01 F<sub>9</sub> had relative higher contents of K, Ca, Na, or Mg. Of the micronutrients, Fe content had the largest relative difference with a ratio of 27.37fold among rice genotypes, and Cu content ranked second with a relative difference of 7.79-fold. In contrast, Zn content had the smallest difference of all of the measured elements, with a ratio of 3.28-fold. Among 274 genotypes, G7/Zhefu 01 F8 and Chunjiang13 contained dense Fe, Zn, and Mn contents, Zhenong 37 and Taizhan were rich in Fe, Zn, and Cu contents, and Heinuomi had relative higher Fe, Cu, and Mn contents. The differences in mineral nutrient contents among genotypes might be due to different genetic resources involved.

**Correlations among the Contents of Eight Mineral Elements in Milled Rice.** To find relationships among 8 mineral element contents in milled rice, Pearson correlation analyses were performed for the accessions (**Table 2**). Among macronutrients of K, Mg, Ca, and Na, there were significantly positive correlations between the contents of K and Mg, between the contents of Ca and Na or Mg, or between the contents of Mg and Na, whereas no close correlations were found between K content and Ca or Na content. Among the micronutrients, closely

Table 3.	Variation	in Rice	Quality	Traits among	Rice	Genotypes
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quality traits	mean	standard deviation	range
GC (mm)	54.38	23.02	2.35-100.00
AC (%)	17.60	8.50	0.07-37.78
ASV (grade)	5.12	1.71	1.42-9.81
aspartic acid (mg/g)	7.67	1.95	2.81-12.03
threonine (mg/g)	3.59	0.69	1.62-5.18
serine (mg/g)	4.91	1.01	1.90-7.27
glutaminic acid (mg/g)	14.69	2.85	6.75-21.92
glycine (mg/g)	7.06	1.27	3.38-10.14
alanine (mg/g)	7.79	1.00	5.72-11.47
cysteine (mg/g)	0.73	0.36	0.03-1.43
valine (mg/g)	6.55	0.94	4.21-9.23
methionine (mg/g)	1.27	0.23	0.69-1.93
isoleucine (mg/g)	3.85	0.77	1.77-5.76
leucine (mg/g)	6.92	1.44	2.92-10.56
tyrosine (mg/g)	2.84	0.90	0.69-5.12
phenylalanine (mg/g)	3.30	0.84	0.97-5.32
lysine (mg/g)	3.02	0.62	1.18-4.34
histidine (mg/g)	2.46	0.34	1.55-3.33
arginine (mg/g)	5.23	1.30	1.50-8.26
proline (mg/g)	4.57	0.89	2.02-6.70
protein (mg/g)	99.06	18.59	57.87-174.21

positive associations were recognized between the contents of Fe and Zn or Mn, between the contents Zn and Mn or Cu, and between the contents Mn and Cu, while there was no visible correlation between Cu and Fe contents. These results suggested that high Fe content might be accompanied with high Zn and Mn contents of rice.

The relationships between macronutrients and micronutrients were indicated that there were significantly negative associations between the contents of Cu and K ( $r = -0.338^{b}$ ) or Mg (r = $-0.193^{b}$ ), whereas visibly positive correlations were recognized among most of the other mineral elements (Table 2). It was probably due to the interaction between ions whose chemical properties were sufficiently similar, and they compete for site of absorption, transport, and function in plant tissues (19–21). Kupper et al. (1998) reported that the heavy metals (Hg, Cu, Cd, Ni, and Pb) might make substitution of Mg, the central atom of chlorophyll. They might take place specifically in PS II reaction centers and lead to a breakdown in photosynthesis (22). According to the result of Gussarsson and Jensen (1992), Cu affected K<sup>+</sup> influx and induced inhibition of active K<sup>+</sup> influx. Passive  $K^+$  influx was also decreased with uptake of Cu (23). Fe content was significantly associated with K, Ca, Na, and Mg contents, and Mn content was closely related to K, Ca, and Mg contents. Likewise, significantly positive correlations were observed between the contents of Zn and Ca or Mg. It implied that Mg content of rice grain might play an important role since the correlations were all significant between Mg and others, which might be explained that Mg regulated the uptake of the other essential elements (24). For example, there was synergistic behavior of Mg on the K concentration. It might be explained as Mg in plants from Mg-ATP complexes, which was required for active absorption of K across cells (25). The result of Kennedy and Gonsalves (1987) showed that  $Zn^{2+}$  with the presence of adequate Mg<sup>2+</sup> could be beneficial to nutrient uptake by maintaining a higher membrane potential (26).

**Relationships between Mineral Element Contents and Quality Traits of Rice.** The mean, SD, and range for 3 cooking quality traits, contents of 17 amino acids, and protein in milled rice evaluated by NIRS are shown in **Table 3**. The results revealed that cooking quality traits varied widely, with GC ranging from 2.35 to 100.00 mm (mean =54.38 mm, SD = 23.02 mm), AC from 0.07% to 37.78% (mean = 17.60%, SD = 8.50%), and ASV from 1.42 to 9.81 grade (mean = 5.12 grade, SD = 1.71 grade). On average, 17 amino acid contents ranged from 0.73 to 14.69 mg/g, with glutaminic acid content having a relatively high mean value (14.69  $\pm$  2.85 mg/g) and cysteine content being the lowest (0.73  $\pm$  0.36 mg/g). The protein content mean was 99.06 mg/g and ranged from 57.87 to 174.21 mg/g.

The relationships between mineral element contents and cooking quality traits were analyzed by the Pearson correlation analysis method (Table 4). GC was significantly positively correlated with K and Mn, whereas it was notably negatively associated with Cu. Significantly positive correlations were found between AC and Na content or Cu content and negative associations between AC and K, Mg, or Mn content. ASV was significantly positively correlated with Ca, Mg, and Mn contents with correlation coefficients (r) of  $0.262^{b}$ ,  $0.146^{a}$ , and  $0.131^{a}$ , respectively. These results suggested that some mineral elements especially for Mn content of milled rice were strongly correlated with cooking quality traits. These relationships between the mineral element contents and quality traits of rice might reflect the physiological function of the mineral elements (24, 27). For example, K is essential for photosynthesis and has a role in carbohydrate metabolism. It also facilitates cell division and growth by helping to move starches and sugars between plant organs. Mg is a constituent of chlorophyll, which is the driving force of photosynthesis. Cu and Mn also act as enzyme cofactors and are thought to be involved in chlorophyll formation, reflecting their important roles in photosynthesis.

In addition to cooking quality traits, some visible relationships between the contents of 17 amino acids and protein and these 8 mineral element contents were also found in milled rice (Table 4). Of the macronutrients of K, Ca, Na, and Mg, Mg content was all significantly positively correlated with each of 17 amino acid contents and also with protein content. There were no close associations between Na content and 17 amino acid contents except for aspartic acid, but a significant negative correlation was found between Na content and protein content (r = $-0.133^{a}$ ). Significantly positive correlations were observed between Ca content and all of the amino acid contents except for methionine content, while K content was only obviously positively associated with aspartic acid, serine, cysteine, methionine, tyrosine, histidine, and proline. Of micronutrients, Zn content had the most closely positive correlations with 17 amino acid contents except for cysteine content. Fe content was significantly positively correlated with some amino acids including alanine, cysteine, methionine, and tyrosine content. Cu content showed closely positive correlation with alanine content and negative association with cysteine or methionine content. Likewise, Mn content was significantly positively related to serine, cysteine, methionine, tyrosine, or histidine content. Furthermore, the contents of Zn, Cu, and Mn also exhibited significantly positive associations with protein content of milled rice. These results suggested that these mineral nutrients especially for micronutrients of rice grain might be effectively improved with some amino acid contents in rice breeding programs. It implied that the mineral elements were involved in nitrogen metabolism (15, 24, 27). Mg is essential for carbohydrate metabolism and also serves as a carrier of phosphate compounds during plant growth. Ca aids in carbohydrate transport and nitrogen uptake. Zn is a component of enzyme systems and is also essential for carbohydrate metabolism and regulating sugar consumption. Cu is involved in protein synthesis and utilization. Mn is a component of enzyme systems involved in carbohydrate breakdown and nitrogen metabolism.

Table 4. Correlation Coefficients between the Mineral Element Contents and Quality Characteristics of Milled Rice

items	K	Ca	Na	Mg	Fe	Zn	Cu	Mn
GC	0.208 <sup>b</sup>	0.038	-0.107	0.060	0.084	-0.097	-0.243 <sup>b</sup>	0.142 <sup>a</sup>
AC	$-0.332^{b}$	0.060	0.153 <sup>b</sup>	-0.170 <sup>b</sup>	-0.119	0.045	0.251 <sup>b</sup>	-0.237 <sup>b</sup>
ASV	0.105	0.262 <sup>b</sup>	0.097	0.146 <sup>a</sup>	0.006	-0.069	0.031	0.131 <sup>a</sup>
aspartic acid	0.183 <sup>a</sup>	0.350 <sup>b</sup>	0.154 <sup>a</sup>	0.508 <sup>b</sup>	0.030	0.228 <sup>b</sup>	-0.065	-0.050
threonine	0.111	0.421 <sup>b</sup>	0.075	0.445 <sup>b</sup>	0.061	0.150 <sup>a</sup>	0.021	0.083
serine	0.133 <sup>a</sup>	0.422 <sup>b</sup>	0.059	0.446 <sup>b</sup>	0.085	0.165 <sup>b</sup>	0.041	0.126 <sup>a</sup>
glutaminic acid	0.089	0.397 <sup>b</sup>	0.050	0.426 <sup>b</sup>	0.067	0.194 <sup>b</sup>	0.067	0.074
glycine	0.110	0.415 <sup>b</sup>	0.076	0.453 <sup>b</sup>	0.063	0.185 <sup>b</sup>	0.032	0.081
alanine	0.053	0.292 <sup>b</sup>	-0.014	0.357 <sup>b</sup>	0.122 <sup>a</sup>	0.297 <sup>b</sup>	0.122 <sup>a</sup>	0.053
cysteine	0.234 <sup>b</sup>	0.285 <sup>b</sup>	-0.002	0.336 <sup>b</sup>	0.124 <sup>a</sup>	0.004	-0.148 <sup>a</sup>	0.234 <sup>b</sup>
valine	0.060	0.365 <sup>b</sup>	0.010	0.401 <sup>b</sup>	0.092	0.242 <sup>b</sup>	0.079	0.060
methionine	0.284 <sup>b</sup>	0.073	-0.053	0.332 <sup>b</sup>	0.208 <sup>b</sup>	0.247 <sup>b</sup>	-0.148 <sup>a</sup>	0.284 <sup>b</sup>
isoleucine	0.115	0.407 <sup>b</sup>	0.040	0.432 <sup>b</sup>	0.097	0.193 <sup>b</sup>	0.067	0.115
leucine	0.097	0.400 <sup>b</sup>	0.042	0.425 <sup>b</sup>	0.084	0.196 <sup>b</sup>	0.072	0.097
tyrosine	0.168 <sup>a</sup>	0.380 <sup>b</sup>	0.002	0.395 <sup>b</sup>	0.143 <sup>a</sup>	0.124 <sup>a</sup>	0.002	0.277 <sup>b</sup>
phenylalanine	0.095	0.391 <sup>b</sup>	0.033	0.431 <sup>b</sup>	0.070	0.170 <sup>b</sup>	0.011	0.074
lysine	0.103	0.422 <sup>b</sup>	0.119	0.421 <sup>b</sup>	0.064	0.138 <sup>a</sup>	0.050	0.110
histidine	0.129 <sup>a</sup>	0.397 <sup>b</sup>	0.037	0.447 <sup>b</sup>	0.106	0.214 <sup>b</sup>	0.058	0.167 <sup>b</sup>
arginine	0.098	0.399 <sup>b</sup>	0.059	0.439 <sup>b</sup>	0.065	0.165 <sup>b</sup>	0.015	0.075
proline	0.125 <sup>a</sup>	0.418 <sup>b</sup>	0.055	0.442 <sup>b</sup>	0.083	0.170 <sup>b</sup>	0.048	0.119
protein	0.005	-0.043	-0.133 <sup>a</sup>	0.125 <sup>a</sup>	0.109	0.300 <sup>b</sup>	0.148 <sup>a</sup>	0.128 <sup>a</sup>

<sup>a</sup> Significant at 0.05 probability level. <sup>b</sup> Significant at 0.01 probability level.

Table 5. Principal Component Analysis for All Principal Components<sup>a</sup>

component	eigenvalue	variance (%)	cumulative variance (%)
1	15.3562	52.9524	52.9524
2	2.9211	10.0727	63.0252
3	2.0672	7.1283	70.1534
4	1.8004	6.2084	76.3618
5	1.3400	4.6207	80.9825
6	1.0209	3.5203	84.5028
7	0.909	3.1354	87.6382
8	0.8200	2.8274	90.4656
9	0.6084	2.0979	92.5635
10	0.5541	1.9108	94.4743
11	0.5199	1.7927	96.2670
12	0.3908	1.3475	97.6145
13	0.3151	1.0865	98.7010
14	0.1742	0.6006	99.3016
15	0.1032	0.3558	99.6573
16	0.0416	0.1435	99.8008
17	0.0206	0.0709	99.8717
18	0.0143	0.0492	99.9209
19	0.0120	0.0414	99.9623
20	0.0051	0.0175	99.9799
21	0.0028	0.0096	99.9895
22	0.0013	0.0045	99.9940
23	0.0011	0.0036	99.9976
24	0.0004	0.0013	99.9989
25	0.0002	0.0006	99.9994
26	0.0001	0.0003	99.9997
27	0.0001	0.0002	99.9999
28	0.0000	0.0001	100.0000
29	0.0000	0.0000	100.0000

<sup>a</sup> On the basis of the correlation matrix, 29 principal components were created. The eigenvalues of the first 6 principal components were all >1.0, and they accounted for over 80% of the total variance. Thus, the first 6 principal components were named PC1, PC2, PC3, PC4, PC5, and PC6.

**Principal Component Analysis of Mineral Element Contents and Quality Traits of Rice.** Principal component analysis was performed on all of the 29 variables, including 8 mineral element contents, 3 cooking quality traits, 17 amino acid contents, and protein content. **Table 5** contains all 29 principal components and their corresponding eigenvalues and variances. The results indicated that the eigenvalues of the first 6 principal components were all >1.0 and they accounted for over 80% of the total variance. The meaning of these 6 components can be distilled from their respective eigenvectors (**Table 6**).

Table 6. Sou	urces of V	Variation	for the	First Six	Principal	Components
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	eigenvectors of principal component							
variable	PC1	PC2	PC3	PC4	PC5	PC6		
К	0.0419	0.2908	0.1871	0.1983	-0.3291	0.3978		
Ca	0.1116	0.0234	0.0793	0.4159	0.2514	-0.1287		
Na	0.0178	-0.0269	0.0772	0.5299	-0.0682	-0.4087		
Mg	0.1261	0.1223	0.2664	0.3198	-0.2678	0.1099		
Fe	0.0297	0.1024	0.2761	0.1245	0.2799	-0.3226		
Zn	0.0573	-0.0573	0.4424	0.0939	-0.0035	0.0297		
Cu	0.0061	-0.2469	0.0911	-0.0820	0.6125	0.0722		
Mn	0.0444	0.2030	0.3700	0.0880	0.3827	0.3612		
GC	0.0411	0.4810	-0.1843	-0.1118	0.1132	-0.0218		
AC	-0.0693	-0.4856	0.0800	0.1595	-0.0691	-0.0782		
ASV	0.1025	-0.0205	-0.2358	0.2691	0.1866	0.5186		
aspartic acid	0.2215	-0.0895	-0.0633	0.1125	-0.2082	-0.0020		
threonine	0.2491	-0.0316	-0.1057	0.0335	-0.0052	-0.0109		
serine	0.2507	-0.0164	-0.0730	0.0097	0.0173	0.0069		
glutaminic acid	0.2474	-0.0802	-0.0598	-0.0119	-0.0147	0.0158		
glycine	0.2502	-0.0644	-0.0689	0.0246	-0.0268	0.0129		
alanine	0.2207	-0.1106	0.1760	-0.1908	-0.0259	-0.0228		
cysteine	0.1522	0.3639	-0.0549	-0.0484	0.1123	-0.2286		
valine	0.2436	-0.0620	0.0424	-0.1178	-0.0060	-0.0554		
methionine	0.1132	0.2270	0.3575	-0.2653	-0.1183	-0.1821		
isoleucine	0.2519	-0.0386	-0.0230	-0.0358	0.0199	0.0059		
leucine	0.2504	-0.0649	-0.0362	-0.0291	0.0052	0.0148		
tyrosine	0.2304	0.1369	0.0000	-0.0768	0.1063	-0.0772		
phenylalanine	0.2481	-0.0059	-0.0883	-0.0306	-0.0138	-0.0633		
lysine	0.2370	-0.0942	-0.0744	0.0962	0.0147	0.0713		
histidine	0.2517	-0.0222	0.0153	-0.0626	0.0060	-0.0208		
arginine	0.2494	-0.0404	-0.0862	0.0075	-0.0207	-0.0188		
proline	0.2504	-0.0252	-0.0709	0.0030	0.0147	0.0049		
protein	0.0888	-0.2434	0.3853	-0.3101	-0.1218	0.1514		

The first principal component (PC1) was the most important, which explained 52.95% of the total variance. Most of the amino acid contents correlated positively with PC1. Thus, PC1 was represented with amino acid contents in milled rice. The second principal component (PC2) accounted for an additional 10.07% of the total variance. The variation was mainly attributed to GC and AC, for which the eigenvectors were 0.4810 and -0.4856, respectively. These suggested that PC2 correlated positively with GC and negatively with AC of rice. Thus, PC2 stood for the cooking quality traits of rice. The third principal component (PC3) accounted for 7.13% of the total variance. The variation was mainly contributed by Zn, Mn, and protein

content and secondly by Mg, Fe, and methionine content, so PC3 was mostly represented with Zn, Mn, and protein content. The fourth principal component (PC4) explained 6.21% of the total variance. The variation was mainly generated by Na, Ca, and Mg contents. At the same time, PC4 had negative correlations with protein and methionine. Thus, PC4 represented macronutrients of Na, Ca, and Mg contents and also stood for protein and methionine content. The fifth principal component (PC5) explained 4.62% of the total variance. The variation was mainly due to Cu content and secondly to Mn and K contents. The eigenvector of Cu content was up to 0.6125, so PC5 primarily took the place of Cu content. The sixth principal component (PC6) accounted for an additional 3.52% of the total variance. The variation was mainly attributed to the alkaline spread value, for which the eigenvector was 0.5186. Taken together, these results suggested that the data collected using the original 29 variables could be condensed and represented by the first six principal components, PC1 through PC6.

With the eigenvectors of the original 29 variables, the score of the principal components could be obtained with each genotype. For example, the PC1 score of Kunshanbuxuejing/ Aixuenuo was the highest among 274 genotypes, which implied abundant amino acid contents. In the present experiment, Kunshanbuxuejing/Aixuenuo contained the highest contents of most of the 17 amino acids, and the mean of the 17 amino acid contents was up to 7.51 mg/g in milled rice. Six genotypes, including Jian 200-88/Zhe 20-110 F8, G8/Zhe20-110 F8, Zheda 025, Shaonuo 01-2, Shaonuo 119, and Yongnuo, obtained the higher scores of PC2. The cooking quality data of these 6 genotypes showed that they had greater GC and lower AC. IR1552, Baliya, RR166-645, G7/Zhefu 01 F<sub>8</sub>, and Chunjiang 13 accounted for the greater PC3 scores. As a matter of fact, IR1552 contained the highest protein content, and Baliya and RR166-645 had the highest Zn contents among 274 genotypes. G7/Zhefu 01 F<sub>8</sub> and Chunjiang 13 had relatively higher Fe, Zn, and Mn content. G7/Zhefu 01  $F_8$  and G99-242/ Zhenong996 F<sub>9</sub> came up to the largest scores of PC4. Actually, G7/Zhefu 01 F<sub>8</sub> and G99-242/Zhenong996 F<sub>9</sub> had relatively higher contents of K, Ca, Na, or Mg. These results suggested that the reasonable score range of the principal components could be used for excellent germplasm selection according to the correlations between the original 29 variables and these 6 principal components.

#### DISCUSSION

The most effective approach for solving the problem of mineral nutrient deficiencies in humans is to develop rice cultivars with abundant mineral nutrients when milled. In this work, the contents of K, Ca, Na, Mg, Fe, Zn, Cu, and Mn in milled rice of 274 genotypes were evaluated. Each of these nutrients also has a critical function in plants and is required in varying amounts among different plant tissues (27). The results showed that 8 mineral element contents of rice were clearly different among genotypes, which implied that genotypic variations might provide opportunities to select for higher mineral element contents (9, 28). The Pearson correlation analysis indicated that significant correlations were found among 8 mineral element contents. In particular, Mg content was closely negatively associated with Cu content and positively correlated with the other 6 mineral element contents. Significantly positive correlations were also recognized between Ca content and Na, Mg, Fe, Zn, or Mn content. These results could probably reflect the fact that Ca provides for normal transport and retention of other elements in plant and that Mg regulates the uptake of the other essential elements (24). These results were identified with the results of previous researchers. Liu et al. (1995) reported that there was a positive relationship between the content of Zn and Ca in brown rice (29). Zeng et al. (2003) also found that Mg content was significantly positively associated with K, Ca, Fe, or Mn content, and Ca content was visibly positively associated with Mg, Fe, Zn, or Mn content (15). However, Cu content had notably negative associations with K and Mg contents of rice in the present experiments, which were quite different from the results shown by Zeng et al. (2003). It was probably due to the interaction between ions whose chemical properties were sufficiently similar and they compete for the site of absorption, transport, and function in plant tissues (19-21). Otherwise, the contents of mineral elements by our laboratory and Zeng et al. (2003) were determined with milled rice and brown rice, respectively. The loss of mineral element contents was quite different among 8 mineral elements during milling processing. In the present study, significantly positive correlations were recognized between Fe and Zn or Mn content and between Zn and Cu or Mn content. These results suggested that higher Fe, Zn, and Mn contents would likely occur simultaneously in milled rice. Wang et al. (2002) found significantly positive correlation between the contents of Zn and Mn in the polished rice (30). Jiang (2002) showed that the content of Mn, Fe, and Cu were significantly correlated with Zn content in milled rice (31). In addition, visibly positive correlation was found between Fe and Zn content (10, 11). Zeng et al. (2003) showed that there were significantly positive correlations between Fe and Zn or Mn content and between Cu and Zn or Mn content (15). These results from previous researchers were supported by the results in the present experiments. Thus, it could enhance the efficiency of selection materials which were abundant in Fe, Zn, and Mn.

Correlation analyses between 8 mineral element contents and 3 cooking quality traits showed that GC correlated significantly with K, Mn, and Cu. AC associated visibly with K, Na, Mg, Cu, and Mn. ASV was significantly positive associated with Ca, Mg, and Mn contents. In addition, Mg, Ca, and Zn contents were significantly positively correlated with most of the amino acid contents. Mg, Zn, Cu, and Mn contents were significantly positively correlated with protein content. These relationships between the mineral element contents and quality traits of rice might reflect the physiological function of the mineral elements (24, 27). These results suggested that it might offer a chance to develop a rice variety rich in mineral nutrients by indirect selection. Zeng et al. (2003) found that GC correlated significantly with K, Ca, Fe, and Zn, AC correlated visibly with K, Ca, and Mn, and ASV was significantly positively associated with K and Mn contents. Protein content had significantly positive association with K, Mg, Cu, and Mn contents (15). Zhou et al. (2003) reported that Cu and Zn content had strong positive correlations with protein and amino acid contents (14). However, Yang et al. (1998) found no close correlation between Fe, Zn, Cu, Mn content and protein, lysine content in polished rice (28). Batten (2002) found that the correlation between mineral element contents and cooking quality traits might be affected by interaction of the genotype  $\times$  environment (32). When the Mg/K ratio of rice grain and the cooking quality from 3 different seasons were analyzed, strong relationships were observed between the Mg/K ratio of brown grain and cooking quality in 2 of 3 seasons. Batten (2002) also indicated that there was not enough evidence to conclude that the Mg/K ratio of brown grain significantly affected the cooking quality of the white rice grain (32). Thus, further investigation is warranted to analyze mineral

element content in rice grains of different genotypes grown in various environments.

In the present experiment, complex correlations among mineral nutrient contents of K, Ca, Na, Mg, Fe, Zn, Cu, and Mn in milled rice (O. stavia L.) of 274 genotypes as well as their relationships with 3 cooking quality traits, protein, and amino acid contents of rice were identified. These correlations may assist future selection of appropriate rice genotypes for specific nutritional needs. Thus, principal component analysis was performed on the basis of the correlation matrix, and six principal components were found to account for a major part of the total variance. Six principal components could explain 84.50% of the total variance. The variance of the fourth principal component (PC4) was generated by contents of Na, Ca, and Mg, whereas the eigenvector of protein and methionine contents had relatively greater negative values, so the score for PC4 for genotype would not be expected to be too large because protein content in milled rice was low. Likewise, PC5 was represented with Cu content, which had a negative correlation with K content. Considering the potential toxicity of high Cu content and the affect on resistance of low K content, rice genotypes should be screened for moderate PC5 score.

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