VARIATIONS IN PROTEIN AND AMINO ACID CONTENTS AMONG GENETIC STOCK OF RICE[1]

W. F. TONG, Y. E. CHU and H. W. LI[2]

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Abstract

Dehulled grains of 17 rice strains were observed for total protein, lysine and methionine contents by the micro-kejldohl method and by using an auto-amino acid analyzer. Crude protein, ranging from about 8% to 18%, was positively correlated with both methionine and lysine contents. But, in percent of crude protein, lysine and methionine content were uncorrelated with the crude protein. Methionine content varied in a wider range than lysine content, and showed significant correlations with morphological grain character.

Introduction

Rice, the main food crop for more than half of the world population, requires improvements in protein content both qualitatively and quantitatively. It is known that animal proteins generally contain essential amino acids in a good balance (Block and Bolling 1951), whereas rice has a low protein content besides having insufficient amount of essential amino acids. To the rice diet, as a general rule, proteins and amino acids must be supplemented to make up the deficiency.

Breeding for higher protein content and better balance in amino acids has been carried out in various crops, *i. e.*, in maize by Nelson (1969), Nelson *et al.* (1965), Alexander (1966) and Alexander *et al.* (1969), in wheat by Johnson *et al.* (1963; 1967; 1968), and in barley by Hagberg and Karlessom (1969) and by Favret and Manghers (1967). In rice, a survey of varieties of crude protein and lysine contents was recently reported by the International Rice Research Institute (1967), and mutation breeding for higher protein content was reported by Swaminathan *et al.* (1969).

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(2) Research Assistant, Associate Research Fellow and Research Fellow, Institute of Botany, Academia Sinica.

The purpose of the present study is to obtain information on the variation in protein and two essential amino acid contents of rice genetic stocks.

Materials and Methods

Fourteen varieties of *Oryza sativa* including seven Indica and seven Japonica strains, and three strains belonging to different wild *Oryza* species, listed in Table 1, were used. Of them, five *sativa* strains showing high protein contents were kindly made available by Dr. G. S. Khush, the International Rice Research Institute.[3]

The plants were grown in 1969, as the first crop in Taipei area. The water content of seeds for analysis were 13.5%.

For measuring protein content, samples of dehulled grain, 0.1 grams each, were analysed by the Micro-Kjeldohl method and the percentage of nitrogen

Table 1. *Contents to total protein, lysine and methionine in the grain of different varieties of* ***Oryza sativa*** *and other Oryza species*

Species & variety	Type	Total protein % (in grains)	Lysine (%)		Methionine (%)	
			in grains	in protein	in grains	in protein
O, sativa,						
Taichung 65	Japonica	8.13	0.26	3.20	0.12	1.48
Tainan 5	Japonica	8.70	0.26	2.99	0.13	1.49
Tainon-Nuo 8	Japonica	9.70	0.33	3.40	0.13	1.34
Tainan 3	Japonica	10.67	0.40	3.75	0.14	1.31
Yunshin 1	Indica	11.23	0.45	4.01	0.12	1.67
Omirt 39	Indica	11.72	0.52	4.44	0.22	1.88
Taichung Shen 2	Indica	11.79	0.45	3.82	0.15	1.27
Yunshin 2	Indica	11.84	0.40	3.38	0.23	1.94
Tongsisai	Japonica	12.13	0.48	3.96	0.17	1.40
Taichung N. 1	Indica	12.28	0.49	3.99	0.28	2.28
Santo	Indica	13.12	0.40	3.05	0.14	1.07
Rikuto Norin 20	Japonica	13.15	0.41	3.12	0.16	1.22
Chok-Jye-bi-chal	Japonica	15.79	0.49	3.10	0.28	1.77
Crosa 2	Indica	18.46	0.60	3.25	0.32	1.73
Mean		12.05	0.42	3.53	0.185	1.52
C. V.*(%)		22.1	22.4	12.9	36.8	23.4
O. officinalis	PO16	13.89	0.56	4.03	0.34	2.45
O. australiensis	W008	12.48	0.44	3.53	0.24	1.92
O. brachyantha	P007	11.85	0.47	3.97	0.38	3.21

* Coefficient of variability

(3) We wish to thank Dr. G. S. Khush for his kind donation of seeds.

obtained was converted into percent protein by multiplying the former with a factor of 6.25 (Horwity 1960).

For determining amino acid contents, 100 mg grain samples were treated with 2 ml of 6 N HCl and sealed in the vacum tube for hydrolysis at 110°C, for 20 hours. After hydrolysis, the aliquot solution was used for the determination of amino acid contents by Auto Amino Acid Analyzer (Yanagimoto model LC–5A). All of the experiments were repated at least twice.

Results

The data for crude protein, lysine and methionine contents obtained from 17 rice strains tested are given in Table 1. Crude protein in per cent of grain weight ranged from 8.13 to 18.46%. An Indica variety, Crosa 2 showed the highest value, and a Japonica variety, Taichung 65, was the lowest.

Methionine content also showed a wide rande among the strains, varying from 0.26% to 0.60%. Lysine content varied in a narrower range, 0.12% t 0.32%. In per cent of total protein, methionine varied from about 1.1% t 2.3%, and lysine varied from about 3% to 4.4%. Methionine had larger variability coefficients than lysine, as shown in Table 1. Indica varieties seemed to contain more lysine and methionine than Japonica varieties.

Correlations among the percentages of protein, lysine and methionine, computed for 14 *sativa* varieties, are given in Table 2. As expected, total protein content was posteitively correlated with the contents of lysine and methionine. Whereas, the proportion of lysine and methionine to total protein were uncorrelated to total protein. Lysine and methionine showed different variation pattern.

Table 2. *Correlations between total protein, lysine and methionine contents and other grain characters (among 14 sativa strains)*

Character	Total Protein	Lysine % grain	Lysine % protein	Methionine % grain	Methionine % protein
Lysine, % of grain wt.	0.83**				
Lysine, % of protein	-0.12	0.45			
Methionine, % of grain wt.	0.78**	0.74**	—		
Methionine, % of protein	0.23	—	0.22	0.79**	
100 grain weight	-0.20	-0.34	-0.10	-0.67**	0.01
Length/width ratio	0.16	0.34	0.25	0.71**	0.28
Length/thickness ratio	0.24	0.38	0.22	0.66**	0.15

** Significant at 1% level

Scatter diagrams showing covariations of the above values are presented in Figs 1 and 2. It is found that among strains with less than 11% protein,

methionine content showed little variation between 0.11% and 0.14%, it varied in a wide range among strains with more than 11% protein. Lysine content did not show such a variation.

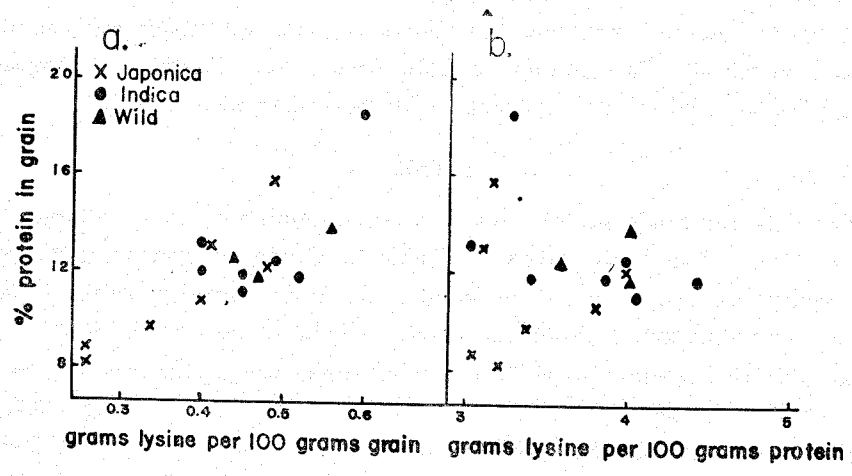

Fig. 1. The covariation relationship between protein and lysine contents

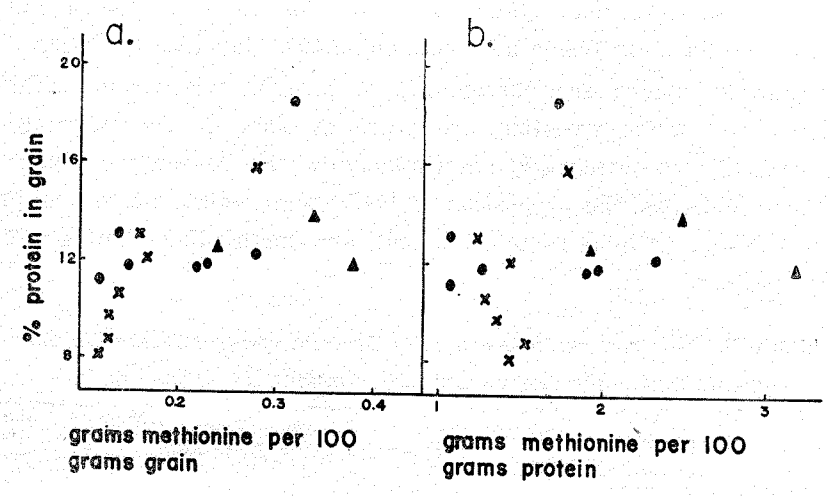

Fig. 2. The covariation relationship between protein and methionine contents

Correlations of protein and amino acid contents with grain characters are also shown in Table 2. Methionine content, in per cent of grain weight, was found to be negatively correlated with single grain weight, and positively correlated with the length/width and length/thickness ratios of grains. Total protein and lysine content did not show significant correlations with these grain characters.

The three strains of wild *Oryza* species observed for comparison had protein and amino acid contents within the range of *sativa* strains.

Discussion

Following the discovery in maize of the opaque-2 and flory-2 genes by Mertz *et al.* (1964) and Nelson *et al.* (1965), variations in protein and amino acid contents of crop varieties have aroused the keen interest of the breeders, with the hope that the nutritive value of grain crops can be improved. In barley, Hagberg and Karlsson (1869) reported that varieties varied in wide ranges of total protein (8.8–10.8%) as well as lysine content (2.8–4.6%). In rice, about 7,000 genetic stocks preserved in the International Rice Research Institute were analyzed for crude protein and 101 of them for lysine content. The concluded that protein content ranged from about 5% to 18% and lysine per unit protein from 2.9% to 4.5%. The data of the present study are consistant with those reported by the I. R. R. I. (1967).

If different genotypes of a crop plant produce the same protein in different quantities, the more the protein is, the higher would be the content of an amino acid composing the protein. Apparently, this is not the case. We find that methionine content varies in a wider range than lysine content and in per cent of total protein, methionine and lysine contents are not correlated. According to Dumanovic and Denic (1969), maize plants with a medium protein content exhibited the highest lysine cotent, and among them lysine in per cent of total protein was negatively correlated with total protein content. This negative correlation was also found among wheat varieties by Johnson *et al.* (1969). The present study also proves this very point. It may be suggested that lysine content tends to remain relatively unchanged despite wide variations in other amino acides.

The above-mentioned experimental results strongly suggest that the synthesis and the deposition of an amino acid in the grain is controlled by a particular gene system. Consequently, their amount per grain weight partly depends upon the shape and size of grains. But, grain characters do not seem to fully account for the wide variations in amino acid contents. It is found that methionine content is highly correlated to grain characters, while lysine content and total protein are not correlated to the latter. Further, the deposit of proteins and amino acids in grains may depend upon environmental conditions, particularly upon those in the period of ripening

稻米品種間蛋白質及氨基酸含量之變異

童武夫 朱耀源 李先聞

使用微量定氮法 (Micro-Kejldohl method) 和氨基酸自動分析器 (Autoamino acid analyzer)分別調查十七種不同之水稻品種種子中蛋白質(Protein)，二氨基已酸 (Lysine)

和甲硫氨基丁酸（Methionine）之含量情形。

蛋白質含量範圍約在百分之八到，百分之十八之間，而與二氨基己酸，甲硫氨基丁酸之含量成正相關。若以蛋白質，二氨基己酸甲硫，氨基丁酸之含量百分率相互比較時，則成負相關。各品種種子中甲硫氨基丁酸之分佈範圍較二氨基己酸爲大，而且和穀粒形態性狀成有意義之相關。

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