Interaction of Genetic and Zn Fertilizer Application on Rice Yield and Grain Zinc Content

U Susanto*, C Gunarsih, and W R Rohaeni

Indonesian Center for Rice Research (ICRR), Indonesian Agency for Agriculture Research and Development (IAARD), Street 9th, Sukamandi, Subang, West Java, Indonesia. Postal Code 41256

Corresponding author: *untungsus2011@gmail.com

Abstract. The prevalence of Zn deficiency in Indonesia is considerably high. Development of variety having high nutrient or called as biofortification supposed to be effective and efficient to combat malnutrition such as Zn deficiency. Zn fertilizer application was reported to be effective to increase rice Zn content. ICRR in collaboration with IRRI and Harvest Plus had initiated testing rice genotypes having high Zn content. The aim of this research was to initially study of Zn fertilizer, especially ZnSO4, effect to Zn content in the rice grains and to select genotypes having high the Zn content. The trial was conducted during Wet Season 2016 in Subang, West Java following split plot design of three replications with main plot of two level ZnSO4 fertilizer applications and sub plot of 24 rice genotypes. Transplanting was conducted at 21 days after sowing into 2 m x 3 m plot with planting space of 20 cm x 20 cm. The results showed that there is interaction between genotype and Zn fertilizer on yield but not on Zn content in rice grains. Zn fertilization tends to increase grain Zn content and yield. IR97477-115-1-CRB-0-SKI-1-SKI-0-2 (5.45 t/ha, 33.65 ppm), IR97477-81-2-CRB-0-SKI-0-SKI-0-2 (5.30 t/ha, 33.40 ppm), and IR97477-115-1-CRB-0-SKI-3-SKI-0-2 (5.56 t/ha, 32.48 ppm) has either high yield or Zn content and prospective for further testing. Correlation analysis showed that Zn is strongly correlated with Fe content. Yield is positively correlated with tiller number and Fe content and negatively correlated with heading date, maturity, plant height, and number of unfilled grain/panicle.

1. Introduction

Around one third of world people are at risk of Zn deficiency [1]. The prevalence of Zn deficiency in Indonesia is also considerably high. The prevalence of Zn deficiency in Indonesia varied according to demographical parameters, ranged from 10% to 90% [2]. It is predicted that Zn deficiency among adult in Indonesia is 74.3% [3]. Zn deficiency occurred evenly across the country, not only in rural but also in the urban area. Zn deficiency caused national loss of 0.04-0.16% of the GDB every year [4]. The loss happened due to decrease productivity, impair immune system, less motoric and intelligent capability, etc.

Development of variety having high nutrient or called as biofortification supposed to be effective and efficient in combating malnutrition such as Zn deficiency [5]. Rice is the ideal plant for Zn biofortification, since rice is staple food for mostly Indonesian people. Rice has not a sufficient Zn content to match human requirement, and mostly eating rice country has high prevalence of Zn deficiency [6]. Development of rice variety having high Zinc content is underway and intermediate
products are obtained. ICRR in collaboration with IRRI and Harvest Plus had initiated testing rice genotypes having high Zn content.

Breeding effort to increase plant capacity to absorb Zn from the soil and accumulate it in the grains is however rely on the Zn availability in the soil. Combining genetic and agronomic effort to increase Zn content in the rice grains is prospective [7, 8]. Zn fertilizer application is reported to be effective to increase rice Zn content [9]. Zn application is also reported increase yield and other agronomic traits [10]. So that, the aim of this research was to initially study of Zn fertilizer, especially ZnSO4, effect to Zn content in the rice grains and to select genotypes having high Zn content.

2. Methodology
The study was conducted during Wet Season (WS) 2016 in Subang, West Java following split plot design of three replications with main plot of two level ZnSO4 fertilizer applications (Zn0 for 0 kg and Zn1 for 3 kg ZnSO4/ha) and sub plot of 24 rice genotypes consisting of 16 elite lines and 8 varieties. Transplanting was conducted at 21 days after sowing into 2 m x 3 m plot with planting space of 20 cm x 20 cm. ZnSO4 was applied together with the third NPK fertilizer application. NPK fertilizers application followed local recommendation. Zinc content was measured into brown rice samples. Zinc content was measured using Oxford XRF Supreme which is calibrated with ICP method. Around 50 g of paddy grains were de-husked using Indian mini de-husking machine, sorted for only good healthy grains for around 10 to 20 g, put in the cups and run in the XRF machine with 1,5 minute reading for each sample.

3. Results and Discussion
The results showed that there is variation among genotypes and interaction between genotype and Zn fertilizer on yield (Table 1). It indicated that Zn application may increase yield in some specific genotypes. This experiment various effect of Zn fertilizer application to plant yield (Figure 1). However, in average Zn application give a little effect to yield and other agronomic traits, unless heading date and number of filled grain/panicle (Figure 2). Zn application caused longer period to reach heading by around three days and increase filled grain per panicle for around 4 panicle. This increase however caused not a significant increase on yield. Zn application, however caused minor effect on the Zn availability in the soil [11]. It may cause not significant increase on yield.

Table 1. Variance analysis of yield of 24 rice genotypes and ZnSO4 fertilizer application, Subang, WS 2016

| No | Source of Variation     | Df | Sum of Square | Mean Square | F Value | P Value |
|----|-------------------------|----|---------------|-------------|---------|---------|
| 1  | Zn Application          | 1  | 1.88631       | 1.88631     | 2.88    | 0.164   |
| 2  | Residual B              | 4  | 2.61563       | 0.653907    | 0.84    | 0.506   |
| 3  | Genotype                | 23 | 221.339       | 9.62344     | 12.34** | 0       |
| 4  | Zn Application x Genotype | 23 | 33.1292       | 1.4404      | 1.85*   | 0.021   |
|    | Residual                | 92 | 71.7411       | 0.779795    |         |         |
| Total |                       | 14 | 330.711       | 2.31267     |         |         |
This experiment showed variation of grain Zn content in the rice grains (Figure 3), but not significant effect of Zn fertilizer application to the grain Zn content. The interaction of genotypes and Zn application is also not present (Table 2). It is different with the previous study which reported that Zn content in rice grains could be increased by Zn application [12]. Different timing and technique of application may cause different effect to Zn content in the grains. Combining of Zn with N fertilizer had been reported increased yield and grain Zn content [13]. This experiment used relatively mild dosage of Zn applied with most practical technique of application i.e. combine with third NPK fertilizers application. It may cause only minor increase in Zn content and other agronomic traits either for naturally having high Zn content rice genotype or the lower Zn content genotypes.

Table 2. Variance analysis of Zn content of 24 rice genotypes and ZnSO4 fertilizer application, Subang, WS 2016

| No | Source of Variation         | DF | Sum of Square | Mean Square | F Value | P Value |
|----|-----------------------------|----|---------------|-------------|---------|---------|
| 1  | Zn Application              | 1  | 23.3611       | 23.3611     | 3.86    | 0.12    |
| 2  | Residual B                  | 4  | 24.2311       | 6.05778     | 1.41    | 0.24    |
| 3  | Genotype                    | 23 | 113.246       | 5.0107      | 26.31** | 0.00    |
| 4  | Zn Application x Genotype   | 23 | 6.78459       | 0.30219     | 1.58    | 0.07    |
|    | Residual                    | 92 | 3.03058       | 0.03292     | 3.03058 | 0.00    |
|    | Total                       | 143| 22.4071       | 0.1592      | 22.4071 | 0.00    |

Figure 1. Yield of 24 rice genotypes with (YLD1) and without (YLD0) ZnSO4 application, Subang, WS 2016

Figure 2. Agronomic performance and Zn/Fe content of 24 rice genotypes, average of with (1) and without (0) ZnSO4 application, Subang, WS 2016
Zn content of the tested lines was mostly higher than the most popular variety Ciherang (27.27 ppm). Nine lines had significantly higher Zn content compared to Ciherang, six lines had comparable ones, but one line lower. Inpari 23, Inpari 25, Inpari 42, and Aek Sibundong had comparable Zn content with Ciherang, but Inpari 5 Merawu, Inpari 32, and Inpari 24 had lower (Figure 3). Ciherang is consistently to have medium Zn content compared to commonly existed varieties and lines [14]. IR97477-115-1-CRB-0-SKI-1-SKI-0-2 (5.45 t/ha, 33.65 ppm), IR97477-81-2-CRB-0-SKI-0-SKI-0-2 (5.30 t/ha, 33.40 ppm), and IR97477-115-1-CRB-0-SKI-3-SKI-0-2 (5.56 t/ha, 32.48 ppm) has either high yield or Zn content and prospective for further testing. Another prospective line is IR99680-3-CRB-0-SKI-1-SKI-2-5 (5.07 t/ha, 32.52 ppm). It further proves that finding of genotypes having high yield and Zn content is technically possible [15]. IR97477-115-1-CRB-0-SKI-1-SKI-0-2 performed well in further testing [16] and had finally been release as a new variety namely Inpari IR Nutri Zinc in 2019 [17].

![Graph showing Zn content of 24 rice genotypes with (Zn1) and without (Zn0) ZnSO4 application, Subang, WS 2016](image)

**Figure 3.** Zn content of 24 rice genotypes with (Zn1) and without (Zn0) ZnSO4 application, Subang, WS 2016

Correlation analysis showed that Zn is strongly correlated with Fe content (0.725) (Table 3). High Zn content genotypes tend to have high Fe content. Similar finding had been reported in testing of Indian germplasm, lines, and released varieties [18, 19]. Zn content had not significant correlation with yield. It further indicated possibility to find high yield and Zn content simultaneously. Nevertheless, study on Philippines elite rice lines material showed negative correlation [19]. Fe content in this study had not a significant correlation with yield. It is similar with study on Philippines elite rice lines material [19]. Yield is positively correlated with tiller number and negatively correlated with heading date, maturity, plant height, and number of unfilled grain/panicle (Table 3).

4. **Conclusions**

There is interaction between genotype and Zn fertilizer on yield but not on Zn content in rice grains. Zn fertilization tend to increase grain Zn content and yield. IR97477-115-1-CRB-0-SKI-1-SKI-0-2 (5.45 t/ha, 33.65 ppm), IR97477-81-2-CRB-0-SKI-0-SKI-0-2 (5.30 t/ha, 33.40 ppm), and IR97477-115-1-CRB-0-SKI-3-SKI-0-2 (5.56 t/ha, 32.48 ppm) has either high yield or Zn content and prospective for further testing.
Table 3. Correlation among agronomic traits and Fe/Zn content of 24 rice genotypes, Subang, WS 2016

|       | Hdg   | hvst  | PH   | TN   | FG   | UG   | TG   | SS   | 1000GW | Fe     | Zn     |
|-------|-------|-------|------|------|------|------|------|------|--------|--------|--------|
| hvst  | 0.679**|       |      |      |      |      |      |      |        |        |        |
| PH    | 0.162 | 0.39  |      |      |      |      |      |      |        |        |        |
| TN    | 0.128 | -0.139| -0.588*|      |      |      |      |      |        |        |        |
| FG    | 0.072 | -0.251| -0.1 | 0.098|      |      |      |      |        |        |        |
| UG    | 0.037 | 0.282 | 0.288| -0.418*| -0.544**|      |      |      |        |        |        |
| TG    | 0.113 | 0.045 | 0.206| -0.348| 0.439*| 0.516**|      |      |        |        |        |
| SS    | 0.088 | -0.263| -0.237| 0.327| 0.828**| -0.906**| -0.125|      |        |        |        |
| 1000GW| 0.105 | 0.186 | 0.356| -0.331| -0.269| 0.312| 0.059| 0.266|        |        |        |
| Fe    | -0.574**|        | -0.237| -0.023| -0.048| -0.063| -0.117| 0.021| -0.09  |        |        |
| Zn    | -0.331| -0.205| -0.047| -0.055| -0.248| 0.175| -0.065| -0.255| -0.357| 0.725**|        |
| Yld   | -0.017| -0.271| 0.637**| 0.447*| -0.622**| -0.209| 0.642**| -0.006| 0.011| -0.297 |        |

Remarks: Hdg = heading date, i.e. days from sowing to 50% of flowering; Hzst = harvesting time (days after sowing); PH = plant height (cm); TN = tiller number; FG = filled grain/panicle; YG = un-filled grain/panicle; TG = total grain per panicle; SS = seed set, i.e. proportion of filled grain form the total gain per panicle; 1000GW = weight of 1000 rice grains; Fe = Fe content in brown rice; Zn = Zn content in brown rice; Yld = yield.

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