The stability of high Zinc rice strain in different agro climate conditions

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Abstract. The production of crops, especially rice, is agitated due to climate change. The change in climate conditions causes the change of growing and harvesting season of rice. Zinc is a micronutrient that is needed by the body for its growth. The symptoms of zinc deficiency, both small and large, can cause stunted growth, eczema, hair loss, delayed sexual maturity and mental development disorders. This study aimed to determine agronomic performance stability and rice genotypes’ yield with high-contain zinc and high yield in different agro-climate conditions. The test was carried out on two different agro-climates, namely, in Karang Village, Karang Pandan, Karanganyar (highlands) with a height of ± 830 meters above sea level (masl) and in Plumbon Village, Mojolahan, Sukoharjo (lowlands) with a height of ± 121 masl, from February to June 2017. A total of ten strains and two comparative varieties (Cibang and Inpari 5 Merawu) were tested using a Randomized Block Design repeated four times on a 4m x 5m plot following a spacing of 25cm x 25cm and using transplanting techniques. The test results showed that the strain of IR 97477-115-1-CRB-0-SKI-1-SKI-0-2, IR 99284-15-3-2, IR 97477-115-1-CRB-0-SKI-3-SKI-0-2, B13884E-MR-22-3-1 and B13884-MR-29-1-1 had a relatively high-contain Zinc of broken rice in the two test sites. However, the strain of IR 97477-115-1-CRB-0-SKI-1-SKI-0-2 and IR 97477-115-1-CRB-0-SKI-3-SKI-0-2 had high stability of yield and Zinc content in different agro-climate conditions. So, it is prospective to provide new high yielding varieties.

1. Introduction
A great need for national food availability continues to increase in line with population growth [1]. The total population of Indonesia in 2015 is 254.9 million, with poor people reaching 28.51 million. Based on BPS (Indonesia Research Institute) data, the total per capita rice consumption is 139 kg per year or 380 grams per day, so the total national rice demand reaches 28 million tons per year. Recent climate change has disrupted the production of food crops, especially rice. This is due to changing climatic conditions resulting in changes in planting time and rice harvest time. In addition, major health problems will arise and develop rapidly in society, impoverished people who live in deprived conditions due to insufficient food needs. Therefore, people experience malnutrition symptoms, iron (Fe) and zinc (Zn) or what is commonly referred to as micronutrient deficiency.

Zinc is a component of more than 300 enzymes needed to repair wounds, maintain fertility, synthesize protein and increase immunity, among many important human health functions [2]. A few or many zinc deficiency symptoms can result in stunted growth, eczema, hair loss, delayed sexual maturity and mental development disorders. Zinc deficiency is more common in developing countries, where
more than 60 percent of the population is Zn deficient [3]. The prevalence of zinc deficiency in East Asia is 7%, Southeast Asia 79%, Eastern Europe 10%, Latin America 33%, Middle East 46% and North Africa, Sub-Saharan Africa 50% and 5% in high-income countries. In 10 provinces in Indonesia, micronutrition studies found the average prevalence of zinc-deficient children was 36.1%, with the highest percentage in West Nusa Tenggara 46.6% and the lowest in West Sumatra 11.7% while for South Sulawesi province at 22.7%. Efforts to overcome micronutrient deficiencies need to be done.

Strategies to thwart the problem of micronutrient deficiency include continuous supplementation (fertilization0 and fortification. Therefore, a new strategy is needed to overcome micro-nutrients malnutrition, namely by bio-fortification. Breeding material is formed conventionally (hybridization and selection) or unconventionally (another culture and gene transformation). Rice, with high Fe and Zn content resulting from breeding activities, can be consumed directly by the middle to lower class communities as energy and Fe [4]. These results provide rice with high micro-nutrient content.

The multi-location test was conducted to determine if there was a strain interaction factor with the environment or not. Multi-location testing provides an estimated value of genetic interactions with the environment, which is indispensable in determining stable and adaptive lines to be released for farmers' cultivation. Both high and low line interaction values with the plant environment form a stable value in different environments. Stability testing is very important because the plant's environment strongly influences several quantitative characters such as crop production. In addition, stability testing can also be used to identify the yield ability in different growing environments. The lines that give small variations in response to a particular growing environment's various conditions are indicated to have good adaptability to the environmental conditions concerned [5]. One way to obtain improved varieties is to test some potential lines in various agro-ecology. It is hoped that this multi-location test will produce a line with high yield and high zinc content, which is stable at two different elevations. This study aimed to determine the agronomic performance and yield of rice lines with high zinc content in both locations and obtain stable lines in both research locations.

2. Methods
The research was carried out in two locations at different elevations, namely in Plumbon Village, Mojolaban District, Sukoharjo Regency with an altitude of 121 meters above sea level and in Karang Village, Karang Pandan District, Karanganyar Regency with an altitude of 831 meters above sea level in February to July 2017. A total of 10 lines and 2 comparison varieties (Ciherang and Inpari 5 Merawu) were tested using a randomized block design repeated four times on a plot measuring 4 m x 5 m with a spacing of 25 cm x 25 cm using transplanting techniques. The cultivation techniques were carried out under the guidelines for integrated plant management. Observations were made on the agronomic characteristics of the plants such as plant height, number of tillers, age of the flower, number of panicles, number of filled grains per panicle, number of empty grains per panicle, weight of filled grains per clump, seed set (fertility), weight of 1000 grains and yield. The data from the observations were inputted and then analyzed using a single-experiment analysis of variance, a combined analysis of variance [6] and analysis of stability parameters using the Finlay and Wilkinson methods [7]. The average difference between lines or genotypes was expressed by the significant difference test (LSD) at 5% level. If the combined test results on the source of genotype x environmental diversity were significantly different at 5% level, the genotypes' appearance was tested by genotype interaction with the environment.

3. Results and discussion
The results of the combined analysis of variants showed that there was an interaction between the tested genotype and the experimental location (Table 1). The interaction between genotype and location affects yield characters, Zn and Fe content in skin-cracked rice, number of vegetative tillers, 50% of flowering age, physiological maturity, number of panicles/clumps, filled grain/panicle, empty grain/panicle, weight of the filled grain/family, seed set and weight of 1000 items but did not affect other characters. This means that each planted line has a different ability to deal with environmental differences [8]. The size of the influence of the genotype x environment interaction greatly depends on the genetic makeup
of a genotype and the complexity of the environment that affects it. The tested genotypes affected yield characters, Zn and Fe content in cracked rice, height of generative plants, number of vegetative tillers, 50% flowering age, physiological maturity, number of panicles/clumps, filled grain/panicle, empty grain/panicle, seeds set and weight of 1000 items and did not affect other characters.

**Table 1.** Analysis of combined yield variants, zinc, iron content and agronomic characters of 10 high zinc content lines and 2 check varieties in two experimental locations

| No | Observation Variables | Genotypes | Location | Genotype x Location |
|----|------------------------|------------|----------|---------------------|
| 1  | Results                | 0.001**    | 0.001**  | 0.000**             |
| 2  | Zinc Content           | 0.000**    | 0.000**  | 0.000**             |
| 3  | Iron Content           | 0.000**    | 0.003**  | 0.004**             |
| 4  | Vegetative Plant Height| 0.075      | 0.066    | 0.350               |
| 5  | Generative Plant Height| 0.010**    | 0.000**  | 0.549               |
| 6  | Number of Vegetative Puppies | 0.000** | 0.488    | 0.026*              |
| 7  | Number of Generative Puppies | 0.535 | 0.019*   | 0.313               |
| 8  | Flowering Age 50%      | 0.000**    | 0.000**  | 0.000**             |
| 9  | Physiological Cooking Age | 0.000**   | 0.000**  | 0.000**             |
| 10 | Number of Malai/clumps | 0.009**    | 0.244    | 0.006**             |
| 11 | Grain Content/Malai    | 0.000**    | 0.000**  | 0.003**             |
| 12 | Hampa/malai Grain      | 0.000**    | 0.007**  | 0.000**             |
| 13 | Weight of Grain Contents /Clumps | 0.192 | 0.001**  | 0.009**             |
| 14 | Seed Set               | 0.000**    | 0.000**  | 0.000**             |
| 15 | Weight 1000 grains     | 0.000**    | 0.538    | 0.004**             |

Note: ** = very significantly different at the 1% error level; * = significantly different at the 5% error rate

The characters' differences were due to genetic differences in varieties and expected lines characterized by different vegetative stages and different plant responses to the growing site [9]. The location of the experiment affected the character of Zn and Fe content in cracked rice, height of generative plants, number of generative tillers, 50% of flowering age, physiological maturity, filled grain/panicle, empty grain/panicle and seed set and had no effect on other characters. This means that the environment in which plants grow affects the agronomic characters and the tested lines' Zn and Fe content. IR 95133 strains: 1-B-16-14-10-GBS, IR 97477-115-1-CRB-0-SKI-1-SKI-0-2, IR 97477-115-1-CRB-0-SKI-3-SKI-0-2 and IR 97646-108-1-CRB-0-SKI-1-SKI-2-2 showed relatively high yields at both trial sites.

The genotypes tested in Karanganyar were lower than those grown in Sukoharjo location because low temperature causes abnormal panicle exertion and increases panicle sterility, irregular maturation of panicles thereby decreasing the grain yield [10]. IR 97477-115-1-CRB-0-SKI-1-SKI-0-2, IR 99284-15-3-2, IR 97477-115-1-CRB-0-SKI-3-SKI-0-2, B13884E-MR-22-3-1 and B13884-MR-29-1-1 had a relatively high zinc content in cracked rice in the two experimental locations. IR 95133 strains: 1-B-16-14-10-GBS, IR 99270-34-2-1, IR 97477-115-1-CRB-0-SKI-1-SKI-0-2, IR 99284-15 -3-2, IR 99680-3-CRB-0-SKI-1-SKI-2-5, B13884E-MR-22-3-1 and B13884-MR-29-1-1 contained Fe in cracked rice was relatively high at both trial sites.
Table 2. Average yields of ten lines with high zinc content and two varieties of check

| No | Genotype                  | Karanganyar | Sukoharjo |
|----|---------------------------|-------------|-----------|
| 1  | IR 95133:1-B-16-14-10-GBS | 1.92 abcd   | 5.1 a     |
| 2  | IR 99270-34-2-1           | 1.52 bcd    | 4.19 bc   |
| 3  | IR 97477-115-1-CRB-0-SKI-1-SKI-0-2 | 2.44 a | 4.42 abc |
| 4  | IR 99284-15-3-2           | 2.01 abcd   | 2.6 d     |
| 5  | IR 97477-115-1-CRB-0-SKI-3-SKI-0-2 | 1.60 abcd | 4.85 ab   |
| 6  | IR 97646-108-1-CRB-0-SKI-1-SKI-1-2-2 | 1.70 abcd | 4.57 ab   |
| 7  | IR 99680-3-CRB-0-SKI-1-SKI-2-5 | 2.28 abc   | 4 bc     |
| 8  | B13884E-MR-22-3-1         | 1.47 cd     | 3.62 c    |
| 9  | B13884-MR-29-1-1          | 1.33 d      | 4.12 bc   |
| 10 | B13884E-MR-30-2           | 1.87 abcd   | 2.75 d    |
| 11 | Inpari 5 Merawu           | 2.01 abcd   | 4.02 bc   |
| 12 | Ciherang                  | 2.34 ab     | 4.07 bc   |

CV 20.74
LSD 5% 0.86

Note: The mean value followed by the same letter in one column shows no significant difference according to the Least Significant Difference (LSD) test at the 5% level.

Table 3. Average zinc and Fe content in ten lines with high zinc content and two check varieties

| No  | Genotype      | Zn   | Fe   |
|-----|---------------|------|------|
|     |               | Karanganyar | Sukoharjo | Karanganyar | Sukoharjo |
| 1   | IR 95133:1-B-16-14-10-GBS | 33.21ef | 25.05 abc | 15.51 abc | 13.12 ab |
| 2   | IR 99270-34-2-1 | 40.92 bc | 24.15 bc | 17.76 a | 13.25 ab |
| 3   | IR 97477-115-1-CRB-0-SKI-1-SKI-0-2 | 43.07 ab | 29.62 abc | 18.41 a | 13.52 ab |
| 4   | IR 99284-15-3-2 | 47.45 a | 27.6 ab | 17.24 ab | 13.45 ab |
| 5   | IR 97477-115-1-CRB-0-SKI-3-SKI-0-2 | 44.24 ab | 27.6 abc | 17.24 a | 12.7 b |
| 6   | IR 97646-108-1-CRB-0-SKI-1-SKI-1-2-2 | 36.9 cde | 26.8 abc | 13.72 c | 12.5 b |
| 7   | IR 99680-3-CRB-0-SKI-1-SKI-2-5 | 40.26 bcd | 27.15 abc | 15.74 ab | 12.87 ab |
| 8   | B13884E-MR-22-3-1 | 47.39 a | 25.75 abc | 18.35 a | 13.42 ab |
| 9   | B13884E-MR-29-1-1 | 44.22 ab | 30.95 a | 17.81 a | 15.85 a |
| 10  | B13884E-MR-30-2 | 34.6 def | 28.27 abc | 14.14 c | 13.27 ab |
| 11  | Inpari 5 Merawu | 29.71 f | 23.65 bc | 14.35 bc | 12.9 ab |
| 12  | Ciherang      | 30.07 f | 23.05 c | 13.74 c | 11.95 b |

CV 7.61 8.63
LSD 5% 3.55 1.79

Note: The mean value followed by the same letter in one column shows no significant difference according to the Least Significant Difference (LSD) test at the 5% level.
Table 4. Stability parameters of rice lines with high zinc content at two different elevations

| Genotype                   | Rata-rata (ton/ha) | CVi   | bi  |
|----------------------------|--------------------|-------|-----|
| IR 95133:1-B-16-14-10-GBS | 3.52               | 64.32 | 1.48|
| IR 99270-34-2-1           | 2.84               | 65.8  | 1.23|
| IR 97477-115-1-CRB-0-SKI-1-SKI-0-2 | 3.42 | 40.48 | 0.91|
| IR 99284-15-3-2           | 2.3                | 17.69 | 0.27|
| IR 97477-115-1-CRB-0-SKI-3-SKI-0-2 | 3.22 | 71.24 | 1.5 |
| IR 97646-108-1-CRB-0-SKI-1-SKI-2-2 | 3.15 | 64.91 | 1.34|
| IR 99680-3-CRB-0-SKI-1-SKI-2-5 | 3.13 | 38.44 | 0.79|
| B13884E-MR-22-3-1         | 2.53               | 61.16 | 1.01|
| B13884-MR-29-1-1          | 2.72               | 72.29 | 1.29|
| B13884E-MR-30-2           | 2.3                | 26.4  | 0.4 |
| Inpari 5 Merawu           | 3.01               | 47.08 | 0.93|
| Cihering                  | 3.21               | 38.11 | 0.8 |

Information: CVi = coefficient of genotype diversity (r = low, t = high); bi = genotype regression coefficient; *= significantly different from 1 at α = 0.05 where bi < 1; ** significantly different from 1 at α = 0.05 where bi > 1; tn = not significantly different from 1 at α = 0.05

Table 5. Stability parameters of zinc content of rice lines with high zinc content at two different elevations

| Genotype                   | Kandungan Zn rata-rata (ppm) | CVi   | bi  |
|----------------------------|-------------------------------|-------|-----|
| IR 95133:1-B-16-14-10-GBS | 29.11                         | 19.83 | 0.65|
| IR 99270-34-2-1           | 33.39                         | 31.9  | 1.2 |
| IR 97477-115-1-CRB-0-SKI-1-SKI-0-2 | 36.33 | 26.15 | 1.07|
| IR 99284-15-3-2           | 37.44                         | 37.75 | 1.59|
| IR 97477-115-1-CRB-0-SKI-3-SKI-0-2 | 35.91 | 32.76 | 1.32|
| IR 97646-108-1-CRB-0-SKI-1-SKI-2-2 | 31.84 | 22.43 | 0.8 |
| IR 99680-3-CRB-0-SKI-1-SKI-2-5 | 33.69 | 27.55 | 1.04|
| B13884E-MR-22-3-1         | 36.55                         | 41.88 | 1.72|
| B13884E-MR-29-1-1         | 37.58                         | 24.98 | 1.06|
| B13884E-MR-30-2           | 31.42                         | 14.26 | 0.5 |
| Inpari 5 Merawu           | 26.65                         | 16.12 | 0.48|
| Cihering                  | 26.56                         | 18.74 | 0.56|

Information: CVi = coefficient of genotype diversity (r = low, t = high); bi = genotype regression coefficient; *= significantly different from 1 at α = 0.05 where bi < 1; **= significantly different from 1 at α = 0.05 where bi > 1; tn = not significantly different from 1 at α = 0.05

The result stability test at two different elevations in the experimental location showed that IR 99284-15-3-2 line had a bi value <1 and was significantly different, so that this line can be categorized as well adapted and stable in the marginal environment. However, IR 95133: 1-B- line 16-14-10-GBS, IR 99270-34-2-1, IR 97477-115-1-CRB-0-SKI-1-SKI-0-2, IR 97477-115-1-CRB-0-SKI-3-SKI-0-2, IR 97646-108-1-CRB-0-SKI-1-SKI-2-2, IR 99680-3-CRB-0-SKI-1-SKI-2-5, B13884E-MR-22-3-1, B13884E-MR-29-1-1, B13884E-MR-30-2, Inpari 5 Merawu and Cihering had bi values that were not significantly different from 1 and were categorized as stable (Table 4).
Zn content stability testing conducted at two different elevations in the experimental location showed that B13884E-MR-22-3-1 line had a bi value > 1 and was significantly different, which means this line had a stable zinc content in the optimal environment. In contrast, the IR 95133: 1 line -B-16-14-10-GBS, IR 99270-34-2-1, IR 97477-115-1-CRB-0-SKI-1-SKI-0-2, IR 99284-15-3-2, IR 97477-115-1-CRB-0-SKI-3-SKI-0-2, IR 97646-108-1-CRB-0-SKI-1-SKI-2-2, IR 99680-3-CRB-0-SKI-1-SKI-2-5, B13884-MR-29-1-1, B13884E-MR-30-2, Inpari 5 Merawu and Ciherang had bi values which were not significantly different from 1 and were categorized as stable (Table 5).

Table 6. Summary of yield, zinc content and regression coefficient of rice lines with high zinc content

| Genotypes            | Results | KRA | SKH | average | Bi | Zn Content | KRA | SKH | average | Bi |
|-----------------------|---------|-----|-----|---------|----|------------|-----|-----|---------|----|
| IR 95133:1-B-16-14-10-GBS | 1.92 abcd | 5.1 a | | 3.52 | 1.48tn | 33.21ef | | 25.05 abc | 29.11 | 0.65tn |
| IR 99270-34-2-1       | 1.52 bcd | 4.19 bc | | 2.84 | 1.23tn | 40.92 bc | | 24.15 bc | 33.39 | 1.2tn |
| IR 97477-115-1-CRB-0-SKI-1-SKI-0-2 | 2.44 a | 4.42 abc | | 3.42 | 0.91tn | 43.07 ab | | 29.62 ab | 36.33 | 1.07tn |
| IR 99284-15-3-2       | 2.01 abcd | 2.6 d | | 2.3 | 0.27* | 47.45 a | | 27.47 abc | 37.44 | 1.59tn |
| IR 97477-115-1-CRB-0-SKI-3-SKI-0-2 | 1.60 abcd | 4.85 ab | | 3.22 | 1.5tn | 44.24 ab | | 27.6 abc | 35.91 | 1.32tn |
| IR 97646-108-1-CRB-0-SKI-1-SKI-2-2 | 1.70 abcd | 4.57 ab | | 3.15 | 1.34tn | 36.9 cde | | 26.8 abc | 31.84 | 0.8tn |
| IR 99680-3-CRB-0-SKI-1-SKI-2-5 | 2.28 abc | 4 bc | | 3.13 | 0.79tn | 40.26 bcd | | 27.15 abc | 33.69 | 1.04tn |
| B13884E-MR-22-3-1     | 1.47 ed | 3.62 c | | 2.53 | 1.01tn | 47.39 a | | 25.75 abc | 36.55 | 1.72* |
| B13884E-MR-29-1-1     | 1.33 d | 4.12 bc | | 2.72 | 1.29tn | 44.22 ab | | 30.95 a | 37.58 | 1.06tn |
| B13884E-MR-30-2       | 1.87 abcd | 2.75 d | | 2.3 | 0.4tn | 34.6 def | | 28.27 abc | 31.42 | 0.5tn |
| Inpari 5 Merawu       | 2.01 abcd | 4.02 bc | | 3.01 | 0.93tn | 29.71 f | | 23.65 bc | 26.65 | 0.48tn |
| Ciherang              | 2.34 ab | 4.07 bc | | 3.21 | 0.8tn | 30.07 f | | 23.05 c | 26.56 | 0.56tn |

Cv 20.74 7.61
LSD 5% 0.86 3.55

Note: The mean value followed by the same letter in one column shows no significant difference according to the Least Significant Difference (LSD) Test at 5% level, bi = Genotype regression coefficient; *= significantly different from 1 at α = 0.05 where bi < 1, **= significantly different from 1 at α = 0.05 where bi > 1; tn = not significantly different from 1 at α = 0.05

The test results showed that the IR 97477-115-1-CRB-0-SKI-1-SKI-0-2 and IR 97477-115-1-CRB-0-SKI-3-SKI-0-2 lines were prospective to be released into new high yielding varieties with high Zn content. These lines had high yield and high Zn content, as observed in the two experimental locations.

4. Conclusion
IR 97477-115-1-CRB-0-SKI-1-SKI-0-2 and IR 97477-115-1-CRB-0-SKI-3-SKI-0-2 are prospective to be released into new high yielding varieties. IR 97477-115-1-CRB-0-SKI-1-SKI-0-2 had an average yield of 4.42 tonnes/ha and average zinc content was 36.33 ppm at both test sites. IR strain 97477-115-1-CRB-0-SKI-3-SKI-0-2 had an average yield of 3.22 tonnes/ha and average zinc content in the two
experimental locations was 35.91 ppm. These lines had high yield and stable zinc content as perceived in the two experimental locations.

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