Evaluation of mungbean lines for resistance to Cercospora leaves spot and powdery mildew (Erysiphe polygoni) diseases in the green house

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ABSTRACT. Mungbean is an important food crop, it is a source of carbohydrates, protein and B vitamine. Therefore the production must be increased. In increasing the production of mungbean, there are several constrains including disease infection. Leaf spot and powdery mildew are the main diseases on mungbean. Planting the resistant varieties to leaf spot / powdery mildew is an effective, easy, and inexpensive way. The aim of research is to get some resistant mungbean lines against leaf spot and powdery mildew disease. The research was conducted in the greenhouse of the Indonesian Legumes and Tuber Crops Research Institute, from May to September 2019. Evaluation of the resistance of mungbean lines to leaf spot and powdery mildew was carried out consecutively. The material tested was 20 mungbean breeding lines from the breeding section, with one resistant variety and one susceptible variety as a check. Inoculation was done by spraying the suspension of Cercospora cruenta spores of leaf spot, or Erysiphe polygoni spores of powdery mildew, with a density of $10^8$ spores / ml in the afternoon. Observation of the intensity of leaf spot or powdery mildew disease is carried out three times, starting at one week after the first symptoms appeared. The results showed that out of 20 mungbean lines tested, there were no line that were resistant, 7 lines were moderately resistant and 13 lines were moderately susceptible to leaf spot disease. Out of 20 lines of mungbeans tested, 8 lines were resistant and 12 lines were moderately resistant to powdery mildew.

1. Introduction
Mungbean is a source of carbohydrate and protein, it was countain 61.0 %, and 23.8% respectively [1]. Compared to soybeans, mungbean is more profitable economically because it harvest is two months only (60 days), the price is higher, the export market is also available, therefore the production of mungbeans must continue to be increased. Increasing the production there are some constrains, including the diseases. Leaf spot and powdery mildew are the main diseases on mungbean. Resistant varieties are one component of cultivation technology that is not expensive, easy, and safe for environmental sustainability. The availibity of resistant varieties can reduce yield loss, so it can increase income.
Leaf spot was often occurred in the rainy season, but powdery mildew was often occurred in the dry season. Leaf spot disease has been spread in Asia such as the Philippines, Malaysia, Thailand, India, Bangladesh, and Pakistan. While this disease has been known in all centers of mungbean production in Indonesia. The yield loss due to leaf spot disease reach up to 40% [2] and 46-61% yield loss in the field [3-4].

Powdery mildew has spread throughout the centre of mungbean production such as India, Pakistan, Thailand, China, Myanmar, and Indonesia [5]. According to Semangun (2005), in Indonesia, powdery mildew is spread in Java, Bali, and Sumatra [6]. The spread of this disease can be through the wind, so it is also possible to spread quickly to other provinces. The yieldloss due to powdery mildew on mungbean in Thailand reach up to 26.21% [7]. The intensity of powdery mildew in the field reaches 44% [8], and yield loss can reach 80% in local varieties. Powdery mildew is widespread throughout the world both on crops such as various bean crops, and on weeds [9]. Symptoms of powdery mildew disease are preceded by white patches on the lower leaves. The white patches are like flour which is a mass of conidia and conidiophores of the fungus. White patches will extend to all leaves, even in susceptible varieties, pods and stems also turn white. The conidium will form a haustorium that develops in leaf cells, sucking up plant nutrient fluids, so that the metabolic process is disrupted. In addition, conidium and conidiophores on the upper surface of the leaf will inhibit photosynthesis and transpiration [10]. Severe infections cause the leaves to dry out and eventually fall out. Severe infections before the flowering phase cause small pods or no pods are formed at all. While in the Philippines 21% if all leaves are covered by powdery mildew when the plants flower [11]. According to Fondevilla and Rubiales (2012), in Spain the loss of peas due to powdery mildew ranged from 25-50%, reducing total biomass, number of pods / plants, number of seeds / pods, number of branches, and plant height [12].

The availability of resistant varieties to leaf spot and powdery mildew in Indonesia is still lacking. Therefore, the breeding of resistant varieties to these disease is needed. The research aims to examined of mungbean lines that are resistant, moderately resistant, moderately susceptible, and susceptible to leaf spot and powdery mildew respectively.

2. Methodology

The research was conducted in a greenhouse of the Indonesian Legumes and Tuber Crops Research Institute (ILETRI) during the dry season (June – Oct 2019). The materials used were 20 mungbean lines, and 3 varieties, Betet as resistant check of leaf spot disease, Vima-I as resistant check of powdery mildew, and Sampeyong as susceptible check of leaf spot and powdery mildew. Each mungbean line was planted in 5 polybags, each polybag contains of 4 plants, 3 replications. These study was arranged two sets, of leaves spot and powdery mildew disease, successively. The mungbean lines were inoculated with spores from Cercospora canescens, and Erysiphe polygoni, respectively. The density of spores was 10⁴. The spores are obtained from the leaves of mungbeans infected with leaf spot or powdery mildew disease. Spores are shed using a brush, so a spore suspension is obtained. Spore suspension is sprayed on the leaves in the evening when the plants were 3 weeks old.

Table 1. Scoring of leaf spot disease and powdery mildew, and the resistant categories [2]

| Score | Arrangement                                           | Categories |
|-------|-------------------------------------------------------|------------|
| 1     | 0 – very resistant                                    | VR         |
| 2     | 1 – 10% leaves covered by leaf spot/ powdery mildew   | R          |
| 3     | 11 – 30% leaves covered by leaf spot/ powdery mildew  | MR         |
| 4     | 31 – 60% leaves covered by leaf spot/ powdery mildew  | MS         |
| 5     | 61 – 80% leaves covered by leaf spot/ powdery mildew  | S          |
| 6     | >80% leaves covered by leaf spot/ powdery mildew      | VS         |
The intensity of leaf spot or powdery mildew disease was scored based on the surface area of the mungbean leaves covered by leaf spot or powdery mildew disease, and referred to Subrahmanyam et al (1995) as followed in appendix 1 [11]. The resistant of leaf spot disease and powdery mildew referred to the AVRDC (1990) as follows Table 1.

3. Result and discussion

3.1. Resistance mungbean lines to leaf spot disease.
The mungbean lines tested from breeders included resistant check for powdery mildew, it was Vima-1, but no resistant check for leaf spot. It was Sampeong variety which has many lovely characters except the resistance to leaf spot and powdery mildew.

The results of leaf spot disease tested was presented in Table 2. At the first observation, the intensity of leaves spot disease were range from 5.70% up to 13.50%, by change the time the disease intensity become heavier, there were 17% up to 41.70% at five weeks after planting. The resistance of mungbeans lines to leaf spot diseases varies from moderately resistant (MR) to moderately susceptible (MS). Out of the 20 mungbean lines tested, only six lines were moderately resistant (MR), the other lines showed moderately susceptible. It was very difficult to obtain the resistant lines for leaf spot because of only one gene controls leaf spot disease, especially recessive genes [13]. The type of monogenic resistance is based on singel gene. It is easy to work with, but it will be break when a new strain of pathogen is coming in.

According to Hartmant et al (2008), nearly 4000 mungbean lines in AVRDC in Taiwan, only 4% are resistant to leaf spot diseases and only 12% lines are resistant to powdery mildew [14]. The results of research at the ILETRI also had difficulty in finding lines that were resistant to leaf spot, the criteria successfully presented were only moderately resistant and moderately susceptible.

| No.  | Mungbean lines     | Leaf spot disease intensity at | Resistance categories |
|------|--------------------|-------------------------------|-----------------------|
|      |                    | Four weeks after planting      | Five weeks after planting |                     |
| 1    | MMC 672-3c-Gt-1/Sampeong-2 | 5.70              | 17.00                  | MR                   |
| 2    | MMC 672-3c-Gt-1/Sampeong-4  | 6.80              | 23.70                  | MR                   |
| 3    | Sampeong/MMC 679-3c-Gt-1-5 | 7.50              | 29.20                  | MR                   |
| 4    | Sampeong/MMC 679-3c-Gt-1-9 | 6.90              | 34.70                  | MS                   |
| 5    | Vima 1/Sampeong-10   | 13.50             | 41.70                  | MS                   |
| 6    | Vima 1/Sampeong//Vima 1-13 | 9.50              | 32.90                  | MS                   |
| 7    | Vima 1/Sampeong//Vima 1-14 | 11.30             | 34.70                  | MS                   |
| 8    | Vima 1/Sampeong//Vima 1-15 | 10.20             | 35.90                  | MS                   |
| 9    | MMC 679-3c-Gt-1/Sampeong-22 | 9.50             | 31.40                  | MS                   |
| 10   | Sampeong/MMC 679-3c-Gt-1-26 | 7.50             | 20.60                  | MR                   |
| 11   | MMC 672-3c-Gt-1/Vima 2-41 | 11.00             | 29.70                  | MR                   |
| 12   | MMC 672-3c-Gt-1/Vima 2-42 | 13.40             | 35.10                  | MS                   |
| 13   | MMC 672-3c-Gt-1/Vima 2-43 | 11.90             | 32.50                  | MS                   |
| 14   | Vima1/MMC 679-3c-Gt-1/Vima 1-46 | 6.90           | 26.30                  | MR                   |
| 15   | Vima1/MMC 679-3c-Gt-1/Vima 1-47 | 7.50           | 33.50                  | MS                   |
The results of leaf spot disease tested was presented in Table 3. At the first observation, the intensity of powdery mildew disease were range from 0% up to 6.70%, by change the time the disease intensity become heavier, there were 3.7% upto 73.30% at six weeks after planting. The resistance of mungbeans lines to powdery mildew diseases varies from moderate resistant (MR) to susceptible (S). Out of the 20 mungbean lines tested, 8 lines were resistant (R), the other lines showed moderately resistant (MR)

Mungbean resistant lines to powdery mildew are more easily to obtained than resistant to leaf spot diseases. This is caused by genes that control resistance to powdery mildew, only two dominant genes (Reddy KS et al. 1994). In an inheretance pattern that is control by twoo or more dominant genes, the chances of getting a resistant genotipe from mungbean are greater than that of recessive single gene pattern. Usually has a lower level of resistance than vertical resistance types.

Table 3. Powdery mildew disease intensity and resistance categories on mungbean lines. Greenhouse. Sept – Oct 2019.

| No | Mungbean lines | Five weeks after planting | Six weeks after planting | Resistance categories |
|----|----------------|--------------------------|--------------------------|-----------------------|
| 1  | MMC 672-3c-Gt-1/Sampeong-2 | 4.70 | 20.00 | MR |
| 2  | MMC 672-3c-Gt-1/Sampeong-4 | 6.70 | 20.00 | MR |
| 3  | Sampeong/MMC 679-3c-Gt-1-5 | 1.30 | 15.00 | MR |
| 4  | Sampeong/MMC 679-3c-Gt-1-9 | 3.30 | 20.00 | MR |
| 5  | Vima 1/Sampeong-10 | 2.70 | 30.00 | MR |
| 6  | Vima 1/Sampeong/Vima 1-13 | 2.30 | 25.00 | MR |
| 7  | Vima 1/Sampeong/Vima 1-14 | 6.00 | 12.50 | MR |
| 8  | Vima 1/Sampeong/Vima 1-15 | 0.00 | 7.30 | R |
| 9  | MMC 679-3c-Gt-1/Sampeong-22 | 4.00 | 12.50 | MR |
| 10 | Sampeong/MMC 679-3c-Gt-1-26 | 3.70 | 5.00 | R |
| 11 | MMC 672-3c-Gt-1/Vima 2-41 | 0.30 | 5.00 | R |
| 12 | MMC 672-3c-Gt-1/Vima 2-42 | 2.70 | 10.00 | R |
| 13 | MMC 672-3c-Gt-1/Vima 2-43 | 0.00 | 12.50 | MR |
| 14 | Vima1/MMC 679-3c-Gt-1/Vima1-46 | 2.30 | 12.50 | MR |
| 15 | Vima1/MMC 679-3c-Gt-1/Vima1-47 | 5.00 | 5.00 | R |
| 16 | MLG 1065/Vima 2-50 | 2.50 | 3.70 | R |
| 17 | MLG 1065/Vima 2-51 | 5.00 | 6.70 | R |
| 18 | MLG 1065/Vima 1-53 | 6.00 | 2.50 | R |
| 19 | MMC 679-3c-Gt-1/MLG 1065-55 | 1.70 | 12.50 | MR |
| 20 | Vima 1/MLG 1065-56 | 1.00 | 12.50 | MR |
3.3. Yield component
Mungbean yield component of the powdery mildew disease tested were observed in the harvested time, it was showed in Table 4. Sum of plants harvested were varies in each mungbean lines, it were also for sum of pods per plant, and grain weight per plant (gr). The lowest of grain yield was 4.67 gram, and the highest was 10.58 gram. Adaptation test of these genotipes showed that the grain yield were varies from 1.58 – 2.03 t/ha when the disease score of powdery mildew was lower, it was 1, as mention on appendix 2 [15].
Table 4. Mungbean yield component. Greenhouse. Dry season (June - Oct 2019)

| No | Mungbean lines                          | Sum of plants harvested | Sum of pods per plant | Grain weight per plant (gr) |
|----|----------------------------------------|-------------------------|-----------------------|----------------------------|
| 1  | MMC 672-3c-Gt-1/Sampeong-2             | 20.5                    | 6.8                   | 9.10                       |
| 2  | MMC 672-3c-Gt-1/Sampeong-4             | 15.6                    | 5.2                   | 6.93                       |
| 3  | Sampeong/MMC 679-3c-Gt-1-5              | 17.9                    | 6.0                   | 7.97                       |
| 4  | Sampeong/MMC 679-3c-Gt-1-9              | 19.5                    | 6.5                   | 8.68                       |
| 5  | Vima 1/Sampeong-10                     | 19.6                    | 6.5                   | 8.73                       |
| 6  | Vima 1/Sampeong//Vima 1-13              | 14.4                    | 4.8                   | 6.40                       |
| 7  | Vima 1/Sampeong//Vima 1-14              | 17.7                    | 5.9                   | 7.85                       |
| 8  | Vima 1/Sampeong//Vima 1-15              | 18.4                    | 6.1                   | 8.18                       |
| 9  | MMC 679-3c-Gt-1/Sampeong-22             | 10.5                    | 3.5                   | 4.67                       |
| 10 | Sampeong/MMC 679-3c-Gt-1-26             | 14.1                    | 4.7                   | 6.27                       |
| 11 | MMC 672-3c-Gt-1/Vima 2-41               | 21.1                    | 7.0                   | 9.39                       |
| 12 | MMC 672-3c-Gt-1/Vima 2-42               | 15.8                    | 5.3                   | 7.01                       |
| 13 | MMC 672-3c-Gt-1/Vima 2-43               | 13.0                    | 4.3                   | 5.76                       |
| 14 | Vimal1/MMC 679-3c-Gt-1/Vima1-46         | 23.8                    | 7.9                   | 10.58                      |
| 15 | Vimal1/MMC 679-3c-Gt-1/Vima1-47         | 11.9                    | 4.0                   | 5.30                       |
| 16 | MLG 1065/Vima 2-50                      | 18.0                    | 6.0                   | 8.00                       |
| 17 | MLG 1065/Vima 2-51                      | 16.7                    | 5.6                   | 7.42                       |
| 18 | MLG 1065/Vima 1-53                      | 18.5                    | 6.2                   | 8.21                       |
| 19 | MMC 679-3c-Gt-1/MLG 1065-55             | 15.5                    | 5.2                   | 6.90                       |
| 20 | Vima 1/MLG 1065-56                      | 20.4                    | 6.8                   | 9.07                       |
| 21 | Sampeong (susceptible check)            | 20.8                    | 6.9                   | 9.23                       |
| 22 | Vima 1 (resistant check to powdery mildew)| 19.4                    | 6.5                   | 8.62                       |
| 23 | Betet (resistant check to leaf spot)     | 21.7                    | 7.2                   | 9.63                       |

4. Conclusion
This study find out from 20 mungbean lines were tested, there was no resistant lines, six lines were moderately resistant, and 14 lines were moderately susceptible to leaf spot disease and from 20 mungbean lines tested, 8 lines were resistant and 12 lines were moderately resistant, to powdery mildew.

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