Increasing resistance of chrysanthemum to white rust disease: the role of mutant genotypes and enzymes activities

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Abstract. Chrysanthemum is an ornamental crop with high economic value and consistently received high demand attributed to its amazing beautifulness. However, white rust disease has drastically suppressed the growth and production of chrysanthemum. The purpose of this study was to obtain genetic and phenotypic diversity characters of peroxidase (pod), polyphenol oxidase (ppo), phenylalanine ammonia lyase (pal) and their resistant component of enzymes activity. The experiment was conducted using a randomized block design. The treatments consisted of 37 mutants and 11 genotype chrysanthemum parents with two replications. Observations were made for pod, ppo and pal enzyme activities. Results showed pod, ppo, and pal enzyme activities had narrow genetic and phenotypic variabilities. Further, chrysanthemum mutant obtained enzyme activities is better than the best control, namely 18.30.068, 10.10.010, 1.30.038, 20.35.056, 18.20.112, 18.25.079, 9.35.162, 18.20.089 and 16.35.086 genotypes. Pod, ppo and pal enzymes activity are more resistant to higher than the susceptible genotype chrysanthemum white rust disease. The other finding is as revealed by negative correlation between disease intensity character and pod, ppo and pal enzymes activity, while disease incidence and the rate disease development characters did not correlate with the third enzymes activity characters.

1. Introduction
Chrysanthemum is one of the ornamental plants attracted many people. Chrysanthemum white rust is an important disease in many countries[1], and the major disease in chrysanthemum[9]. These diseases cause yield losses of 30% [25] to 80% [10], even in the northeast United States cause yield losses of up to 100% [12].

The use of resistant varieties is the most reliable, economical in the long term and does not require specific tools and expertise in application, so efficient to suppress and control the white rust disease on chrysanthemum[3]. This step aside can reduce the cost of production for the application of pesticides, also can reduce the risk of negative effects of using chemicals on the environment[21].

Mutation breeding method is one approach that can be taken to obtain new genotypes with different phenotypic appearance with the parent. According to Miller[19], mutation techniques can be done to increase genetic variability and obtain new cultivars in a shorter time. Therefore, the need search genetic and phenotypic variability of the characters which can be sued as a more effective and efficient toll for the selection of chrysanthemum.
Metabolic defense is one of the mechanisms of plant against pathogen attack. Metabolic defense formed in the cells and tissues of plants likewise producing toxic substances to pathogens or create conditions that inhibit the development of pathogens [1]. Li and Steffens [18] reported that, over expression of polyphenol oxidase occurs in transgenic tomato with a 10-fold increase, this increase led to increased resistance to bacterial disease. In addition, the ppo in the plant will increase when the plant is injured or infected [26].

Fang [6] reported that the content of the peroxidase (pod), polyphenol oxidase (ppo) and phenylalanine ammonia lyase (pal) enzyme activities on resistant chrysanthemum genotype white rust disease is higher than the susceptible genotype. These inform that the activities of the pal, ppo and pod enzymes activities increased during the attack of certain diseases and this increase was in line with the increase in plant resistance to attack the disease. Thus, there is a closed link between the pal, ppo and pod enzyme activities with an increase in plant resistance to certain diseases. Thus, the pod, ppo and pal enzyme activities are defensive enzymes in plants resistance, especially of chrysanthemum.

2. Materials and Methods
The genotypes used were 37 chrysanthemum mutant genotypes MV_{4} generation and 11 parents varieties as a control. The numbers used genotypes are presented in Table 1. The genotypes were planted and inoculated with of P. horiana spores fungus. The observations are pod, ppo and pal enzyme activities. Observations to obtain information of genetic and phenotypic variability third enzyme activities and to obtain information correlation between resistance component with these enzymes activity characters.

Table 1. The Genotypes Used in Research

| No. | Genotypes   | No. | Genotypes   | No. | Genotypes   | No. | Genotypes   |
|-----|-------------|-----|-------------|-----|-------------|-----|-------------|
| 1   | 16.25.105   | 13  | 9.25.322    | 25  | 9.10.132    | 37  | 18.20.112   |
| 2   | 20.20.043   | 14  | 10.0        | 26  | 20.35.056   | 38  | 18.25.079   |
| 3   | 15.25.067   | 15  | 7.0         | 27  | 9.25.051    | 39  | 9.35.162    |
| 4   | 10.10.010   | 16  | 20.10.025   | 28  | 21.10.046   | 40  | 9.25.075    |
| 5   | 18.15.033   | 17  | 18.15.021   | 29  | 5.15.045    | 41  | 1.15.017    |
| 6   | 9.0         | 18  | 13.0        | 30  | 13.15.002   | 42  | 15.0        |
| 7   | 20.0        | 19  | 1.25.087    | 31  | 18.0        | 43  | 7.10.096    |
| 8   | 13.25.004   | 20  | 20.25.067   | 32  | 5.0         | 44  | 1.0         |
| 9   | 18.30.068   | 21  | 15.15.172   | 33  | 20.35.022   | 45  | 18.20.089   |
| 10  | 1.30.038    | 22  | 21.0        | 34  | 20.30.061   | 46  | 20.15.113   |
| 11  | 1.25.163    | 23  | 15.20.176   | 35  | 16.0        | 47  | 1.25.072    |
| 12  | 1.35.084    | 24  | 18.10.082   | 36  | 21.20.061   | 48  | 16.35.086   |

The experiment was designed using a randomized block design consisting of 48 treatments with two replications. Absorbance values are calculated by subtracting the absorbance values obtained from the spectrophotometer with absorbance values of the reference solution. The average value of absorbance (ΔOD = b) of the observations obtained using the regression equation (Y = a + bx). The unit enzyme activity (UEA) is calculated with the following formula:

\[
\text{UEA} = \frac{b \times e \times p \times t}{F_d}
\]

where:
- \(b\) = average absorbance
- \(e\) = enzyme preparations (ml)
- \(p\) = total protein content
- \(t\) = time of observation (minute)
- \(F_d\) = dilution factor
Data was analysed using ANOVA with a linear model Gaspersz[8]. Genetic variability is obtained from the expected means square (EMS). Genetic variance ($\sigma^2_g$) and phenotypic variance ($\sigma^2_f$), were calculated using the following formulas:

$$\sigma^2_g = \frac{\text{MS}_{\text{treatment}} - \text{MS}_{\text{error treatment}}}{r}$$  \hfill (2)

$$\sigma^2_f = \sigma^2_g + \text{MS}_{\text{error treatment}}$$  \hfill (3)

Broad and narrow variability a character is determined by the deviation standard value of the genetic variance. The deviation standard value is the square root of the genetic variance ($\sigma_g$). If the genetic variance ($\sigma^2_g$) value is greater than twice the deviation standard ($\sigma_g$), then the character is broad variability[11]. Differences between genotypes were using the F test at 5% level. The Least Significant Increase (LSI) test is performed as advanced test if there are significant differences among treatments[22]. LSI test is defined as follows:

$$\text{LSI} = t_\alpha \left( \frac{2 \text{MS}_{\text{error treatment}}}{r} \right)^{\frac{1}{2}}$$  \hfill (4)

where :
- $t_\alpha$ = one direction t test value at 48 degrees of freedom with a confidence level of 5%.
- MS = mean square
- $r$ = the number replication of treatment

Data process were done with Microsoft EXCEL. Observations were made on the total protein content, peroxidase, polyphenol oxidase and phenylalanine ammonia lyase enzymes activities characters.

3. Results and Discussion

3.1 Genetic and phenotypic variabilities enzymes activities characters

All populations were infected by inoculant of *P. horiana*. The results of variance analysis on peroxidase (pod), polyphenol oxidase (ppo) and phenylalanine ammonia lyase (pal) enzymes activities characters showed significant difference between treatments (Table 2) implying that chrysanthemum genotypes in term of enzyme activities and response to white rust disease are very diverse.

| No. | Characters                     | F      | CV  | $\sigma^2_g$ | $\sigma_g$ | $\sigma^2_f$ | $\sigma_f$ | $C_1$   | $C_2$   |
|-----|--------------------------------|--------|-----|-------------|------------|-------------|------------|---------|---------|
| 1.  | Peroxidase                     | 1422.051** | 3.086 | 2.147-06    | 2.931-03   | 2.150-06    | 2.933-03   | N       |         |
| 2.  | Polyphenol oxidase             | 199.273** | 6.639 | 2.058-07    | 9.072-04   | 2.078-07    | 9.118-04   | N       |         |
| 3.  | Phenylalanine ammonia lyase    | 34.858**  | 12.804 | 0.00041     | 0.04048    | 0.00043     | 0.042      | N       |         |

** Highly significant based on F test.

$F_{(0.05/0.01;48)} : 1.616 - 1.972$  

$\sigma_g$ = Deviation standard of the genetic variance  

$\sigma_f$ = Deviation standard of the phenotypic variance  

$C_1$ = Criteria genetic variability  

$C_2$ = Criteria phenotypic variability  

CV = Coefficient of variation
The large genetic variability criteria, when the genetic variability value is greater than 2 times genetic deviation standard ($\sigma_g$).
The large phenotypic variability criteria, when the phenotypic variability value is greater than 2 times phenotypic deviation standard ($\sigma_p$).

Genetic and phenotypic variabilities criteria of pod, ppo and pal enzyme activities are low. The narrow variability indicates that chrysanthemum genotypes had the same phenotypic performance relatively. Selection of the pod, ppo and pal enzymes activity characters has a small chance to obtain chrysanthemum genotypes resistant, so the selection of resistant genotype to white rust disease through the third characters is not easy to do. The genetic background of parent and irradiation treatment to chrysanthemum genotypes are assumed not to give a real effect to change that characters, to be the cause of narrow genetic variability.

3.2 Enzymes activity characters of 37 chrysanthemum mutant genotypes performances.

Peroxidase enzyme activity of chrysanthemum mutant genotypes tended to be higher than the parent (control), although the differences were relatively small. Least Significance Increase (LSI) test was performed to compare the best mean value of chrysanthemum mutant genotypes and control. LSI test results are presented in Table 3.

Relatively high peroxidase enzyme activities was shown by genotypes 18.30.068 (0.0077), 9.35.162 (0.0052) and genotypes 1.35.084 (0.0059). However, LSI test showed that genotypes mutants have a better performance than the best control (7.0) with the value of enzyme activity above 0.0022 UEA (units enzyme activity). These genotypes are 16.25.105, 10.10.010, 18.30.068, 1.30.038, 1.35.084, 9.25.322, 15.20.176, 18.10.082, 20.35.056, 18.20.112, 18.25.079, 9.35.162, 7.10.096, 18.20.089 and 16.35.086.

Genotype 18.30.068 has immune criteria status, whereas genotype 9.35.162 and 1.35.084 had susceptible criteria status. This suggests that the chrysanthemum mutant genotypes are more resistant and have a greater ability to increase peroxidase enzymes activities. This was in line with the results of Fang [6] reported that chrysanthemum resistant genotypes has higher peroxidase activity than the susceptible chrysanthemum to white rust disease. Similar results were also reported by Pudjihartati on peanut plant [23].

Buonario and Montalbini [4], argues that peroxidase enzyme plays a role in the process that occurs in the extracellular matrix, which is a process associated with the formation of the cell wall. Lignifikasi induction of cell walls in infected tissue fungus is one of the structural defense systems of plants [13]. Lignin content increased can inhibit penetration and invasion of pathogens physically block the spread of toxins and enzymes released by pathogens, as well as restricting the nutrients supply needed pathogens [27].

Peroxidase enzyme is plays a role of catalyzing oxidation reaction phenolic compound become a quinone compound to produce H$_2$O$_2$ which is toxic to the pathogen [5]. Furthermore Lebeda [16] reported that peroxidase enzyme plays a role in the phenolic compounds synthesis and intermolecular bonds form in the cell walls infected of pathogens. Then, peroxidase enzyme will form ROS (reactive oxygen species) and converts it into lignin which serves to resist pathogenic spore infections [6]. Thus, peroxidase enzyme activity has a function as anti fungus and one of the enzymes that play a role in plant defense against disease system [24].

The various activities polyphenol oxidase enzyme in the chrysannemum genotypes showed different genotypes response to treatment white rust disease spores inoculation and the various of genotypes resistance level to white rust disease. The polyphenol oxidase activities in the chrysanthemum mutant genotypes showed higher than the parents, and chrysanthemum mutant genotypes have white rust disease intensity lower than the parent. This condition in line with Li and Steffens [18] reports that, over expression of polyphenol oxidase in transgenic tomato with increased 10 times and increased expression of these enzymes lead to increased resistance to bacterial disease.
Table 3. Least Significance Increase (LSI) Enzyme Activities.

| No. | Genotypes | Peroxidase (UEA) | Polyphenol Oxidase (UEA) | Phenylalanin Ammonia Lyase (UEA) |
|-----|-----------|------------------|--------------------------|---------------------------------|
| 1.  | 16.25.105 | 0.0024 +         | 0.0008 +                 | 0.0427                          |
| 2.  | 20.20.043 | 0.0011           | 0.0005                   | 0.0273                          |
| 3.  | 15.25.067 | 0.0009           | 0.0004                   | 0.0266                          |
| 4.  | 10.10.010 | 0.0026 +         | 0.0009 +                 | 0.0515 +                        |
| 5.  | 18.15.033 | 0.0014           | 0.0005                   | 0.0327                          |
| 6.  | 13.25.004 | 0.0022           | 0.0007                   | 0.0405                          |
| 7.  | 18.30.068 | 0.0077 +         | 0.0026 +                 | 0.1178 +                        |
| 8.  | 1.30.038  | 0.0024 +         | 0.0009 +                 | 0.0499 +                        |
| 9.  | 1.25.163  | 0.0017           | 0.0006                   | 0.0382                          |
| 10. | 1.35.084  | 0.0059 +         | 0.0020 +                 | 0.1036                          |
| 11. | 9.25.322  | 0.0026 +         | 0.0009 +                 | 0.0536 +                        |
| 12. | 20.10.025 | 0.0008           | 0.0004                   | 0.0266                          |
| 13. | 18.15.021 | 0.0012           | 0.0005                   | 0.0306                          |
| 14. | 1.25.087  | 0.0013           | 0.0005                   | 0.0311                          |
| 15. | 20.25.067 | 0.0012           | 0.0005                   | 0.0310                          |
| 16. | 15.15.172 | 0.0007           | 0.0004                   | 0.0254                          |
| 17. | 15.20.176 | 0.0023 +         | 0.0008 +                 | 0.0414                          |
| 18. | 18.10.082 | 0.0009 +         | 0.0005                   | 0.0269                          |
| 19. | 9.10.132  | 0.0005           | 0.0003                   | 0.0193                          |
| 20. | 20.35.056 | 0.0034 +         | 0.0013 +                 | 0.0671 +                        |
| 21. | 9.25.051  | 0.0015           | 0.0006                   | 0.0329                          |
| 22. | 21.10.046 | 0.0006           | 0.0004                   | 0.0217                          |
| 23. | 5.15.045  | 0.0012           | 0.0005                   | 0.0276                          |
| 24. | 13.15.002 | 0.0008           | 0.0004                   | 0.0257                          |
| 25. | 20.35.022 | 0.0021           | 0.0006                   | 0.0401                          |
| 26. | 20.30.061 | 0.0016           | 0.0006                   | 0.0356                          |
| 27. | 21.20.061 | 0.0016           | 0.0006                   | 0.0357                          |
| 28. | 18.20.112 | 0.0027 +         | 0.0012 +                 | 0.0569 +                        |
| 29. | 18.25.079 | 0.0031 +         | 0.0012 +                 | 0.0636 +                        |
| 30. | 9.35.162  | 0.0052 +         | 0.0015 +                 | 0.0675 +                        |
| 31. | 9.25.075  | 0.0014           | 0.0005                   | 0.0312                          |
| 32. | 1.15.017  | 0.0012           | 0.0005                   | 0.0307                          |
| 33. | 7.10.096  | 0.0023 +         | 0.0007                   | 0.0408                          |
| 34. | 18.20.089 | 0.0026 +         | 0.0012 +                 | 0.0545 +                        |
| 35. | 20.15.113 | 0.0006           | 0.0004                   | 0.0215                          |
| 36. | 1.25.072  | 0.0016           | 0.0006                   | 0.0371                          |
| 37. | 16.35.086 | 0.0032 +         | 0.0012 +                 | 0.0643 +                        |
| 38. | 9.0       | 0.0007           | 0.0004                   | 0.0252                          |
| 39. | 20.0      | 0.0007           | 0.0004                   | 0.0230                          |
| 40. | 10.0      | 0.0016           | 0.0006                   | 0.0359                          |
| 41. | 7.0       | 0.0021           | 0.0006                   | 0.0390                          |
| 42. | 13.0      | 0.0003           | 0.0002                   | 0.0152                          |
| 43. | 21.0      | 0.0002           | 0.0002                   | 0.0140                          |
| 44. | 18.0      | 0.0007           | 0.0004                   | 0.0225                          |
| 45. | 5.0       | 0.0005           | 0.0003                   | 0.0214                          |
| 46. | 16.0      | 0.0017           | 0.0006                   | 0.0386                          |
| 47. | 15.0      | 0.0004           | 0.0003                   | 0.0191                          |
| 48. | 1.0       | 0.0003           | 0.0003                   | 0.0188                          |

|                  | LSI        | Comparison + LSI |
|------------------|-----------|-----------------|
| **(U)**          | 0.0001    | 0.0001          |
| **Phenylalanin Ammonia Lyase (UEA)** | 0.0100 | 0.0490 |

Values in the same column followed by the symbol + indicate higher than the best control (P > 0.05) based on Least Significance Increase (LSI) test.
LSI test of polyphenol oxidase enzyme activities character indicates that there are 11 mutant genotypes has a better performance than the best control (16.0) with a value of enzyme activity higher than 0.0007 UEA (units enzyme activity). The genotypes are 16.25.105, 10.10.010, 18.30.068, 1.30.038, 15.20.176, 20.35.056, 18.20.112, 18.25.079, 9.35.162, 18.20.089, and 16.35.086. Fang [6] reported that polyphenol oxidase enzyme activity increased. Catalyzing will increase quinones compounds formation. These compounds play a role in limiting the multiplication and spread of pathogens. Do [5] and Fang [6], report the peroxidase and polyphenol oxidase enzymes were quinones compounds catalyzing. This reinforces the opinion Krzywanski and Kozłowska [15] which found, increased polyphenol oxidase activity is closely related to increased phenolic acids and peroxidase activity. Thus, the pod and ppo activity was allegedly involved in plant defense reactions together.

Peroxiadase and polyphenol oxidase enzymes activities in plants can increase plant resistance to disease. Increased resistance occurs when crops suffer from injury and infection. This is due to the phenols oxidation in phytoalexin biosynthesis or other toxic compounds derived from pathogens [15]. Therefore, after pathogens infection in the plant tissue, then there will be an increase in enzyme activity and increased endurance.

The chrysanthemum genotypes has phenylalanine ammonia lyase activities were diverse. That difference shows that each genotype had different responses to the biotic stress. Biotic stress in question is P. horiana fungus spores inoculation treatment of conducted 14 to 20 days before phenylalanine ammonia lyase enzyme activity measurement.

LSI test of phenylalanine ammonia lyase enzyme activity character showed that, 10 mutants genotypes has phenylalanine ammonia lyase enzyme activity value higher than the best control (7.0) with activity above 0.0490 UEA. The genotypes are 10.10.010, 18.30.068, 1.30.038, 20.35.056, 18.20.112, 18.25.079, 9.35.162, 18.20.089, and 16.35.086.

The phenylalanine ammonia lyase enzyme activity obtained from 18.30.68 genotype is the highest activity (0.1178). Be reviewed from the disease intensity 18.30.68 genotype has has 0% disease intensity value, meaning that this genotype is not white rust disease against. This shows that perhaps phenylalanine ammonia lyase is one of the factors that play a role in the plant's defense system against white rust disease.

Fang [6] reported that phenylalanine ammonia lyase enzyme is a key enzyme in lignin synthesis. Pal activity will increase the lignin produced plants. thus preventing nutrients absorption pathogen mycelia in leaf tissue. In addition, this enzyme acts as a catalyst in the early stages phenylpropanoid pathway that produces trans cinnamic acid derivatives and phenylalanine form [7]. Phenylpropanoid compounds are precursors in the formation penolik compounds such as flavonoids, isoflavonoids, anthocyanin. hormones phytoalexin and lignin [17].

Based on above description, Peroxidase, polyphenol oxidase, and phenylalanine ammonia lyase enzyme activities of in the more resistant genotypes will be higher than the susceptible genotypes. This was in line with Fang[6] reported that resistant chrysanthemum genotypes have peroxidase activity higher than the white rust disease susceptible chrysanthemum.

3.3 Correlation between resistance component and enzyme activities characters

Peroxidase, polyphenol oxidase and phenylalanine ammonia lyase enzyme activities highly significant negative correlation with white rust disease intensity (Table 4). This means that the higher the third enzymes activities, then disease intensity in mutant genotypes the lower and mutants genotypes increasingly resistant.

The results of this study are consistent with the Harrison [14] reported that peroxidase enzyme activity correlates with disease severity and red pepper virus concentration. Thus, the character peroxidase, polyphenol oxidase and phenylalanine ammonia lyase enzyme activity of chrysanthemum mutant genotypes can be used as the basis for selection to obtain varieties chrysanthemum resistant white rust disease. In addition, the selection white rust disease resistance character can be done at the beginning of the growth phase, so that the selection can be done faster and more efficiently.
The rate of disease development did not correlate with the third enzymes activities. This was in line with the opinion of Vanitha [26] that polyphenol oxidase activity in the plant will increase when the plant is injured or infected by the pathogen. Furthermore Ngadze [20] reported that peroxidase, polyphenols oxidase and phenylalanine ammonia lyase activities increased significantly in potato tubers were wounded and inoculated. Then, on a different potato varieties, high polyphenol oxidase activity indicated in the potato varieties that have high activity of phenylalanine ammonia lyase [20]. Then, phenylalanine ammonia lyase activity on all varieties of potatoes has increased significantly eight hours after cutting the tubers and after inoculation to P. carotovorum subsp. brasiensis. Phenylalanine ammonia lyase enzyme activity was higher in potato clones that are resistant to Phytophthora infection palmivora.

The rate of disease development disease characters did not correlate with peroxidase, polyphenol oxidase and phenylalanine ammonia lyase enzymes activities. This is due to chrysanthemum mutant genotypes showed enzyme activity increased after the disease incidence, hence infection rate can be reduced or even stopped. This situation causes no relationship between the rate of disease development with the third enzymes activities.

Based on description aboveshowed genetic and phenotypic variabilities are narrow for third enzymes activities. Then, enzyme activities of chrysanthemum mutant genotype is better than the best control. Namely 18.30.068, 10.10.010, 1.30.038, 20.35.056, 18.20.112, 18.25.079, 9.35.162, 18.20.089 and 16.35.086. Peroxidase, polyphenol oxidase and phenylalanine ammonia lyase enzymes activities in the more resistant genotypes was higher than the susceptible genotype. Very real negative correlation between disease intensity with peroxidase enzyme, polyphenol oxidase and phenylalanine ammonia lyase enzymes activities, while disease incidence and rate of disease development characters did not correlate with the third enzymes activities.

4. Conclusions

Peroxidase, polyphenol oxidase and phenylalanine ammonia lyase enzyme activities in chrysanthemum genotypes mutant had narrow genetic and phenotypic variabilities. Chrysanthemum genotypes mutant obtained enzyme activity is better than the best control. Namely 18.30.068, 10.10.010, 1.30.038, 20.35.056, 18.20.112, 18.25.079, 9.35.162, 18.20.089 and 16.35.086 genotypes. Peroxidase, polyphenol oxidase and phenylalanine ammonia lyase enzymes activities in chrysanthemum genotypes mutant resistant to white rust disease higher than the susceptible genotype chrysanthemum. Very real negative correlation between disease intensity with peroxidase, polyphenol oxidase and phenylalanine ammonia lyase enzymes activities, while disease incidence and the rate disease development did not correlate with the third enzymes activities.

| No. | Characters                          | Disease intensity | Disease incidence | The rate of disease development |
|-----|------------------------------------|-------------------|-------------------|---------------------------------|
| 1.  | Disease intensity                  | 1                 |                   |                                 |
| 2.  | Disease incidence                  | -0.1651ns         | 1                 |                                 |
| 3.  | The rate of disease development    | 0.0805ns          | 0.6886**          | 1                               |
| 4.  | Peroxidase                         | -0.9374**         | 0.1212ns          | -0.1622ns                       |
| 5.  | Polyphenol Oxidase                 | -0.9290**         | 0.1428ns          | 0.1296ns                        |
| 6.  | Phenylalanine Ammonia Lyase        | -0.9287**         | 0.1742ns          | 0.1125ns                        |

** highly significant
ns nonsignificant

Table 4. Correlation between resistance component with enzyme activities characters.

Disease incidence characters are not correlated with peroxidase, polyphenol oxidase and phenylalanine ammonia lyase enzymes activities characters. This happens because in the event of disease incidence. The third enzymes activities are still low, then shortly after the attacks of white rust disease the third enzymes activities will increase. This was in line with the opinion of Vanitha [26] that polyphenol oxidase activity in the plant will increase when the plant is injured or infected by the pathogen. Furthermore Ngadze [20] reported that peroxidase, polyphenols oxidase and phenylalanine ammonia lyase activities increased significantly in potato tubers were wounded and inoculated. Then, on a different potato varieties, high polyphenol oxidase activity indicated in the potato varieties that have high activity of phenylalanine ammonia lyase [20]. Then, phenylalanine ammonia lyase activity on all varieties of potatoes has increased significantly eight hours after cutting the tubers and after inoculation to P. carotovorum subsp. brasiensis. Phenylalanine ammonia lyase enzyme activity was higher in potato clones that are resistant to Phytophthora infection palmivora.

The rate of disease development disease characters did not correlate with peroxidase, polyphenol oxidase and phenylalanine ammonia lyase enzymes activities. This is due to chrysanthemum mutant genotypes showed enzyme activity increased after the disease incidence, hence infection rate can be reduced or even stopped. This situation causes no relationship between the rate of disease development with the third enzymes activities.
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