Growth and development of pigeon pea (*Cajanus cajan*) on the differences of Fitosan concentration

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**Abstract.** Pigeon pea is *Leguminosae* plant that survives in drought conditions and able to grow in various soil types. Pigeon pea is potentially used for soy substitution, but has not been intensively cultivated in Indonesia. Fitosan is result of chitosan irradiation. This research was conducted to determine the effect of soaking pigeon pea seeds in various Fitosan concentrations. The research was designed as Randomized Complete Block Design factorial with two factors. The first factor was Fitosan concentration, namely control (without Fitosan), 2 ml l⁻¹, 4 ml l⁻¹, and 6 ml l⁻¹. The second factor was color of pigeon pea namely black, white, and brown. The results showed that best Fitosan concentration was 2 ml l⁻¹ which accelerate shoots appear and increase plant height up to 4 weeks after planting. The fastest flowered was in black pigeon pea that soaked with Fitosan 2 ml l⁻¹ and yields the most pods, white pigeon pea flowers appear fastest in soaking with 6 ml l⁻¹, while the brown pigeon pea has not flowered and produces pods until the end of the observation. There is no interaction between Fitosan concentration and the differences of color pigeon pea seeds on the growth and development plants.

1. **Introduction**

Pigeon pea (*Cajanus cajan*) has long been cultivated in Indonesia and has potential to be developed, even in barren areas with thin soil layers such as in Gunung Kidul and Jeneponto. Pigeon pea has production nearly of 2.5-3.3 tons ha⁻¹ of wet beans [1], therefore Pigeon pea can be used to improve food security, through food diversification. Pigeon pea can be used as a food crop, i.e. dry beans, flour, green beans, and as a cover crop [2], while other study reporting that pigeon pea beans can be used for soy substitution [3]. Pigeon pea beans contain 20-22% protein, 1.2% fat, 65% carbohydrate, and 3.8% ash [4], and the following minerals such as phosphorus 28.2%, potassium 17.2%, zinc 14.7%, copper 20.9%, iron 14.7%, calcium more than 19.2%, and manganese more than 10.8% [5].

Fitosan is produced by BATAN (Badan Tenaga Nuklir Nasional) as the result of chitosan irradiation. Fitosan is a low molecular weight chitosan [6] and can be obtained by irradiation using gamma rays at doses of 75 kGy. Chitosan and its derivatives can be used as antibiotic microorganisms [7], suggested that oligo chitosan exhibits better antifungal activity than chitosan in large units [8]. While Fitosan can accelerate growth, improve yield and protect plants from viruses, fungi and bacteria, i.e. as vaccines and antibiotics [9]. The difference of chitosan irradiation with chitosan without irradiation is chitosan oligo chitosan chemical is simpler than chitosan, so it is more easily absorbed and utilized plant. Oligo chitosan contains several hormones, namely IAA, kinetin, zeanin, GA3, GA5, and GA7 [10]. Pigeon pea plant growth and development need to be considered, to
increase the production. The purpose of this research was to know the effect of soaking of Fitosan on black, white, and brown seeds so that the best Fitosan concentration can increase the growth and development of pigeon pea, to know growth and development response of pigeon pea, and the interaction between pigeon pea seed color in some concentration of Fitosan.

2. Methods
The research was conducted at Greenhouse Faculty of Agriculture, Universitas Sebelas Maret Surakarta in April 2017 until August 2017. The research design was research used Randomized Complete Block Design factorial consisting of two factors. The first factor is concentration of soaking Fitosan, i.e. control (without Fitosan), concentration 2 ml l\(^{-1}\), 4 ml l\(^{-1}\), and 6 ml l\(^{-1}\). The second factor is the color of pigeon pea seeds, i.e. black, white, and brown. Based on these two factors, then obtained 12 combinations treatment and repeated as much 3 times, with each sample there are 3 sub samples, so obtained 108 plants. 108 plants. The observation variable consists of the shoots appear, plant height, number of leaves, leaf area, leaf color, number of branches, stem diameter, flower initiation, and number of pods black, white, and brown pigeon pea. Data were analyzed using F test followed with DMRT (Duncan's Multiple Range Test) at 5%.

3. Results and discussions
3.1. Shoots appear
Fitosan soaking aims to accelerate the emergence of pigeon pea shoots. Different colors of pigeon pea seeds do not affect when shoots appear, but the difference in Fitosan concentration affects the time of buds, and there is no interaction between the two (Table 1).

Table 1. Effect of Fitosan concentration and color of seed at shoot appear of pigeon pea

| Pigeon pea | Control | Fitosan (ml l\(^{-1}\)) | Average |
|-----------|---------|-------------------------|---------|
|           |         | 2          | 4        | 6        |         |
| Black     | 3.22    | 2.67       | 3.33     | 3.33     | 3.14\(^{a}\) |
| White     | 2.89    | 2.33       | 3.11     | 4.00     | 3.08\(^{a}\) |
| Brown     | 3.11    | 2.67       | 3.22     | 3.22     | 3.06\(^{a}\) |
| Average   | 3.07\(^{b}\) | 2.56\(^{a}\) | 3.22\(^{bc}\) | 3.52\(^{c}\) | - |

Notes : Control : without Fitosan and negative sign (-) : no interaction. The average values followed by the same letter on the Fitosan row and the type pigeon pea column show no significant difference according to the DMRT of 5% level.

Shoots are part of newly grown plants and appear to the soil after the germination process. Based on Table 1, it can be seen that the average of black, white and brown pigeon pea shoots appeared is not significantly different, i.e. at 3 HST. Black, white, and brown pigeon pea seeds have almost the same germination process, because the germination is determined by the food reserves in each seed and water. Soaking the seeds can accelerate germination and the appearance of shoots. Water is important in activating embryonic cells, softening the seed shell, as an oxygen entry facility, leading to the expansion of cotyledons, and enzyme activation [11].

Soaking seeds with 2 ml l\(^{-1}\)Fitosan concentration showed the fastest shoots appeared compared to control treatment, 4 ml l\(^{-1}\), and 6 ml l\(^{-1}\). Fitosan contains plant growth regulator, that is Gibberellin (GA) which is Giberelin, IAA from auxin, and Zeatin from cytokinin. GA, IAA, and Zeatin can regulate the growth of plants and affect directly on when shoots appear. Giberellin regulate some plant development processes that start from seed germination [12]. GA and auxins support each other in root formation, while GA and cytokinin reciprocally in shoots, apical meristem activity requires high cytokinin.

Fitosan can affect the balance of internal hormones in the seed. External giberellin needs to be given to accelerate germination and increase seed vigority [13]. Soaking the seeds with higher concentration of Fitosan, i.e. 4 and 6 ml l\(^{-1}\) can decrease the shoots appeared speed, although all the
seeds can still grow. This is because the cytokinin are too high to suppress gibberellin in germination, in addition to excessive auxin also inhibits plant growth. Auxin with excessive concentrations given to plants can reduce inhibiting the development of buds even to reduce the percentage of plant life [14].

3.2. Plant height
Soaking pigeon pea seeds with Fitosan aims to increase plant growth, one of them through the indicator of plant height. The high growth of the pigeon pea plant was shown in Figure 1.

![Figure 1. Chart of height pigeon pea plants](image)

Figure 1 showed that black, white, and brown pigeon pea have similar plant height increase, while data analysis shows no interaction between seed color difference at some of Fitosan concentrations used. Plant height at the end of observation was between 135 cm to 164 cm. Vange and Moses [15] suggested that pigeon pea plant height varies between 135 cm to 466 cm. Soaking the seeds using Fitosan with different concentrations affect the plant height at 1 to 4 weeks after planting, with the highest plant height in soaking with 2 ml l⁻¹. Fitosan contains GA, IAA, and Zeatin so it can increase the height of the pigeon pea plants. Gibberellin can affect the plant height, because gibberellin supports cell extension. IAA can increase plant height, because its role can increase the elongation of the cell, especially in the vertical direction [16], but if the concentration of IAA is not optimal, it can inhibit the growth of plants, while cytokinin can increase division cells in meristematic tissues and stimulate shoot growth [17].

Plant height at 1 to 4 weeks after planting on soaking of the seeds using 6 ml l⁻¹ concentration showed a similar height to the controls, suggesting excessive concentrations of Fitosan may decrease plant height. Plant height at 4 to 8 weeks after planting had a large increase (Fig. 1), but based on the analysis of variance, the difference in concentration of Fitosan did not affect plant height at 5 to 9 weeks after planting. Increased plant height occurs at the beginning of growth can be caused by giving Fitosan only done at the beginning of planting. The treatment of hormone in plants will be beneficial to plants when applied to the appropriate concentration and time or plant growth phase [18].

3.3. Numbers of leaves
Leaves are important organ of plants, especially its role in photosynthesis. The numbers of leaves calculated every week. The growth chart of the numbers of pigeon pea leaves was shown in Figure 2.
The numbers of black, white, and brown pigeon pea leaves were not significantly different, it was indicating a similar growth response of black, white, and brown pigeon pea. Fitosan can support the increase in the numbers of leaves because it contains zeatin, IAA, and GA, but the numbers of leaves shows the same result, although with the soaking of Fitosan. Soaking the pigeon pea seeds with Fitosan does not affect the numbers of leaves, can be caused by the endogenous hormone that can be self-produced in plants. Plants need plant growth regulator from the outside to stimulate growth, but the level of plant growth regulator concentration needed every plant organ will be different, when the endogenous hormone is sufficient, the hormone exogenously has no significant effect [19].

3.4. Leaf area

Leaf area was observed because of its association with photosynthesis and transpiration. The wider a leaf, it will enlarge photosynthesis and transpiration. The calculation of leaf area using gravimetric method. Fitosan soaking in black, white, and brown seeds with control treatment and concentration of 2 ml l⁻¹, 4 ml l⁻¹, and 6 ml l⁻¹ and pigeon pea seeds color difference is not significant to the large leaf area, while there is no interaction of both factor (Table 2).

Table 2. Effect of Fitosan concentration and color of seed at leaf area of pigeon pea

| Pigeon pea | Fitosan (ml l⁻¹) | Average |
|------------|------------------|---------|
|            | Control 2 4 6    |         |
| Black      | 49.31 51.06 49.31 50.10 | 49.95⁻  |
| White      | 49.42 49.05 48.62 50.95 | 49.51⁻  |
| Brown      | 49.53 48.36 49.36 49.26 | 49.13⁻  |
| Average    | 49.42⁻ 49.49⁻ 49.10⁻ 50.11⁻ | -       |

Notes: Control : without Fitosan and negative sign (-) : no interaction. The average values followed by the same letter on the Fitosan row and the type pigeon pea column show no significant difference according to the DMRT of 5% level.

Soaking pigeon pea seeds in Fitosan with control treatment and concentration 2 ml l⁻¹, 4 ml l⁻¹, or 6 ml l⁻¹ and color difference pigeon pea not significant to leaf area, while there is also no interaction of both factors. Giving of Fitosan for pigeon pea seeds aims to increase the leaf area, while the use of different colored pigeon pea seeds aims to compare the leaf area of black, white, and brown pigeon pea. Leaf area of black, white, and brown pigeon pea almost the same that ranges from 49-50 cm².
Giving Fitosan by soaking the pigeon pea seeds with concentration of 2 ml 1⁻¹, 4 ml 1⁻¹, 6 ml 1⁻¹, and control gave no significant difference. This can be due to the measured leaf samples from each plant is the 4th, 5th, and 6th from the branches end, so that it is at almost the same leaf growth rate. The largest leaf area is a leaf in the middle, while the leaf is located on the below and the tip is usually smaller [20]. Another factor is that environmental factors are almost the same, especially related to the intensity of sunlight, then the leaf size will be the same. The shaded plants have leaves more widely than those exposed to the sun directly [21].

3.5. Leaf color

Leaf color observation was done to find out the pigment of pigeon pea leaf. Pigeon pea leaf color mode of each treatment was shown in Table 3.

Table 3. Mode of leaf color from pigeon pea with different color seeds and Fitosan concentration

| Pigeon pea | Control | 2 | 4 | 6 |
|------------|---------|---|---|---|
| Black      | *C 75 Y 80 | C 80 Y 90 | C 75 Y 70 | *C 75 Y 80 |
| White      | M 60 K 30  | M 60 K 30  | M 60 K 30  | M 60 K 30  |
| Brown      | *C 75 Y 80 | M 60 K 30  | M 60 K 30  | *C 75 Y 80 |

Notes: Control : without Fitosan, C : cyan, Y : yellow, M : magenta, and K : black. M and K in %.
Asterisk (*) : same color.

The leaf color observations used the Process Color Manual Book from Michael and Pat Rogodino. This book showed the appropriate combination of color compositions. Based on Table 3, it was known that pigeon pea leaves had different colors that was affected by the color pigment, but the difference was not too large. The color can be used as the basis of identification pigments, blue green from chlorophyll a, yellow green from chlorophyll b, orange from carotenoid, gray from feofitin a [22]. Table 3 showed that most pigeon pea leaf color is in combination C (cyan) 75, Y (yellow) 80, M (magenta) 60%, and K (black) 30%, it was dark green and slightly tanned.

3.6. Numbers of branches

Counted the numbers of branches was done on the primary branch. The growth chart of the number of pigeon pea branches was shown in Figure 3.

![Figure 3. Chart of numbers of branches pigeon pea plants](chart.png)
Pigeon pea pods form at the end of the branches and main stems, so that more branches were expected to increase the numbers of pigeon pea pods. The increment of pigeon pea branches varies every week. The numbers of branches in each plant is different, however, soaking the seeds with some concentration of Fitosan and difference color seeds is not significant to the numbers of branches. The interaction between some Fitosan concentration and skin color difference was only available at week 7, that was the most branches at pigeon pea with black seed color with control treatment (without Fitosan), but the interaction did not occur overall. Branch formation is influenced by cytokinin hormone, because it functions in activation of cell division and stimulates shoot formation [23]. Fitosan was not affect the numbers of branches of the plant can be caused due to endogenous hormone for the formation of branches in sufficient condition, so the giving of Fitosan do not give effect, that when the endogenous hormone is sufficient, then the exogenous hormone does not affect [19].

3.7. Stem diameters
Measurement of stem diameter was done using the Vernier caliper, so that the result obtained by cm unit. Stem diameter of black, white, and brown pigeon pea were 1.1 cm (Table 4).

Table 4. Effect of Fitosan concentration and color of seed at stem diameter of pigeon pea

| Pigeon pea | Control | 2 | 4 | 6 | Average |
|------------|---------|---|---|---|---------|
| Black      | 1.07    | 1.16 | 1.15 | 1.13 | 1.13*   |
| White      | 1.12    | 1.11 | 1.05 | 1.14 | 1.11*   |
| Brown      | 1.08    | 1.11 | 1.09 | 1.16 | 1.11*   |
| Average    | 1.09*   | 1.13* | 1.10* | 1.14* | -       |

Notes: Control: without Fitosan and negative sign (-) : no interaction. The average values followed by the same letter on the Fitosan row and the type pigeon pea column show no significant difference according to the DMRT of 5% level.

Table 4 showed the different concentrations of Fitosan used to soak the pigeon pea seeds not affecting the large diameter of the stem, while the difference in the pigeon pea seeds color also has no effect and no interaction of the two. It showed that black, white, and brown pigeon pea had same large diameter of stems, while the given Fitosan does not increase the diameter of the stem. There are some things to be considered in give plant growth regulator, it was plant growth regulator is given in concentration and the right way [24].

3.8. Flower initiation
Observations when flowers appear was done when the initiation of flower. Flower initiation was the earliest stadia of the flower development process, beginning from the appearance of the bud on the flower stalk to the beginning of a small bud phase marked by the emergence of a cluster structure compound flower [25]. The average flower initiation of each treatment was shown in Table 5.

Table 5. Effect of Fitosan concentration and color of seed at flower initiation of pigeon pea

| Pigeon pea | Control | 2 | 4 | 6 | Average |
|------------|---------|---|---|---|---------|
| Black      | 71.78   | 67.56 | 70.67 | 74.33 | 71.09   |
| White      | 77.44   | 75.44 | 76.00 | 74.78 | 75.92   |
| Brown      | -       | -    | -    | -    | -       |

Notes: Control: without Fitosan, unit: days after planting, and negative sign (-) : no flowers yet.

Differences color of pigeon pea seeds that indicate different types of pigeon pea can cause different characteristics in flowering. The black pigeon pea flowered at 71 days after planting, white pigeon pea at 76 days after planting, while the brown pigeon pea had not flowered until the end of the observation. White and black pigeon pea were more tolerant than reddish-brown types [26].
The black pigeon pea flower appeared fastest in plants whose seeds was soaked by 2 ml l$^{-1}$ Fitosan concentration, while the fastest white pigeon pea flower appeared on the plant whose seeds was soaked by 6 ml l$^{-1}$ Fitosan concentration. Fitosan has not been able to accelerate the appearance of flowers at brown pigeon pea. Fitosan given before planting is expected to indirectly affect the appearance of interest. Mullen et al [27] states that pigeon pea flowering at the age of 60 to 80 days, but flowering can be delayed because of the period of extreme or moisture stress.

3.9. Numbers of pods

The pod observation was done by counted the numbers of pods formed at 12 weeks after planting. The black, white, and brown pigeon pea pods at 12 weeks after planting showed different number (Table 6).

| Pigeon pea | Fitosan (ml l$^{-1}$) | Average |
|------------|----------------------|---------|
|            | Control | 2 | 4 | 6 |         |
| Black      | 4.81    | 5.27 | 4.74 | 3.74 | 4.64$^a$ |
| White      | 3.52    | 3.87 | 3.75 | 3.81 | 3.74$^b$ |
| Brown      | 0.71    | 0.71 | 0.71 | 0.71 | 0.71$^c$ |
| Average    | 3.02$^a$ | 3.28$^a$ | 3.06$^a$ | 2.75$^b$ |         |

Notes : Control : without Fitosan and negative sign (-) : no interaction. The analyzed data has been transformed by $\sqrt{x + 0.5}$. The average values followed by the same letter on the Fitosan row and the type pigeon pea column show no significat difference according to the DMRT of 5%.

Soaking the pigeon pea seeds with Fitosan at concentrations of 2 ml l$^{-1}$, 4 ml l$^{-1}$, and 6 ml l$^{-1}$, or control (without Fitosan) did not affect the number of pods, while the pigeon pea seed color was significant to the numbers of pods, and there was no interaction between the two factors. Fitosan given to pigeon pea seeds before planting is expected to affect the physiological process of the plant, thus indirectly increasing the numbers of plant pods, but Fitosan has not been able to affect the numbers of pigeon pea pods formed. The largest numbers of pods were in black pigeon pea. It related to the flower initiation, the pigeon pea with the black seeds color more quickly flowering, so the pods formed at 12 weeks after planting had a lot. Another factor was the presence of different skin color pigeon pea seeds indicate the existence of different types of plants that can affect the productivity of the plant. Kurnia et al. [28] stated that in soybeans, varieties affect the numbers of pods in plants.

4. Conclusion

Based on the research and discussion, it could be concluded that the best Fitosan concentration to increase the growth and development of pigeon pea plants is 2 ml l$^{-1}$. Fitosan concentration of 2 ml l$^{-1}$ given by soaking the seeds can accelerate when shoots appear and increase plant height up to 4 weeks after planting. The growth and development response of black, white, and brown pigeon pea showed the difference, that was the fastest flowering black pigeon pea on soaking in Fitosan concentration of 2 ml l$^{-1}$ and yielded the most pods, while white pigeon pea flower appeared fastest in soaking with concentration Fitosan 6 ml l$^{-1}$, while the brown pigeon pea has not flowered and produces pod until the end of observation. There is no correlation between the concentration of Fitosan used in soaking the seeds and the difference of pigeon pea color on the growth and development of pigeon pea.

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