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## Allelopathic effects of lai (*Durio kutejensis* Hassk. Becc) leaf extract, on germination and early growth of weeds and crops

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### ABSTRACT

<sup>1</sup>Lai (*Durio kutejensis* Hassk. Becc) an endemic plant of Kalimantan-Indonesia leaves contain secondary metabolites as allelochemicals (alkaloids, flavonoids, saponins, phenols, and steroids). We investigated the allelopathic effects of lai leaf extract on the seed germination and early growth of weeds (*Ruellia tuberosa* L. and *Amaranthus spinosus* L.) and crops (*Oryza sativa* L. and *Zea mays* L.). We conducted two experiments: Germination bioassay and Early growth greenhouse bioassay consisting of 0 (Control), 25, 50 and 75 % leaf extracts. The leaf extracts significantly decreased the germination and early growth of both test weeds and crops: Germination (%) (100 to 26.66 %), shoot length (9.40 to 3.16 cm); germination index (3.30 to 0.36); vigour index (9.40 to 0.84); biomass 1.43 to 0.14 g; and chlorophyll a (0.45 to 0.016 mg/L), b (0.077 to 0.024 mg/L), and total chlorophyll (0.122 to 0.038 mg/L) contents. In contrast, the extract increased the germination time (3.00 to 7.00 h), mortality (0 to 40 %), and phytotoxicity (0 to 2.33 %) of test plants. Higher concentrations of plant extracts were more inhibitory to germination and early growth in all test crops. Therefore, *D. kutejensis* leaf extract contains a natural compound that can be potentially suitable as an allelopathic-natural herbicide. In future, the results can be used to reduce the use of chemical herbicides in crop production.

**Keywords:** Allelochemical, allelopathy, *Amaranthus spinosus*, bioherbicide, crops, *Durio kutejensis*, germination, *Oryza sativa* L., *Ruellia tuberosa* L., secondary metabolites, seedling growth, weeds, *Zea mays*.

### INTRODUCTION

The abundant biodiversity in Indonesia is strong potential for food, medicines, cosmetics and other bioactivities. However, the potential wealth attributable to this flora has not been utilized and managed optimally. Balikpapan ginger (*Etilingera balikpapanensis*) and lai (*Durio kutejensis* Hassk. Becc) plants are endemic to the Kalimantan region, and they contain certain phytochemicals which promote several bioactivities (31,32). Its fruits are less commonly used than *Durio zibethinus* and *D. kutejensis* plants (28). Besides its leaves have the potential as medicinal, cosmetic and natural bio-herbicides. Its fruit peel extract contained secondary metabolites (flavonoids and saponins), which inhibited the bacterial growth (35). Furthermore, Manurung *et al.* (32) reported phytochemicals compounds (alkaloids, flavonoids, phenolics, saponins, and steroids) and 43 active chemicals in *D. kutejensis* leaf methanol extract. Some of these active compounds showed antioxidant, antibacterial, antimicrobial, antifungal and herbicidal activities. In addition,

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Perez *et al.* (37) reported that its flavonoids and saponins inhibited the growth of other plants. Alegore (5) found that flavonoids and saponins in ketapang (*Terminalia catappa*) leaf extract inhibited the growth of *Cyperus rotundus* L. Likewise, *Leucaena leucocephala* Lam. leaf extract, containing flavonoid and tannins inhibits the growth of weeds such as *Cyperus iria*, *Echinochloa crus-galli*, and *Amaranthus spinosus* (2).

Weeds can produce allelopathic toxic substances that poison neighboring cultivated plants and inhibit their growth. The presence of weeds in a field reduces the absorption of water and nutrients available for cultivated plants, thereby reducing growth of target crop (46). Generally, weed management methods include physical, mechanical, chemical, and biological strategies. Mostly, the use of chemical herbicides has a harmful impact on the environment, animal health, and human health (8,11). Herbicides are cost-effective to manage weeds; but they are quickly losing efficiency due to plant resistance to herbicides (26). weed control on cultivated land involves the use of chemical herbicides, but their use over a long period causes pollution, environmental damage and decrease the biodiversity due to the toxicity and death of microorganisms. In addition, further studies must be done to identify methods to eradicate weeds that are resistant to chemical herbicides. Therefore, research involving plants with natural herbicidal potential (bio-herbicides) that are environmentally friendly are currently being developed to avoid these issues.

Further, biological herbicides has proven to be an effective method to control wide range of weeds (10). These biological herbicides have the potential to minimize the usage of synthetic herbicides, while also reducing their negative environmental effect. In addition, ecological, organic, and family-based farms are increasingly using natural herbicides and allelochemical-based products.

*Durio kutejensis* has bio-herbicide potential, as allelochemical compounds and secondary metabolites found in its leaf extract inhibits weed germination and growth on cultivated land (32,58). However, no published reports have identified and discussed the herbicidal potential of *D. kutejensis* leaves. Thus, this research aimed to determine the allelopathic effects of *D. kutejensis* Hassk. Becc leaf extract on the germination and early growth of weeds and crops.

## MATERIALS AND METHODS

The studies were done in laboratory and greenhouse of Plant Physiology and Development Laboratory the Biology Department, Faculty of Mathematics and Natural Sciences at Mulawarman University in Samarinda, East Kalimantan, Indonesia (Coordinates: 0°21'18"-1°09'16" South and 116°15'36"-117°24'16" East). The leaves were collected in June 2022 from the area surrounding Mulawarman University.

### Seeds and planting media preparation

Pletekan (*Ruellia tuberosa*), thorny spinach (*Amaranthus spinosus* L), rice (*Oryza sativa* L), and corn (*Zea mays* L) seeds were used in this study. *Ruellia tuberosa* L and *A. spinosus* seeds were collected from farms and *O. sativa* and *Z. mays* seeds were purchased from local markets in Samarinda, East Kalimantan, Indonesia. The seeds used were fully mature, these were air-dried for 3 d and stored in plastic pots before use. Seed selection was done by immersing seeds in water; seeds that sink indicated that the seeds are good and were

used in the germination test stage. For germination, 10 seeds of each test plant were used. The planting media consisted of mixture of topsoil and humus (1:1 ratio) and then placed in 12 cm dia polybags.

#### Preparation of leaf extract

The mature *D. kutejensis* leaves were rinsed with distilled water and allowed to air-dry in our Lab. for 14 d. The dried leaves were chopped into small pieces and pulverized in an electric blender. The *D. kutejensis* leaf powder (500 g) was macerated in 98 % methanol for 3d, thereafter it was incubated for 24 h and shaken at 150 rpm. The leaf extract was filtered using Whatman No. 1 filter paper and the filtrate was dried in rotary evaporator at 40 °C. Then allowed to cool to 4°C, and diluted to 25, 50 and 75 % concentrations for further usage. These leaf extracts were used to determine their effects on germination and early growth of pletekan (*Ruellia tuberosa*), thorny spinach (*Amaranthus spinosus*), rice (*Oryza sativa*), and corn (*Zea mays*).

#### Seed Germination Bioassay

All test weed and crop seeds were treated with 0, 25, 50, 75 % leaf extract (distilled water was used as control). Ten cotton-coated Petri plates were sown with 10-seeds of each crop, and 10 mL extract or distilled water was added to each Petri plate, which were placed in germinator box at 20 °C for 10 d. During this period, seed germination was regularly monitored and extract water was applied to each Petri dish as required. Mean germination time (MGT), germination (%), shoot length (cm), germination index, and seedling vigor index (SVI) were measured throughout the germination process. Germination was deemed to occur when the emerged radical size from seeds was 1-2 mm and the germinated seeds were counted daily.

The proportion of germinated seeds of each test crop was estimated 10-days after sowing using the following formula :

(i). Germination (%) = Germinated seed/total seed ×100.

(ii). Mean germination time (MGT) was calculated using the formula of Tanveer *et al.* (47) :

$$MGT = \frac{\sum(Dn)}{\sum n}$$

Where n : Total number of seeds emerging on day D and D : Total days from the start of germination.

(iii). Germination Index (GI) was counted by following equation below:

$$GI = \frac{\text{No. of germinated seeds}}{\text{Days of first count}} + \dots + \frac{\text{No. of germinated seeds}}{\text{Days of final count}}$$

(iv). Seedling vigour index (SVI) was counted based on equation of Abdul-baki and Anderson (1).

$$SVI = \frac{\text{Germination}}{\text{Emergence}} \% \times \text{Radical length (mm)}$$

### Greenhouse bioassay

A second bioassay was done in greenhouse, to determine the effects of *D. kutejensis* leaf extract on the test crops. Five seeds of each test crop were sown in polybags filled with soil and grown and maintained for 3 weeks. During the 4<sup>th</sup> week after emergence, the entire leaf surfaces of the seedlings were sprayed with 10 mL of 25, 50, or 75 % *D. kutejensis* extract or distilled water (as a control). Spraying was done daily for 7 d, thereafter, the biomass (fresh and dry weight), phytotoxicity, mortality, chlorophyll a, chlorophyll b, and total chlorophyll concentrations were determined.

The chlorophyll contents were estimated using the method of Harborne (18). Fresh leaves (1 g) were cut into small pieces, crushed, dissolved in 100 mL of 95 % alcohol and homogenized. The solution was filtered using Whatman No. 1 filter paper. The chlorophyll concentration was measured using a UV-VIS spectrophotometer at optical absorbances of 663 nm and 646 nm. The various chlorophyll contents were calculated using the following equations:

$$\begin{aligned}\text{Chlorophyll a (mg/L)} &= (12.21 \times A_{663}) - (2.81 \times A_{646}) \\ \text{Chlorophyll b (mg/L)} &= (20.13 \times A_{646}) - (5.03 \times A_{663}) \\ \text{Total chlorophyll (mg/L)} &= (17.3 \times A_{646}) + (7.18 \times A_{663})\end{aligned}$$

### Data analysis

The SPSS version 24 (SPSS, Inc., USA) was used to examine the collected data. ANOVA and Duncan's multiple range tests were performed to evaluate the significant difference among samples. A significance level of 0.05 was applied.

## RESULTS AND DISCUSSION

### Seed germination bioassay

We tested whether *D. kutejensis* leaf extract can allelopathically affect the germination and early growth of test crop weeds (*Ruellia tuberosa* and *Amaranthus spinosus*) and crop plants (*Zea mays* and *Oryza sativa*). During the germination stage (seed germination bioassay), the *D. kutejensis* leaf extract slowed the mean germination time (MGT) and reduced the germination (%), shoot length, germination index (GI), and seedling vigor index (SVI). Furthermore, the greenhouse bioassay revealed that the extract reduced the biomass and the contents of chlorophyll a, chlorophyll b, and total chlorophyll, and increased the plant mortality and phytotoxicity during the early stages of growth.

### Mean germination time and germination (%)

Methanolic leaf extract of *D. kutejensis* significantly affected the MGT and germination (%) of test weeds and crops, with inhibition of MGT and germination (%) at all test concentrations than the control. The higher extract concentrations corresponded to greater inhibition of MGT and germination percentage (Table 1). The inhibitory allelopathic effect of *D. kutejensis* leaf extract on germination of all test crops are shown in Figure 1.

Table 1. Allelopathic effects of *D. kutejensis* leaf extract on mean germination time (MGT) (days) of test crop.

Test crop	Extract concentration (%)			
	0 (control)	25	50	75
	MGT			
<i>R. tuberosa</i>	4.00 <sup>d</sup>	5.33 <sup>bc</sup>	5.66 <sup>b</sup>	7.00 <sup>a</sup>
<i>A. spinosus</i>	3.66 <sup>d</sup>	5.00 <sup>bc</sup>	5.66 <sup>ab</sup>	6.33 <sup>a</sup>
<i>Z. mays</i>	3.00 <sup>d</sup>	4.33 <sup>bc</sup>	4.66 <sup>b</sup>	6.00 <sup>a</sup>
<i>O. sativa</i>	3.00 <sup>d</sup>	4.66 <sup>bc</sup>	5.00 <sup>b</sup>	6.33 <sup>a</sup>
	Germination (%)			
<i>R. tuberosa</i>	100.00 <sup>a</sup>	86.66 <sup>b</sup>	66.66 <sup>c</sup>	36.66 <sup>d</sup>
<i>A. spinosus</i>	100.00 <sup>a</sup>	73.33 <sup>b</sup>	53.33 <sup>c</sup>	26.66 <sup>d</sup>
<i>Z. mays</i>	100.00 <sup>a</sup>	70.00 <sup>b</sup>	46.66 <sup>c</sup>	30.00 <sup>d</sup>
<i>O. sativa</i>	100.00 <sup>a</sup>	66.66 <sup>b</sup>	43.33 <sup>c</sup>	26.66 <sup>d</sup>

Note: Different letters within a row indicate a significant difference ( $P < 0.05$ ) based on Duncan's multiple range test.

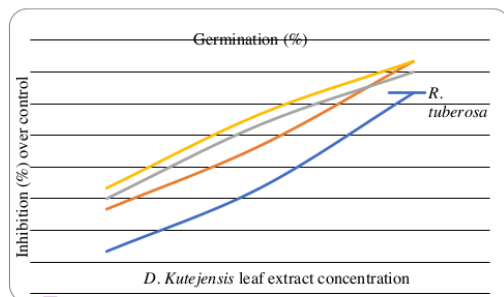


Figure 1. Inhibitory allelopathic effects of *D. kutejensis* leaf extract on germination (%) of test crop.

The longer MGTs and lower germination (%) indicated that the leaf extract of *D. kutejensis* may contain phytotoxins that affects the germination process. Certain phytotoxins released from plants can inhibit the seedling development and growth and the present findings corroborate those of previous studies in which allelopathic compounds (phytotoxin) that entered into the seeds with water inhibited the formation of growth hormones such as gibberellins and indole acetic acid (33,34,57). Phytotoxins affects and decreases the performance of enzymes involved in the hydrolyzation of starch into glucose in the endosperm or cotyledons, leading to reduced growth energy that can inhibit the germination process (6,33). Allelochemicals have the potential to harm feeble seeds, prevent the absorption of nutrients and stunt the growth and development of seedlings. The higher the extract concentration, the longer it takes for the seeds to germinate and the lower the

germination (%). This finding was supported by Senjaya and Surakusumah (42), who reported that the allelochemical compounds found in various extracts of *Pinus merkusii* inhibited the germination of *Echinochloa colonum* and *Amaranthus viridis* and could be used as inhibitory bio-herbicides against seed germination of these species. Similarly, Kurniati *et al.* (30) found that extracts of *Imperata cylindrica* that contained allelopathic compounds inhibited the germination (%) of *Oryza sativa*. Wang *et al.* (54) also reported that herbs (*Achnatherum splendens*, *Artemisia frigida*, *Stellera chamaejasme*) aqueous extract inhibited lettuce seed germination and root length. The agriculture industry will benefit greatly, if a plant extract can prevent weed seeds from germination because this will slow down the growth of weeds and increase the yield of cultivated plants. In addition, application of plant extracts of velvetleaf (*Abutilon theophrasti*), ragweed (*Ambrosia artemisiifolia*) and cocklebur (*Xanthium strumarium*) to seeds of lettuce and tomato their seeds significantly inhibited germination (concentration of 50 %) and early seedling growth (49).

#### Shoot length, Germination index (GI), and Seedling vigor index (SVI)

All concentrations of *D. kutejensis* leaf extract significantly affected the shoot length, GI, and SVI in both test groups (weed and crop plant), when compared to the control (Table 2). As the concentration of methanolic leaf extract increased, the shoot length, GI, and SVI decreased. The presence of phytotoxin in higher concentrations of *D. kutejensis* leaf extract could explain the lower values of these growth factors. Figure 2 illustrates the inhibitory allelopathic effects of *D. kutejensis* leaf extract on test crops shoot length (cm).

Table 2. Allelopathic effects of *D. kutejensis* leaf extract on germination index (GI), and seedling vigour index (SVI) of test crops.

Test crop	Extract concentration (%)			
	0 (control)	25	50	75
<b>Germination Index (GI)</b>				
<i>R. tuberosa</i>	2.50 <sup>a</sup>	1.63 <sup>b</sup>	1.17 <sup>c</sup>	0.52 <sup>d</sup>
<i>A. spinosus</i>	2.76 <sup>a</sup>	1.46 <sup>b</sup>	0.94 <sup>c</sup>	0.41 <sup>d</sup>
<i>Z. mays</i>	3.30 <sup>a</sup>	1.65 <sup>b</sup>	1.01 <sup>c</sup>	0.50 <sup>d</sup>
<i>O. sativa</i>	3.30 <sup>a</sup>	1.46 <sup>b</sup>	0.86 <sup>c</sup>	0.36 <sup>d</sup>
<b>Seedling Vigor Index (SVI)</b>				
<i>R. tuberosa</i>	7.86 <sup>a</sup>	5.31 <sup>b</sup>	3.14 <sup>c</sup>	1.23 <sup>d</sup>
<i>A. spinosus</i>	6.23 <sup>a</sup>	3.58 <sup>b</sup>	2.21 <sup>c</sup>	0.84 <sup>d</sup>
<i>Z. mays</i>	9.40 <sup>a</sup>	5.32 <sup>b</sup>	2.60 <sup>c</sup>	1.10 <sup>d</sup>
<i>O. sativa</i>	7.70 <sup>a</sup>	4.14 <sup>b</sup>	2.06 <sup>c</sup>	0.89 <sup>d</sup>

Note: Different letters within a row indicate a significant difference ( $P < 0.05$ ) based on Duncan's multiple range test.

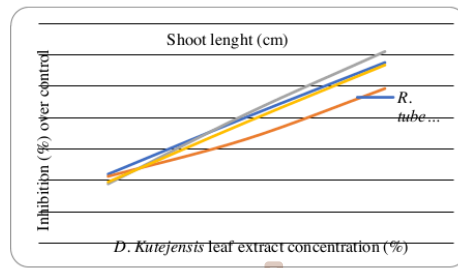


Figure 2. Inhibitory allelopathic effects of *D. kutejensis* leaf extract on shoot length (cm) of test crops.

The growth reduction occurred because allelopathic compounds that entered the plant in the form of phenolics inhibited the process of cell mitosis, through destruction of spindle threads in the cell at the metaphase stage. Cell division in the plant was consequently hindered, which inhibited growth and reduced shoot length (56). Manurung *et al.* (33) stated that the methanolic leaf extract of *D. kutejensis* has secondary metabolites (alkaloids, flavonoids, phenolics, saponins, and steroids) called allelochemicals or allelopathic compounds. These metabolites can be phytotoxic or act as stimulants for neighbouring plants (15,40). Studies have reported that allelopathic plant extracts inhibited the germination and growth of test plants. For example, the allelopathic compounds found in Acacia extract inhibited the shoot growth of *Echinochloa crus-galli* (7). The present research showed that the highest extract concentration (75 %) caused the greatest reduction in shoot length, possibly because of the higher concentration of allelochemicals compared to the 50 % and 25 % extracts and the control. These findings corroborates Julio *et al.* (22), who reported that increasing concentrations of the allelochemical compounds found in *Chromolaena odorata* extract inhibited the *Vigna radiata* germination and seedling growth.

Furthermore, allelopathic compounds can interfere with or stop the action of enzymes in the cell wall, which disrupts energy formation and results in a slowing of the growth process and an inhibition of seed germination, which negatively affects the germination index and germination time (15,16,45). These results correspond to those of Respatie *et al.* (39), who used the *Cosmos sulphureus* flower as a source of allelochemical compounds to inhibit the growth and development of *Cyperus rotundus*. Similarly, El-Rokiek *et al.* (16) identified that pea seed powder had allelopathic and phytotoxic effects that prevented the growth of weeds in wheat.

The present findings are consistent with Channappagoudar *et al.* (9), who reported that allelochemical-containing extracts of *Cyperus rotundus* and *Commelina benghalensis* inhibited the germination, seedling length, and seedling vigour index of wheat, sorghum, and soybean. Stavrianakou *et al.* (44), Dongre and Yadav (12), and Agarwal *et al.* (3) reported suppression of plumule and radicle length and biomass losses in wheat, peas and lentils using aqueous extracts of several weeds. According to Shukla *et al.* (43), the unique



action of the extract and the dramatic slowing down of seedling growth might be explained by the presence of high concentrations of inhibitory chemicals in the leaves. Seedling emergence, seedling vigour index, growth rate, and total dry weight were significantly reduced, when wheat, chickpea and lentil seedlings were germinated and grown in soil containing water extracts of various organs (root, stem, leaf, and fruit) of *Euphorbia helioscopia* L. (47). In addition, El-Mergawi and Al-Humaid (14) revealed that *Tamarix mannifera* and *Lactuca virosa* extracts inhibited the seed germination and seedling development in target species (*Phalaris minor*, *Echinochloa crusgalli*, *Portulaca oleracea*, and *Lactuca sativa*), at high concentrations.

### GREENHOUSE BIOASSAY

#### Chlorophyll content

The effects of lai leaf methanol extract on the chlorophyll contents of weeds and cultivated plants are shown in Table 3. All concentrations (25, 50, and 75 %) of extract significantly affected the chlorophyll content compared to control (0 %) group. Furthermore, chlorophyll production in the weeds and crops decreased as the extract concentration increased. These results indicated that the allelopathic compounds in *D. kutejensis* leaves inhibited the chlorophyll formation.

Table 3. Allelopathic effects of *D. kutejensis* leaf extract on chlorophyll a, chlorophyll b, and total chlorophyll (mg/L) on test crops.

Test crop	Extract Concentration (%)			
	0	25	50	75
<b>Chlorophyll a</b>				
<i>R. tuberosa</i>	0.045 <sup>a</sup>	0.042 <sup>b</sup>	0.039 <sup>c</sup>	0.032 <sup>d</sup>
<i>A. spinosus</i>	0.037 <sup>a</sup>	0.024 <sup>b</sup>	0.020 <sup>c</sup>	0.016 <sup>d</sup>
<i>Z. mays</i>	0.033 <sup>a</sup>	0.028 <sup>b</sup>	0.023 <sup>cd</sup>	0.021 <sup>d</sup>
<i>O. sativa</i>	0.023 <sup>a</sup>	0.021 <sup>ab</sup>	0.019 <sup>c</sup>	0.014 <sup>d</sup>
<b>Chlorophyll b</b>				
<i>R. tuberosa</i>	0.077 <sup>e</sup>	0.074 <sup>e</sup>	0.063 <sup>f</sup>	0.056 <sup>c</sup>
<i>A. spinosus</i>	0.062 <sup>f</sup>	0.040 <sup>c</sup>	0.032 <sup>b</sup>	0.024 <sup>a</sup>
<i>Z. mays</i>	0.057 <sup>c</sup>	0.048 <sup>d</sup>	0.039 <sup>c</sup>	0.038 <sup>c</sup>
<i>O. sativa</i>	0.040 <sup>c</sup>	0.037 <sup>c</sup>	0.033 <sup>b</sup>	0.024 <sup>a</sup>
<b>Total Chlorophyll</b>				
<i>R. tuberosa</i>	0.122 <sup>a</sup>	0.117 <sup>b</sup>	0.101 <sup>c</sup>	0.088 <sup>d</sup>
<i>A. spinosus</i>	0.099 <sup>a</sup>	0.064 <sup>b</sup>	0.052 <sup>c</sup>	0.040 <sup>d</sup>
<i>Z. mays</i>	0.090 <sup>a</sup>	0.077 <sup>b</sup>	0.062 <sup>c</sup>	0.060 <sup>c</sup>
<i>O. sativa</i>	0.064 <sup>a</sup>	0.059 <sup>ab</sup>	0.052 <sup>c</sup>	0.038 <sup>d</sup>

Note: Different letters within a row indicate a significant difference ( $P < 0.05$ ) based on Duncan's multiple range test.

Allelopathic compounds such as phenols can damage the chlorophyll structure, including that of chlorophyll a, chlorophyll b, and total chlorophyll, thereby disrupting the photosynthesis process and inhibiting the formation of glucose compounds, resulting in growth decline or death of plants (20,25, 291). Narwal (36) reported that the allelopathic potential of *Helianthus annuus* can reduce the germination, growth and chlorophyll content

of *Portulaca oleracea* and *Flaveria australasia*. In addition, allelopathic compounds that enter the soil affect the absorption of nutrients by plants, which leads deficiency of nutrients for chlorophyll formation, thereby indirectly affecting plant chlorophyll levels (17).

#### Biomass, Mortality percentage, and Phytotoxicity

The allelopathic effects of *D. kutejensis* leaf extract on the biomass and mortality (%) and the phytotoxicity effects on the test plants are listed in Table 4. The application of *D. kutejensis* leaf extract significantly affected all three components in both weeds (*R. tuberosa* and *A. spinosus*) and cultivated plants (*Z. mays* and *O. sativa*). The 75 % extract concentration produced the lowest biomass and significantly different results than other treatments for both the weed and cultivated plant test crops. The highest percentage of mortality and phytotoxicity was found with the 75 % extract, which significantly differed from 50 % and 25 % extract and the control. Higher concentrations of leaf extract corresponded to higher levels of phytotoxicity. The allelochemical compounds contained in the methanol extract of *D. kutejensis* leaves inhibited the plant biomass in all test crops. The higher the concentration of *D. kutejensis* leaf extract, the lower the biomass of all test crops produced. Inhibitory allelopathic effects of *D. kutejensis* leaf extract on the biomass-dry weight (g) of test crops are shown in Figure 3.

Table 4. Allelopathic effects of *D. kutejensis* leaf extract on mortality (%), and phytotoxicity (%) of test crops.

Test crop	Extract concentration (%)			
	0 (control)	25	50	75
<b>Mortality (%)</b>				
<i>R. tuberosa</i>	0.00 <sup>a</sup>	0.00 <sup>a</sup>	0.00 <sup>a</sup>	26.66 <sup>b</sup>
<i>A. spinosus</i>	0.00 <sup>a</sup>	0.00 <sup>a</sup>	0.00 <sup>a</sup>	26.66 <sup>b</sup>
<i>Z. mays</i>	0.00 <sup>a</sup>	0.00 <sup>a</sup>	0.00 <sup>a</sup>	6.66 <sup>b</sup>
<i>O. sativa</i>	0.00 <sup>a</sup>	0.00 <sup>a</sup>	0.00 <sup>a</sup>	40.00 <sup>b</sup>
<b>Phytotoxicity</b>				
<i>R. tuberosa</i>	0.00 <sup>a</sup>	0.00 <sup>a</sup>	0.33 <sup>b</sup>	1.33 <sup>c</sup>
<i>A. spinosus</i>	0.00 <sup>a</sup>	0.00 <sup>a</sup>	1.00 <sup>b</sup>	2.00 <sup>c</sup>
<i>Z. mays</i>	0.00 <sup>a</sup>	0.00 <sup>a</sup>	1.00 <sup>b</sup>	1.67 <sup>c</sup>
<i>O. sativa</i>	0.00 <sup>a</sup>	0.00 <sup>a</sup>	0.67 <sup>b</sup>	2.33 <sup>c</sup>

Note: Different letters within a row indicate a significant difference ( $P < 0.05$ ) based on Duncan's multiple range test.

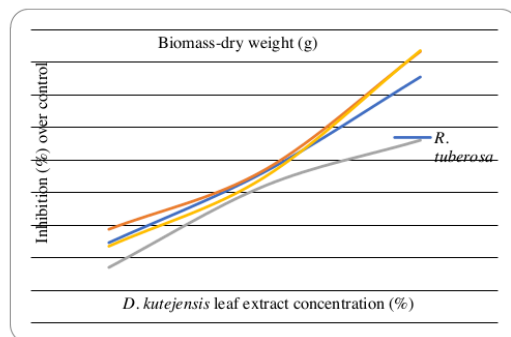


Figure 3. Inhibitory allelopathic effects of *D. kutejensis* leaf extract on biomass-dry weight (g), of test crops.

Allelochemical compounds reduces the ability of plants to absorb ions from the soil and those from the phenol group can inhibit potassium absorption from the soil. Potassium deficiencies can lead to plant growth inhibitions and affect plant weight and size (41). Manurung *et al.* (32) showed that methanolic leaf extract of *D. kutejensis* contained secondary metabolites consisting of 43 bioactive compounds, including legal; 2-(1,1-dimethyl ethyl)-phenol; phenol, 4-(3-hydroxy-1-propenyl)-2-methoxy (coniferyl alcohol); E7-decenylacetate; and pentadecanoic acid, 14-methyl-, methyl ester. These bioactive compounds are toxic to plant cells and are present in herbicides, pesticides, nematocides, larvicides, antifungals and antimicrobials (4,13,51,52). Using plant extracts such as *D. kutejensis* leaf extract as herbicide is safer and more environmentally friendly than chemical herbicides (chlorophenols, glyphosate, picloram, atrazine). Chemical herbicide use poses a serious risk to people, the environment, and nearby living organisms. Herbicide residues from chemical herbicides can be inhaled, consumed through food, applied topically during the application, or encountered, when one enters an area where the compounds have recently been used. Chemical herbicide exposure can cause nausea, vomiting, diarrhea, headaches, disorientation, strange or aggressive behavior, strange breath odor, muscle weakness, peripheral neuropathy, and loss of reflexes. US EPA (50) reported that chemical herbicide exposure can cause plants to produce fewer amino acids, divide fewer cells, and have higher internal herbicide concentrations. By altering habitat and food availability, effects on aquatic plants can have an indirect impact on fish and invertebrates.

The allelochemical compounds in *D. kutejensis* leaf extract could inhibit water absorption from the soil, break down chlorophyll, cause stomatal closure, and damage cell plasma membranes. This reduction of these functions decreases the ability of plants to photosynthesize and form organic matter, which decreases the plant dry weight (20,25). Allelopathic compounds in plants can cause disorders such as the inhibition of germination or growth, wilting, or death of the plants. Adawiah (2) reported that allelopathic compounds found in leaf extract of *Leucaena leucocephala* decreased the biomass of weeds (*Cyperus iria*, *Echinochloa crus-galli*, and *Amaranthus spinosus*). Ketapang leaf extract (*Terminalia*

*catappa*) which also contains allelopathic compounds, was used as a bioherbicide against *Cyperus rotundus* weed and the extract of this plant increased the mortality percentage (5).

The reduction in phytotoxicity was proportional to the concentration of lai leaf extract used because allelopathic compounds contained in *D. kutejensis* leaves cause a poisoning effects [yellowing of leaves, wilting, tissue damage or necrosis, and death in the affected plants (48,59)]. This finding is consistent with Hussain *et al.* (21), who determined that the extract of the aerial foliage (flowers and phyllodes) of *Acacia melanoxylon* had a detrimental influence on the growth and biomass of *Lactuca sativa* seedlings. Furthermore, aqueous and methanolic extracts of *Sapindus rarax* prevented the growth of *L. chinensis* and *F. mileacea* weeds at concentrations of 25 %, 50 %, and 75 % (38). In addition, Kaab *et al.* (23) demonstrated that the methanolic extract of *Cynara cardunculus* prevented the weeds germination, hindered seedling development, and induced necrosis or chlorosis in the affected plants. The current study found that the leaf extract of *D. kutejensis* could decrease germination and growth and increase the mortality (%) in weeds and cultivated plants, suggesting that this extract could be effective as a bio-herbicide to replace chemical herbicides. Utilizing plant extracts with bio-herbicide potential has the benefit of minimal toxicity, easy environmental degradation, and some weed control potential, which lowers the likelihood of weed resistance to herbicides. For a broad adoption of bio-herbicides, appropriate formulas and usage methods that enable a uniform application of the biological agent in the desired location without wasting or excessive consumption of the bio-herbicide that would increase the cost of treatment and the risk of effects on non-organisms are required (19,27). To be successful, bio-herbicides must be competitive in price, as well as efficient and consistent (high virulence and appropriate formulations). Competitive pricing is achieved by the optimization of fermentation processes, efficient bioreactors, and the use of low-added-value raw materials (24). Moreover, to commercialize bio-herbicides, might arise a specific toxin compound, for example, mycotoxins which are highly hazardous to mammals. The formation of mycotoxins by putative bio-herbicides might impede or prevent the commercialization of such bacteria (55).

### CONCLUSIONS

The leaf extract of *Durio kutejensis* Hassk. Becc inhibited seed germination and early growth of the tested weeds (*Ruellia tuberosa* L. and *Amaranthus spinosus* L.) and crops (*Oryza sativa* L. and *Zea mays* L.). Higher concentrations of plant extract increased the inhibition of germination and early growth in all test crops. In addition, higher concentrations of the extract created greater phytotoxicities. These results can be used to improve agriculture and reduce the use of herbicides in plant cultivation. Therefore, in future, a natural compound in *D. kutejensis* leaf extract can be potentially suitable as an allelopathic-natural bio-herbicide to replace chemical herbicides. To confirm these results, more research is needed under field conditions.

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### DECLARATION

We declare that all authors of this Ms. have made substantial contributions. We did not exclude any author who substantially contributed to this Ms. We have followed the ethical norms established by our respective institutions.

### CONFLICT OF INTEREST

The authors announce that they have no conflict of interest.

### ETHICAL APPROVAL

The authors declare that the study was carried out following scientific ethics and conduct. However, this study did not involve any use of animals, hence no ethical approval has been obtained from the concerned committee.

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