# *In Vitro* Sensitivity of *Phytophthora infestans* (Mont.) de Bary Isolated from Cikajang, Garut, to Several Fungicides

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**ABSTRACT/ABSTRAK**

**Evaluasi Secara *In Vitro* Sensitivitas *Phytophthora infestans* (Mont.) de Bary Isolat Cikajang Garut terhadap Beberapa Fungisida**

Late blight disease caused by oomycete *Phytophthora infestans* is one of the major diseases on potatoes. The standard way used by farmers to control late blight disease is by using synthetic fungicides. However, continuous usage of similar fungicides can lead to the emergence of resistant pathogen populations and therefore reduce the fungicide's effectiveness. This study evaluated the sensitivity of *P. infestans* isolated from a potato plantation in Cikajang Village, Garut Regency, to several commonly used fungicides (metalaxyl, mancozeb, dimetomorph, chlorothalonil, and oxathiapiprolin). The experiment was conducted using poisoned food assay and detached-leaf assay. The results demonstrated that *P. infestans* was sensitive to metalaxyl and mancozeb at concentration of 2.000 ppm with the colony growth suppression of 93.3%. Furthermore, based on the detached leaf assay, metalaxyl caused lesion suppression by 100%, whilst mancozeb 70.89%. The intermediate sensitivity of *P. infestans* to dimetomorph was shown at the concentration of 1.000 ppm and chlorothalonil at the concentration of 2,000 ppm with each colony growth suppression of 65.5% and 75.8%, and lesion suppression of 54.7% and 59.1%. The study also found an indication of reduced sensitivity of *P. infestans* to oxathiapiprolin as shown by the *P. infestans* colony growth suppression of only 8.5% and lowest necrotic lesion suppression compared to the other four fungicides at the value of 48.8%.

Kata Kunci:
Dimetomorf,
Klorotalonil,
Mankozeb,
Metalaksil,
Oksatiapiprolin

Penyakit hawar daun yang disebabkan oleh oomycete *Phytphthora infestans* merupakan salah satu penyakit penting pada tanaman kentang. Metode pengendalian utama yang digunakan oleh petani adalah dengan menggunakan fungisida sintetis. Akan tetapi, penggunaan fungisida secara terus menerus dapat menyebabkan timbulnya populasi patogen yang resisten sehingga dapat menurunkan keefektifan fungisida tersebut. Penelitian ini dilaksanakan dengan tujuan untuk mengevaluasi sensitivitas *P. infestans* yang diisolasi dari pertanaman kentang di Desa Cikajang, Kabupaten Garut terhadap beberapa fungisida berbahan aktif metalaksil, mankozeb, dimetomorf, klorotalonil dan oksatiapiprolin. Eksperimen dilaksanakan dengan menggunakan metode makanan beracun dan *detached-leaf assay*. Hasil eksperimen menunjukkan bahwa *P. infestans* sensitif terhadap metalaksil dan mankozeb pada konsentrasi 2000 ppm dengan penghambatan pertumbuhan koloni masing-masing mencapai 93,3%. Lebih lanjut, berdasarkan hasil *detached leaf assay*, penghambatan terbentuknya lesi mencapai 100% pada perlakuan fungisida

berbahan aktif metalaksil. Sementara penghambatan sebesar 70% diperoleh pada perlakuan fungisida berbahan aktif mankozeb. Sensitivitas *P. infestans* dikategorikan sebagai sedang terhadap fungisida berbahan aktif dimetomorf pada konsentrasi 1000 ppm dan klorotalonil pada konsentrasi 2000 ppm dengan penghambatan koloni masing-masing sebesar 65,5% dan 75,8% serta penghambatan pembentukan lesi 54,7% dan 59,1%. Eksperimen ini juga menemukan adanya indikasi penurunan sensitivitas *P. infestans* terhadap oksatiapiprolin yang ditunjukkan dengan rendahnya penghambatan pertumbuhan yang hanya sebesar 8,5% dengan penghambatan pembentukan lesi sebesar 48,8% ketika dibandingkan dengan keempat jenis fungisida lainnya.

## INTRODUCTION

Fungicide is essential in agriculture to control plant-disrupting organisms such as pathogens. Using fungicides to control plant pathogens cannot be removed completely in plant cultivation practices (Wismaningsih *et al*., 2016). The dosage and concentration of fungicides become one of the important aspects in the effectiveness of fungicides. Application of sub-lethal dose or concentration can affect the fungicides in controlling plant pathogens (Damicone, 2008). In addition, the reduction of effectiveness of fungicides can also be caused by the reduction in sensitivity of pathogens to fungicides. Reduction in sensitivity occurs due to the emergence of resistant pathogen population triggered by continuous usage of similar fungicides (Byamukama *et al*., 2019).

*Phytophthora infestans* is one among many plant pathogens that has been reported resistant to various fungicides (Brent & Hollomon, 2007; Deising *et al.,* 2008). Potato late blight disease is commonly controlled by fungicides with various active ingredients such as metalaxyl, azoxystrobin, dimetomorph, fluazinam, chlorothalonil, mancozeb, and cymoxanyl (Pobedinskaya *et a*l., 2012; Yuan *et al*., 2006). Repeated use of similar doses and types of fungicides contributed to the emergence of resistant population of pathogens (Sumardiyono, 2008). The resistance of *P. infestans* to fungicides can also be caused by heterothallic pathogens in which causing diverse genetic variation of *P. infestans* (Purwanti, 2002).

In Indonesia, a reduction of *P. infestans* sensitivity to chlorothalonil was reported in potato plantation in Kerinci Regency, Jambi (Yusuf, 2018). A similar case was also found on *P. infestans* that infect tomato crops in Karo Highlands. It was reported that the application of mancozeb was no longer effective in controlling leaf blight disease in tomato crops (Syahputra, 2018). Dangi *et al*. (2020) also stated an indication of reduction sensitivity of *P. infestans* at potato plantations in Wonosobo and Pasuruan to metalaxyl with a concentration of 5 mg/l. This study aimed to detect the reduction of several fungicide's effectiveness in suppressing the growth of *P. infestans* isolated from Cikajang, Garut.

## MATERIAL AND METHODS

### Isolation, Purification, and Identification of *Phytophthora infestans*

The isolate of *Phytophthora infestans* was collected from Cikajang, Garut, West Java. Samples were put inside a labeled container and kept in a polystyrene box with ice until they were isolated in the laboratory. *P. infestans* was isolated from the infected leaf with potato sandwich method (Tumwine *et al*., 2000). Healthy potato tubers were peeled and surface sterilized in 200 ml sterile distilled with chloramphenicol 1mg/l and nystatin 1 mg/l for about five minutes and dried for about two minutes with sterile tissue paper. The potato tubers were cut into 2-3 mm thick slices using a sterile knife and sterile slicing board. Pieces of infected potato leaves of approximately 1 $cm^2$ were cut from a leading edge of the necrotic lesion. The infected discs were then covered by the sliced potato tuber and incubated at 18-20°C. The suspected growth mycelium was aseptically transferred onto BSEA (Bean Sprout Extract Agar) and incubated at 18-20°C for five days. The suspected colony was identified macroscopically and microscopically. The result of *P. infestans* isolation was determined based on microscopic morphology using the key of pathogen determination on CABI and necrotic formation that showed on potato leaves.

### Fungicide Testing Using Poisoned Food Assay

This experiment aimed to determine the sensitivity of *P. infestans* Cikajang Garut isolate to several commonly used fungicides with different active ingredients based on the response of the colony growth in BSEA containing several levels of fungicides concentrations. This experiment was conducted using poisoned food assay (Dhingra & Sinclair, 1985). Culture medium (BSEA) was mixed with fungicide dilution in different concentrations (X, 1/2X, 1/4X, and 1/8X, with X being the recommended concentration as advised by the manufacturer) and without fungicide treatment as the control. The recommended concentration of metalaxyl was 2,000 ppm, mancozeb 2,000 ppm, dimetomorph 1,000 ppm, chlorothalonil 2,000 ppm, and oxathiapiprolin 3,000 ppm.

A Purified colony of *P. infestans* was cut into plugs using a 6 mm cork borer and an inoculum plug was put in the middle of medium in petri dishes. The colony growth was observed from day 1 of incubation until the control treatment had filled the surface of the culture medium at 24 hrs intervals. The percentage of the colony growth inhibition of *P. infestans* by fungicides were calculated by the formula developed by McKinney (1923):

$$I = \frac{(C - T)}{C} \times 100\%$$

Where $I$ represents the percentage of growth inhibition of the *P. infestans* (%), C and T indicate radial growth of *P. infestans* in control (C) and treatment (T) (cm).

The data were analyzed with Microsoft Excel 2007 to determine the effective concentration to suppress 50% of *P. infestans* colony growth ($EC_{50}$).

### Fungicide Testing Using Detached-leaf Assay

This test was conducted to determine the rate of damage caused by *P. infestans* on the potato leaves after being given fungicides with several concentrations. This test was done using detached-leaf assay by Goth & Keane (1997). The healthy potato leaves were washed with sterilized distilled water and keep it air dried before being dipped into fungicide solutions with respective concentrations (X, 1/2X, 1/4X, and 1/8X, with X being the recommended concentration as advised by the manufacturer). The leaves were then air dried and placed abaxial surface in a moist chamber made from transparent plastic containers containing layers of moist tissue paper and some straws to separate the leaves and the tissue paper. The leaves were pricked with a sterilized needle, then *P. infestans* was inoculated to each treatment on the leaves by placing a 10 mm inoculum plug on the injured part. The assessments were calculated based on the latent period, the rate of necrotic development, and the diameter of necrotic lesion caused by *P. infestans*.

The latent period was measured by counting the days from inoculation until the first day that necrotic lesion appeared. The rate of necrotic development was observed 3-5 days after inoculation and calculated by this formula (Rosmayati *et al.*, 2006):

$$\mathrm{r} = \frac{xj - xi}{tj - ti}$$

Where $r$ represents the rate of necrotic development, $Xi$ and $Xj$ indicate the diameter of necrotic lesion, $tj$ *and* $ti$ indicate the day of observation.

The necrotic lesion was observed 3-5 days after inoculation and calculated by this formula (Vleeshouwers *et al.*, 1999):

$$A = \frac{\pi \times p \times l}{4}$$

Where $A$ represents the necrotic lesion width whereas $p$ and $l$ represent the diameter of the vertical and horizontal lesion.

### Statistical Analysis

The data were analyzed with ANOVA and followed by Duncan's Multiple Range Test and at a significant level of 0.05 using SPSS version 26.0 and Scott Knott Test using Microsoft Excel 2007 to compare each treatment.

## RESULTS AND DISCUSSION

### History of Fungicides Used on Potato Plantation in Cikajang Garut

*Phytophthora infestans* isolate that was used in this experiment (Figure 1a) was isolated from potato plantation in Cikandang Village, Cikajang, Garut (Figure 1b), which grows potatoes almost every season. Therefore, *P. infestans* inoculum is available year-round. Samples of infected potato leaves by *P. infestans* were taken from a potato crop Granola variety with a lifespan of ±115 days (Figure

1c). Fungicides commonly used at the sampling site were fungicides with active ingredients dimetomorph, chlorothalonil, and oxathiapiprolin. However, over the past two years, oxathiapiprolin fungicide was more commonly applied by many farmers as it was considered an effective fungicide in controlling *Phytophthora* leaf blight.


Figure 1. *Phytophthora infestans* isolated from Cikajang Garut. (a) *P. infestans* colony. (b, c) Potato plantation infected by *P. infestans* in Cikajang Garut

Table 1. Colony growth of *P. infestans* isolated from Cikajang Garut on BSEA media containing various fungicides with different concentrations

| Treatments | Concentrations (ppm) | Diameter of colony ($cm^2$) ± SD | | | Percentage of colony inhibition (%) | $EC_{50}$ (ppm) |
|---|---|---|---|---|---|---|
| | | 24 h | 48 h | 72 h | | |
| **C** | **-** | **1.73±0.21** | **4.83±0.38** | **9.00±0.00** | **0.0 a** | **-** |
| **MT X** | **2,000** | **0.60±0.00** | **0.60±0.00** | **0.60±0.00** | **93.3 e** | **759** |
| MT 1/2X | 1,000 | 0.60±0.00 | 0.60±0.00 | 0.60±0.00 | 93.3 e | |
| MT 1/4X | 500 | 0.60±0.00 | 0.60±0.00 | 0.60±0.00 | 93.3 e | |
| MT 1/8X | 250 | 0.60±0.00 | 0.60±0.00 | 0.60±0.00 | 93.3 e | |
| **MC X** | **2,000** | **0.60±0.00** | **0.60±0.00** | **0.60±0.00** | **93.3 e** | **760** |
| MC 1/2X | 1,000 | 0.60±0.00 | 0.60±0.00 | 0.60±0.00 | 93.3 e | |
| MC 1/4X | 500 | 0.60±0.00 | 0.60±0.00 | 0.60±0.00 | 93.3 e | |
| MC 1/8X | 250 | 0.60±0.00 | 0.60±0.00 | 0.77±0.15 | 91.4 e | |
| **DM X** | **1,000** | **0.60±0.00** | **1.57±0.58** | **3.10±0.10** | **65.5 cd** | **791** |
| DM 1/2X | 500 | 0.97±0.21 | 3.23±0.32 | 6.80±0.40 | 24.3 b | |
| DM 1/4X | 250 | 1.10±0.20 | 3.33±0.23 | 6.87±0.23 | 23.7 b | |
| DM 1/8X | 125 | 1.40±0.26 | 4.67±0.06 | 8.83±0.06 | 1.8 a | |
| **CL X** | **2,000** | **0.60±0.00** | **0.87±0.06** | **2.17±0.12** | **75.8 d** | **1,029** |
| CL 1/2X | 1,000 | 0.60±0.00 | 1.20±0.36 | 2.60±0.35 | 71.0 d | |
| CL 1/4X | 500 | 0.60±0.00 | 1.57±0.55 | 3.97±0.91 | 55.9 c | |
| CL 1/8X | 250 | 0.73±0.23 | 2.40±0.10 | 6.50±1.99 | 27.7 b | |
| **OX X** | **3,000** | **1.17±0.06** | **3.43±0.06** | **8.23±0.68** | **8.5 a** | **14,706** |
| OX 1/2X | 1,500 | 1.47±0.06 | 4.33±0.06 | 8.33±0.12 | 7.3 a | |
| OX 1/4X | 750 | 1.53±0.06 | 4.37±0.06 | 8.70±0.17 | 3.3 a | |
| OX 1/8X | 375 | 1.70±0.06 | 4.37±0.12 | 8.70±0.10 | 3.3 a | |

Note: C: Control; MT: Metalaxyl; MC: Mancozeb; DM: Dimetomorph; CL: Chlorothalonil; OX: Oxathiapiprolin; EC: *Effective Concentration*; Means not followed by the same letters were significantly different (P≤ 0.05) according to Duncan's test.

### The Inhibition of *P. infestans* Colony Growth in Agar Media Containing Fungicides

Colony diameter of *P. infestans* grown on media containing various fungicides with several concentrations were varied (Figure 2; Table 1). Metalaxyl and mancozeb treatments at recommended concentration of 2,000 ppm caused the highest growth inhibition of *P. infestans* at 93.3% with each $EC_{50}$ value of 759 ppm and 760 ppm respectively. Meanwhile, oxathiapiprolin caused the lowest growth inhibition result with the percentage of colony growth inhibition at 8.5% and

the $EC_{50}$ value of 14,706 ppm. The $EC_{50}$ value of oxathiapiprolin treatment was much higher than its recommended concentration. $EC_{50}$ value represents the effective concentration to suppress 50% of pathogen colony growth and oxathiapiprolin fungicide cannot suppress 50% of *P. infestans* colony growth in recommended concentration or lower. Therefore, it indicated that there had been a sensitivity reduction of *P. infestans* Cikajang Garut isolate to oxathiapiprolin.

Furthermore, the colony of *P. infestans* had grown fully on agar medium of control treatment in three days both in PDA and BSEA media. The growth rate of *P. infestans* Cikajang Garut isolate was different compared to similar studies. Under similar growing conditions, Gómez-González *et al.* (2020) mentioned that the growth of *P. infestans* isolated from Colombia in 2018 reached its maximum growth on a 9 cm petri dish within 7 to 10 days. Meanwhile, *P. infestans* isolated from several regions in Indonesia including Garut in 2019 took about 7 to 10 days to fully cover the surface of the culture medium in a 9 cm petri dish (Dangi *et al.*, 2020).

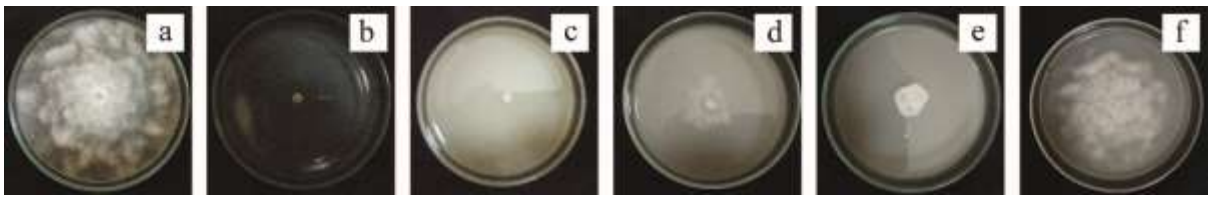


Figure 2. *P. infestans* grown on BSEA media containing fungicides. (a) BSEA media without fungicide. (b) Metalaxyl 2000 ppm. (c) Mancozeb 2,000 ppm. (d) Dimetomorph 1,000 ppm. (e) Chlorothalonil 2,000 ppm. (f) Oxathiapiprolin 3,000 ppm

As the best treatment for suppressing the colony growth of *P. infestans*, metalaxyl is a fungicide with a mode of action inhibiting nucleic acid synthesis. This fungicide is commonly used to control blight diseases caused by *Pythium* sp., *Phytophthora* sp., and other oomycetes (Urech *et al.*, 1977). Sajid & Shah (2018) stated that metalaxyl fungicide is still effective for controlling potato leaf blight with a disease suppression rate of 93.34%. Furthermore, mancozeb is a contact fungicide categorized as a wide-spectrum or multi-site fungicide, thus the possibility of resistance mechanism in pathogen due to the use of mancozeb is relatively low (Gullino *et al.*, 2010). Mancozeb fungicide has a good effect in suppressing *Phytophthora* blight disease thet reduced 77.99% of infection on 80% active ingredient and 4 g/l concentration (Fitria & Rachmawati, 2019).

Chlorothalonil is used as an alternative fungicide to control *P. infestans* because it has a broad spectrum with a mode of action that interferes with the enzymatic process of fungi, so fungi will not be able to perform essential functions related to their growth (Yamamoto *et al.*, 2009). Several necessary enzymes in cellular respiration and metabolic processes involving large molecules will break apart and the energy will not be available (Cox, 1997). Dimetomorph is also an alternative fungicide to control *P. infestans*. This fungicide is a systemic fungicide that functioned as a protectant, curative, and antisporulant to oomycetes, especially the genus of Phytophthora such as *Phytophthora infestans* (Cohen *et al.*, 1995). Dimetomorph has a mode of action inhibiting sterol synthesis. Sterol is a vital substance for the structure and function of pathogen cell membranes (Hudayya & Jayanti, 2013).

Oxathiapiprolin is a systemic fungicide made specifically to control oomycetes (Cohen *et al.*, 2018; Pasteris *et al.*, 2015). However, this study demonstrated that oxathiapiprolin could not inhibit the in vitro growth of *P. infestans* as shown by the lowest percentage of colony growth inhibition of *P. infestans* Cikajang Garut isolate. The result is contradicted to the study by Evenhuis *et al.* (2019) that oxathiapiprolin was a fungicide with the best efficacy rate in controlling *P. infestans*, which was 4.9/5 or 98%.

A similar mode of action of oxathiapiprolin and dimetomorph that inhibits sterol synthesis might contribute to the reduction of oxathiapiprolin ability to inhibit the growth of *P. infestans*. The simultaneous use of similar mode of action fungicides might lead to the emergence of resistant pathogen population despite being rotated. This is possible as the pathogens can evolve against the fungicide stress to preserve their existence.

**The Inhibition of Necrotic Lesion on Potato Leaves**

The necrotic lesion appeared within 24 hrs at 18°C on all potato leaves treated with fungicides and control except for leaves treated with metalaxyl (Figure 3). Up to 120 hrs incubation, no necrotic symptom found on those leaves. Prabawa & Damanhuri (2018) stated that disease symptoms faster when crops are applied with pesticides. It might due to either plants being susceptible to the pathogen infection or an indication of the fungicide efficacy reduction due to a resistant pathogen population.

The highest necrotic lesion of 2.46 $cm^2$ was found on potato leaves treated with oxathiapiprolin, whereas none was found on leaves treated with metalaxyl (Table 2). It indicates that *P. infestans* Cikajang Garut isolate is still sensitive to metalaxyl fungicides.

Table 2. Necrotic lesion area on potato leaves treated with several types of fungicide and inoculated with *P. infestans*

| Concentrations | Latent Period | | | | | Necrotic Lesions [Average ($cm^2$) ± SD)] | | |
|---|---|---|---|---|---|---|---|---|
| | 24 h | 48 h | 72 h | 96 h | 120 h | 72 h | 96 h | 120 h |
| C | ✔ | - | - | - | - | 1,21±0,20 | 2,64±0,32 | **4,81±0,66 d** |
| MT X | - | - | - | - | - | **0,00±0,00** | **0,00±0,00** | **0,00±0,00 a** |
| MT 1/2X | - | - | - | - | - | 0,00±0,00 | 0,00±0,00 | 0,00±0,00 a |
| MT 1/4X | - | - | - | - | - | 0,00±0,00 | 0,00±0,00 | 0,00±0,00 a |
| MT 1/8X | - | - | - | - | - | 0,00±0,00 | 0,00±0,00 | 0,00±0,00 a |
| MC X | ✔ | - | - | - | - | 0,62±0,15 | 1,18±0,32 | **1,40±0,22 b** |
| MC 1/2X | ✔ | - | - | - | - | 0,69±0,22 | 1,35±0,57 | 1,60±0,48 b |
| MC 1/4X | ✔ | - | - | - | - | 0,97±0,35 | 1,67±0,58 | 1,81±0,71 b |
| MC 1/8X | ✔ | - | - | - | - | 0,99±0,57 | 2,10±0,67 | 2,75±0,90 c |
| DM X | ✔ | - | - | - | - | 0,38±0,21 | 1,25±0,19 | **2,18±0,32 b** |
| DM 1/2X | ✔ | - | - | - | - | 0,60±0,40 | 1,45±0,89 | 2,39±0,84 c |
| DM 1/4X | ✔ | - | - | - | - | 0,68±0,12 | 1,77±0,12 | 3,25±0,10 c |
| DM 1/8X | ✔ | - | - | - | - | 1,06±0,42 | 2,27±0,59 | 3,76±0,22 c |
| CL X | ✔ | - | - | - | - | 0,62±0,40 | 1,17±0,69 | **1,97±0,66 b** |
| CL 1/2X | ✔ | - | - | - | - | 0,68±0,09 | 1,63±0,76 | 2,00±0,77 c |
| CL 1/4X | ✔ | - | - | - | - | 0,82±0,12 | 1,88±0,37 | 3,09±0,63 c |
| CL 1/8X | ✔ | - | - | - | - | 0,82±0,28 | 2,18±0,12 | 3,43±0,79 c |
| OX X | ✔ | - | - | - | - | 0,53±0,40 | 1,39±0,61 | **2,46±0,92 c** |
| OX 1/2X | ✔ | - | - | - | - | 0,64±0,12 | 1,91±0,55 | 3,48±0,43 c |
| OX 1/4X | ✔ | - | - | - | - | 0,81±0,12 | 2,37±0,56 | 3,52±0,34 c |
| OX 1/8X | ✔ | - | - | - | - | 0,95±0,15 | 2,63±0,53 | 4,52±0,98 c |

Note: C: Control; MT: Metalaxyl; MC: Mancozeb; DM: Dimetomorph; CL: Chlorothalonil; OX: Oxathiapiprolin; Means not followed by the same letters were significantly different according to Scott Knott's test.

Matson *et al*. (2015) stated that metalaxyl fungicide showed high-moderate effectiveness in suppressing the growth of *P. infestans* both in the artificial medium and in plants as shown in this study. Meanwhile, oxathiapiprolin fungicide treatment showed the highest necrotic lesion area compared to other fungicide treatments regardless of the use of the fungicide to specifically control oomycetes and reported to have an efficacy value of 98% to *P. infestans* (Cohen *et al*., 2018; Evenhuis *et al*., 2019). This study showed that there had been a reduced sensitivity of *P. infestans* to oxathiapiprolin fungicide.

Pathogen resistance to fungicides indicates the presence of adaptive pathogen to the applied fungicide. Pathogen resistance is influenced by many factors such as the mode of action of a fungicide, the cycle of pathogens, the way fungicides are applied, and the planting system (Damicone, 2008). Pathogens genetically regulate the process of resistance to lower their sensitivity to the given type of fungicide. The emergence of resistant strains in a pathogen population can affect the process of pathogen selection in a population (Damicone, 2008). The selection process affects the increased population of resistant pathogen strains due to genetic drift (McDonald & Linde, 2002). Resistant

pathogens will continue to increase when the similar fungicides mode of action are used continuously for a long time and eventually the entire population of pathogens becomes resistant (Deising *et al.,* 2008).

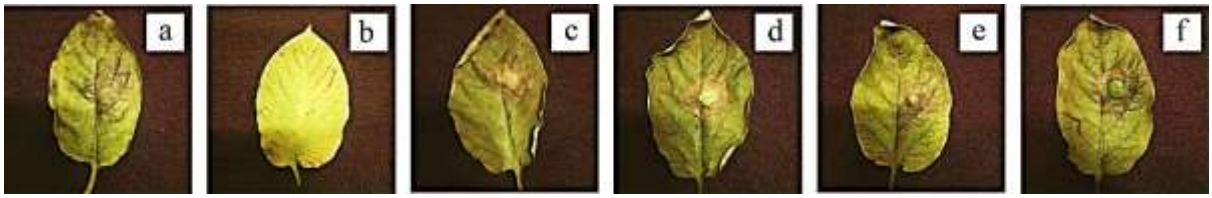


Figure 3. Necrotic lesion on potato leaves treated with various fungicide. (a) Control treatment. (b) Metalaxyl 2000 ppm. (c) Mancozeb 2,000 ppm. (d) Dimetomorph 1,000 ppm. (e) Chlorothalonil 2,000 ppm. (f) Oxathiapiprolin 3,000 ppm

## CONCLUSIONS

*Phytophthora infestans* Cikajang Garut isolate was still sensitive to metalaxyl and mancozeb. Meanwhile, *P. infestans* was moderate-sensitive to dimetomorph and chlorothalonil. However, there were indications of sensitivity reduction of *P. infestans* to oxathiapiprolin.

## REFRENCES

Brent, KJ and DW Hollomon. 2007. Fungicide Resistance in Crop Pathogens: How Can It Be Managed? 2nd Edition. Fungicide Resistance Action Committee. Belgium.

Byamukama, E, D Yabwalo, S Ali, C Tande, C Strunk, R Hopkins, N Braun, and F Mathew. 2019. Fungicide Resistance: Risk and Management. Journal of Agronomy. 1(1): 1–4.

Cohen, Y, A Baider, and BH Cohen. 1995. Dimetomorph activity against oomycete fungal plant pathogens. Phytopathology. 85(12): 1500–1506.

Cohen, Y, AE Rubin, and M Galperin. 2018. Oxathiapiprolin-based fungicides provide enhanced control of tomato late blight induced by mefenoxam-insensitive *Phytophthora infestans*. Plos One. 13(9): 1–22.

Cox, C. 1997. Chlorothalonil. Journal of Pesticide Reform. 17(4): 14–20.

Damicone, J. 2008. Fungicide Resistance Management. Division of Agricultural Sciences and Natural Resources. Oklahoma State University.

Dangi, S, P Wharton, AD Ambarwati, TJ Santoso, Kusmana, I Sulastrini, J Medendorp, K Hokanson, and DS Douches. 2020. Genotypic and phenotypic characterization of *Phytophthora infestans* populations on Java, Indonesia. Plant Pathology. 70: 61–73.

Deising, HB, S Reimann, and SF Pascholati. 2008. Mechanisms and significance of fungicide resistance. Brazillian Journal of Microbiology. 39(2): 286–295.

Dhingra, OD, and JB Sinclair. 1985. Basic Plant Pathology Methods. CRC Press. Inc. Florida.

Evenhuis, A, R Bain, BJ Nielsen, W van den Berg, and HTAM Schepers. 2019. Fungicide Evaluation to Rate the Efficacy to Control Tuber Blight. Wageningen Applied Plant Research. Wageningen.

Fitria, RU, dan D Rachmawati. 2019. Keefektifan fungisida bahan aktif mankozeb untuk mengendalikan hawar daun kentang (*Phytopthora infestans*). Agrika. 13(2): 90–100.

Gómez-González, S, D Castañeda-Sánchez, and J Morales-Osorio. 2020. Media preferences, micro-morphometric analysis, and cardinal growth temperature determination for *Phytophthora infestans* Sensu Lato isolated from different hosts in Colombia. Brazilian Journal of Biology. 80(1): 167–179.

Goth, RW, and JA Keane. 1997. A detached-leaf method to evaluate late blight resistance in potato and tomato. American Potato Journal. 74(5): 347–352.

Gullino, ML, F Tinivella, A Garibaldi, GM Kemmitt, L Bacci, and B Sheppard. 2010. Mancozeb: Past, present, and future. Plant Disease. 94(9): 1077–1087.

Hudayya, A, and H Jayanti. 2012. Pengelompokan Pestisida Berdasarkan Cara Kerjanya (Mode of Action). Balai Penelitian Tanaman Sayuran, Pusat Penelitian dan Pengembangan

Hortikultura, Badan Penelitian dan Pengembangan Pertanian, Kementerian Pertanian Republik Indonesia.

Matson, MEH, IM Small, WE Fry, and HS Judelson. 2015. Metalaxyl resistance in *Phytophthora infestans*: assessing role of RPA190 gene and diversity within clonal lineages. Phytopathology. 105(12): 1594–1600.

McDonald, BA, and C Linde. 2002. Pathogen population genetics, evolutionary potential, and durable resistance. Annual Review of Phytopathology. 40(1): 349–379.

McKinney, HH. 1923. A new system of grading plant diseases. Journal of Agricultural Research. 26: 195–218.

Pasteris, RJ, MA Hanagan, JJ Bisaha, BL Finkelstein, LE Hoffman, V Gregory, JL Andreassi, JA Sweigard, BA Klyashchitsky, YT Henry, and RA Berger. 2015. Discovery of oxathiapiprolin, a new oomycete fungicide that targets an oxysterol binding protein. Bioorganic and Medicinal Chemistry. 24(3): 354–361.

Pobedinskaya, MA, SN Elansky, NV Statsyuk, and MP Plyakhnevich. 2012. Fungicide resistance of Russian *Phytophthora infestans* strains. EuroBlight. 15: 243–248.

Prabawa, P, dan Damanhuri. 2018. Evaluasi Ketahanan genotip padi beras merah (*Oryza sativa* L.) terhadap penyakit blas daun (*Pyricularia oryzae* Cav.) Ras 173. Agro Bali (Agricultural Journal). 1(2): 82–87.

Purwanti, H. 2002. Penyakit hawar daun (*Phytophthora infestans* (Mont.) de Bary) pada kentang dan tomat: Identifikasi permasalahan di Indonesia. Jurnal Agrobiogen. 5(2): 67–72.

Rosmayati, GA Wattimena, MS Sinaga, dan M Machmud. 2006. Identifikasi ras fisiologi *Phytophthora infestans* pada sentra produksi kentang di Kabupaten Karo. Jurnal Ilmiah Pertanian KULTURA. 41(1): 9–18.

Sajid, MN, and S Shah. 2018. Effectiveness of some fungicides against late blight of potato caused by *Phytophthora infestans*. Plant Protection. 2(1): 31–34.

Sumardiyono, C. 2008. Ketahanan jamur terhadap fungisida di Indonesia. Jurnal Perlindungan Tanaman Indonesia. 14(1): 1–5.

Syahputra, TS. 2018. Evaluasi Efektivitas Fungisida Berbahan Aktif Mankozeb terhadap *Phytophthora infestans* Penyebab Penyakit Hawar Daun Tomat di Dataran Tinggi Karo. [Skripsi] Universitas Sumatera Utara. Medan.

Tumwine, J, HD Frinking, and MJ Jeger. 2000. Isolation techniques and cultural media for *Phytophthora infestans* from tomatoes. Mycologist. 14(3): 137–139.

Urech, PA, F Schwinn, and F Staub. 1977. CGA 48988, a novel fungicide for the control of late blight, downy mildew, and related soil borne disease. In Proceedings of the British Crop Protection Conference - Pests and Diseases Vol 2, pp 671-678. Brighton, Nottingham.

Vleeshouwers, VGAA, WV Dooijeweert, LCP Keizer, L Sijpkes, F Govers, and LT Colon. 1999. A laboratory assay for *Phytophthora Infestans* resistance in various solanum species reflects the field situation. European Journal of Plant Pathology. 105(3): 241–250.

Wismaningsih, ER, and DI Oktaviasari. 2016. Identifikasi jenis pestisida dan penggunaan APD pada petani penyemprot di Kecamatan Ngantru Kabupaten Tulungagung. Jurnal Wiyata. 3(1): 100–105.

Yamamoto, A, I Miyamoto, M Kitagawa, H Moriwaki, H Miyakoda, H Kawasaki, and R Arakawa. 2009. Analysis of chlorothalonil by Liquid Chromatography/Mass Spectrometry using negative-ion atmospheric pressure photoionization. Analytical Sciences. 25(5): 693–697.

Yuan, SK, XL Liu, NG Si, J Dong, BG Gu, and H Jiang. 2006. Sensitivity of *Phytophthora infestans* to flumorph: In vitro determination of baseline sensitivity and the risk of resistance. Plant Patology. 55(2): 258–263.

Yusuf, AM. 2018. Analisis Resistensi *Phytophthora infestans* (Patogen Hawar Daun Kentang) terhadap Fungisida di Kecamatan Kayu Aro Barat Kabupaten Kerinci Provinsi Jambi. [Skripsi]. Universitas Mercu Buana. Yogyakarta.