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Endemic Bacteria Potential for Decomposing Pesticide *Prophenophos* and *Chlorantraniliprole* from Pesticide-Exposed Soybean Field in East Java

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Abstract— Pest control in soybean field in general still uses synthetic pesticide. Soil bacteria from pesticide-exposed field are potentially able to decompose pesticide residue in during certain period of time. This research aimed to find out the potency of isolated endemic bacteria from soybean field in Jombang, Lamongan, and Probolinggo in East Java for decomposing pesticide prophenophos and chlorantraniliprole. This research used observation method. Bacteria sample was obtained from soybeans fields exposed by pesticide in nutrient agar medium with addition prophenophos and chlorantraniliprole using pour plate method. Testing medium used was nutrient agar with added prophenophos (1 g/l) and chlorantraniliprole (2 g/l), in which bacteria was incubated for 1 day. The area of the clear zone was determined by subtraction of clear zone diameter with well diffusion diameter. Results showed that endemic soil bacteria isolates grown in medium with addition of prophenophos showed clear zone were amounted to 15 (Jombang), 19 (Lamongan), and 16 (Probolinggo), while in chlorantraniliprole-added medium were 21 (Jombang), 23 (Probolinggo), and 25 (Lamongan) isolates. The total number of endemic bacteria isolates obtained from the soybean fields in those 3 areas able to decompose prophenophos was 50 isolates, while chlorantraniliprole was 69 isolates. Further test of pesticide-decomposing potency found that 18 isolates able to decompose prophenophos with clear zone diameter of 1.03 – 3.15 cm, 13 isolates were able to decompose chlorantraniliprole with clear zone diameter of 0.2 – 3.1 cm, while 5 isolates were able to decompose both pesticides with clear zone of 1.25 – 4.0 cm.

Keywords— endemic bacteria, pesticide decomposer, prophenophos, chlorantraniliprole, East Java

I. INTRODUCTION

Pest control in crops agroecosystem generally still uses synthetic pesticide [1] Unmanaged insecticide application can affect soil physical and chemical structure [2], in addition to microbe diversity. Two groups of widely used pesticides by farmer are organophosphate and amide. Prophenophos is an organophosphate with gastric-contact toxicity, has wide spectrum, and can react quickly in controlling various pest attack [3] [4]. This pesticide toxin also affects nerve system, inhibits enzyme acetylcholinesterase, induces respiration failure, and blocks the heart [5]. Meanwhile, chlorantraniliprole is included in diamide group that can

affect nerves and muscles, induce paralysis in Coleoptera, Diptera, and Isoptera pest insect, also in Lepidoptera, such as *Spodoptera exigua* [6]. Insects exposed to this pesticide have their appetite highly lowered; body contracted, inactivity, and finally died. This insecticide was found to left residue in cabbage post-harvest up to 0.698 mg/kg and after being soaked in tap water up to 0.526 mg/kg [7]

The application of both insecticides can cause decrease of soil physical and chemical quality. Based on previous study, pesticide-applied land in East Java (Jombang, Lamongan, and Probolinggo) had chemically very low C-organic level (0.58-0.94%), low N-total (0.10-0.15%), and low C/N ratio (6-9) beside low content of organic materials and varying P level from low (11.90 mgkg⁻¹ – 11.94 mgkg⁻¹), mid, up to very high (36.82-38.48 mgkg⁻¹) [2]. Based on soil physical analysis, it was found that these lands had soil porosity approaching ideal level at 46.08-49.30%, water content was above land capacity at 0.34-0.45 pF, water situation was available, while soil samples were categorized as clay, clay loam, and silty clay.

Lowering land condition due to pesticide application can be improved using endemic bacteria isolated from local lands. Several farming lands in East Java has already used pesticide which can be easily decomposed by microbe. In this area, pesticide-decomposing microbes can be found easily. These endemic bacteria can be isolated and subcultured in laboratory to be then introduced to farming land exposed to synthetic pesticide to minimalize or eliminate pesticide residue. Based on previous study, no pesticide residue of aceptate, chlorantraniliprole, dimehypo, phenobucarb, methomyl, prophenophos, chlorothalonil, and Chlorpyrifos was found in field in Jombang. and Lamongan. Meanwhile in Probolinggo, chlorantraniliproleresidue was found at 0.025 mg/kg [2]. Based on those pesticide-applied soil conditions, endemic microbes found in the three area of East Java were isolated and tested against synthetic pesticide prophenophos and chlorantraniliprole.

II. METHODS

Methods used in the current study were observation. Bacteria sample was taken from soybean-planted lands

TABLE 1. CLEAR ZONE OF PESTICIDE-DECOMPOSING BACTERIA FOUND IN PESTICIDE-EXPOSED SOYBEAN FIELD IN JOMBANG, LAMONGAN, AND PROBOLINGGO

Field location	Number of decomposing bacteria		Testing sample number	Isolate code	clear zone diameter (cm)	
	Propheno-phos	Chlorantraniliprole			Propheno-phos	Chlorantraniliprole
Lamongan	15	23	1	L1*/10 ⁴ A3	2.25	-
			2	L1/10 ⁴ A4	-	3.1
			3	L1/10 ⁵ A2	2.2	2.4
			4	L1*/10 ⁵ A3	-	2.3
			6	L2*/10 ³ A2	-	1.9
			7	L2/10 ³ A2	-	0.1
			8	L2*/10 ⁴ A1	3.9	4.0
			9	L3/10 ³ A2	2.0	-
			10	L3*/10 ⁴ A1	-	2.0
			Jombang	19	21	11
12	J1*/10 ⁴ A3	2.35				2.0
13	J1/10 ⁵ A1	-				3.0
14	J2*/10 ⁵ A2	3.15				3.1
15	J2/10 ⁴ A1	2.25				-
16	J3/10 ⁴ A2	2.65				-
17	J3*/10 ⁵ A1	-				-
Probolinggo	16	25	18	P1*/10 ⁴ A1	2.0	-
			19	P1*/10 ⁴ A2	3.0	-
			20	P1*/10 ⁴ A3	1.05	-
			21	P2/10 ⁴ A1	3.0	-
			22	P3/10 ⁴ A2	2.55	-
			23	P3*/10 ⁵ A2	3.0	0.2
			24	P3*/10 ⁵ A3	2.75	1.25
			25	P3*/10 ⁵ A4	2.75	-
			26	P3/10 ⁵ A4	2.50	2.7
Total	50	69			18	13

located in Jombang, Lamongan, and Probolinggo, East Java which had been applied pesticide on 3 points; water entry, middle, and water exit points. Bacteria were isolated from soil sample. As much as 1 g soil sample was homogenized into 9 ml distilled water using shaker and then diluted. Next, 1 ml of dissolved soil sample was grown on nutrient agar medium (Merck) with additional pesticide 1 g/l prophenophos and 2 g/l chlorantraniliprole using pour plate method. Grown bacteria showing clear zone surrounding colonies were isolated in slant nutrient agar medium. These isolates were then tested their ability in decomposing prophenophos and/or chlorantraniliprole separately using well diffusion method in nutrient agar medium with added 1 g/L prophenophos and 2 g/l chlorantraniliprole. Bacteria were incubated for 1 day. Clear zone diameter was measured using ruler at precision of 1 mm. Inhibition zone was determined by subtracting well diameter from clear zone diameter.

III. RESULT AND DISCUSSION

Isolation result from soil sample of 3 areas in East Java (Jombang, Lamongan, and Probolinggo) found bacteria which could survive on medium with added chlorantraniliprole from Jombang soil were amounted to 21

isolates, from Probolinggo soil 23 isolates, and from Lamongan soil 25 isolates. In the other hand, bacteria

survived on medium with added prophenophos from Jombang soil was amounted to 19 isolates, from Lamongan soil was 15 isolates, and from Probolinggo soil was 16 isolat (Table 1). Bacteria colonies survived on the medium were too dense, thus clear zone observed was overlapped. As much as 18 bacteria isolates survived on prophenophos medium and 13 isolates on chlorantraniliprole medium were further tested their ability in decomposing both chemicals by using well-diffusion method. Resulting clear zone were presented in Table 1.

Out of 119 bacteria isolated from soybean field in Jombang, Lamongan, and Probolinggo previously being applied pesticide prophenophos and/or chlorantraniliprole, 31 of them were able to decompose pesticide prophenophos and/or chlorantraniliprole. As much as 18 isolates were able to decompose prophenophos, 13 isolates were able to decompose chlorantraniliprole, and 5 of them were able to decompose both chemicals, indicated by producing clear zone around colonies. Based on [8], 7 bacteria isolated from farming field in Baturiti, Tabanan, Bali were tolerant to pesticide with Chlorantraniliprole active compound. In the other hand, [3] [4] found 7 pesticide bacteria which were able to degrade persistent organic pollutants (POPs) insecticide

residue, including 5 isolates that degraded lindan, heptachlor, DDT, and dieldrin, and 2 isolates that degraded aldrin. Bacteria have density of $10^6 - 10^9$ cells/g in soil and commonly abundant in surface of clay soil at water content 50-75%, pH neutral, and temperature 25-35°C [9]. This is in accord with water content of soil sample used in the current study which had clay structure and water content above field capacity at 0.34-0.45 pF [2], thus suitable condition for bacteria was fulfilled and numerous isolates could be cultured (119 isolates). *Pseudomonas putida* could be isolated in medium containing organophosphate insecticide (including prophenophos) [1]. In addition, from soil exposed to insecticide prophenophos, *Bacillus* sp., *Pseudomonas* sp., *Enterobacter* sp., *Citrobacter* sp., *Azotobacter* sp., and *Azospirillum* sp. could be isolated [11].

Based on table 1, as much as 18 isolates were able to degrade prophenophos at 24 hours incubation time with resulting clear zone of 1.05 – 3.15 cm, 13 isolates were able to decompose chlorantraniliprole with clear zone of 0.2 – 3.1 cm, while 5 isolates were able to degrade both chemicals with clear zone of 1.25–4.0 cm. Clear zone indicated that bacteria was able to degrade prophenophos and/or chlorantraniliprole after 24 hours incubation with degradation area of 0.2 – 4.0 cm around colonies. *Oceanobacillus iheyis*, *Exiguobacterium* *7ofundus*, and *Bacillus formis* isolated from Rawa Pening were able to degrade malathion and prophenophos at 192 hours [12].

IV. CONCLUSION

Endemic bacteria found in soybean field in Jombang, Lamongan, and Probolinggo, East Java potentially able to decompose pesticide prophenophos, chlorantraniliprole, or both of them. Bacteria able to decompose prophenophos were 18 isolates with clear zone of 1.03 – 3.9 cm, 13 isolates were able to decompose chlorantraniliprole with clear zone of 0.2 – 4.0 cm, and 5 isolates were able to decompose both pesticides with clear zone of 1.25 - 4.0cm

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