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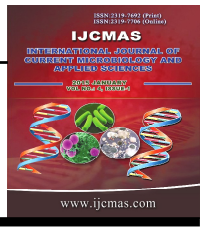

Original Research Article

The ability phosphate solubilization of bacteria rhizosphere of potato Var. Hartapel from Buru Island

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Keywords

Potato var. Hartapel, Buru Island, Phosphate solubilization, Rhizosphere, Plant growth promoter

A B S T R A C T

The ability of microorganisms to solubilize phosphates is detected by production of halo around colonies on media containing insoluble mineral phosphate. The halos formed around the colonies are as a result of pH drop produced by the release of organic acids, which are responsible for phosphate solubilization. The aim of this study was to isolate bacteria of rhizosphere origin of potato cv. Hartapel which has the ability to phosphate solubilization. In this study, the isolate that showed the best capability in solubilizing phosphate was isolate HB3 (14.2376 mg l-1) and isolate HB18 also was able to solubilize phosphate but resulted in lower concentration of soluble phosphate (4.457 mg l -1). Bacteria rhizosphere phosphate solubilization as agents of plant growth promoters, which along its use as inoculants can increase phosphate uptake by plants.

Introduction

Potatos is the third most common food crop in the word, with a high yield per hectare; however, the potato has been relatively sensitive to yield losess resulting from soil salinity, drought, and low nutrient availability (van der Linden, 2011). Improved soil fertility is one of the most important strategies to increase agricultural production. Nitrogen fixation is essential in improving soil fertility. In addition to nitrogen fixation, phosphate dissolving as important for plants by microorganisms. Phosphate is essential macronutrients for plant growth and development. Microorganisms are biological rescue system capable of dissolving inorganic phosphate soil and make it available to plants. The ability of some microorganisms to convert the phosphate solution to form available, such as orthophosphate, is an important property of microorganisms solvent phosphate in increasing crop yields (Rodriguez et al., 2006; Chen et al., 2006).

Phosphate rhizosphere bacteria utilize a source of plant growth to spur the agent. The

use of phosphate dissolving bacteria as inoculants can improve phosphate uptake by plants (Chen et al., 2006; Igual et al., 2001). Recent developments in the understanding of the functional diversity, ability rhizosphere mengkoni, application mode tends to facilitate the use of microorganisms as a reliable component in the management of sustainable agricultural systems (Zaidi et al., 2009a).

Because bacteria are most abundant in the rhizosphere microorganisms, very likely affect the physiology of plants, especially on the competitiveness of bacteria in root colonization (Barriuso et al., 2008). Bacteria phosphate solubilization plays help provide nutrients for plants because it can alter phosphate forms that are not available to the plants into a form that is available. One alternative to improve the efficiency of phosphate fertilizer is to utilize a group of bacteria that can dissolve phosphate, so it can be absorbed by plants. The purpose of this study is to get the plant rhizosphere bacterial isolates origin of potato cv. Hartapel which has the ability to dissolve phosphate.

Materials and Methods

Source of bacteria

Bacteria were isolated from the rhizosphere soil samples Hartapel varieties of potato plants that grow on the altitude of 700 m above sea level in Leksula, Buru South (Buru Island) Maluku, Indonesia. In each sampling point, one sample consisted of rhizosphere soil (soil around the root zone) plants. Soil samples have been taken at a depth of 0-20 cm in the four quadrants stands Hartapel potato varieties then were combined.

Isolation of rhizosphere soil samples of potato

Isolation of rhizosphere bacteria carried by serial dilution method. Ten grams of rhizosphere soil was weighed and dissolved in 90 ml of sterile water, then shaked for 30 minutes. One ml of rhizosphere soil suspension was added to a test tube containing 9 ml of sterile water to obtain a suspension with a 10^{-2} dilution level. Dilution was done so in the same manner until a 10^{-8} suspension. Subsequently 0.1 ml of the suspension was grown on NA medium in a petri dish. NA medium which already contains rhizosphere bacteria were incubated for 24 hours at room temperature. Every single colonies were grown to reisolated and made as pure culture.

Phosphate Solubilization Test

Solubilization of phosphate was tested following the method of described by Pikovskaya (Sundara and Shinha 1962; Subba Rao 1982). Suspension 24-hour-old bacterial isolates grown on solid media containing tricalcium phosphate Phikovkaya (Ca_3PO_4) with dispersive method. The formation of transparent halos around each bacterial colonies showed solubilization activity. The resulting halos zone around the colonies after incubation for 3 days showed the presence of bacterial activity in solubilization phosphate. The ability of phosphate dissolving bacteria isolates characterized by halo zone, dissolution efficiency phosphate and phosphate leaching index. Measurement of the efficiency of dissolving phosphate (EP) is done by using the formula :

$$EP = \frac{\text{Diameter of halo zone}}{\text{Diameter colony}} \quad X\ 100$$

Against dissolution index phosphate (IP) was measured by using the formula:

$$IP = \frac{\text{Diameter of halo zone - Diamater of colony}}{\text{Diameter of colony}}$$

Result and Discussion

From rhizosphere soil samples of varities Hartapel from South Buru at an altitude of 700 m above sea level as much as 70 bacterial isolates were obtained. Of these isolates, 36 isolates were capable of phosphate solubilization. Phosphate solubilization ability is marked by the formation of transparent halos around the colony bacteria in media containing Phikovskaya tricalcium phosphate (Ca3PO4) (Figure 1). Of the 70 isolates tested encountered a total of 36 (51.43 %) isolates were capable of dissolving phosphate contained in Phikovskaya media. The isolates were different concentration of dissolving phosphate. The isolate that showed the best capability in solubilization phosphate was isolate HB3 (14.237 mg l^{-1}) with 2.08 dilution index and 307.69 dissolution efficiency at pH 5.32. While the lowest solubilization phosphate was isolate HB18 (4.457 mg l^{-1}) with 0.54 dilution index and 153.85 dissolution efficiency at pH 4.67 (Table 1). Generally phosphate solubilization occurs at pH acidic or under 6. There is a tendency that the pH of the dissolution rates of phosphate sour varies or differs from each other in value of the isolates tested. Isolates can potentially superior dindikasikan with a high concentration of phosphate solubilization. From the test results in a row is encountered isolates HB3, HB8, HB34, and HB32.

The diameter of halo zone, index and efficiency dissolution referred as indicator phosphate dissolution. The diameter of halo zone, index dissolving phosphate and phosphate leaching efficiency is highly dependent on the growth of the colony. Colony growth rate the stronger the higher the concentration of phosphate and generally dissolution occurs at the level of liquid sour acidity Pikovskaya media (pH 4:34 - 5.71).

Phosphate is the most important key elements in plant nutrients other than nitrogen, since it plays an important role in almost all the major metabolic processes in plants, including photosynthesis, energy transfer, signal transduction, macro-molecule biosynthesis and respiration (Khan et al., 2010). Although phosphate is abundant in the soil in the form of organic and inorganic compounds, phosphate is a major factor that limits the growth of plants as a form not available for root uptake. Some researchers reported the ability of various species of bacteria to dissolve inorganic phosphate, such as tricalcium phosphate, dicalcium phosphate, hydroxyapatite, and rock phosphate (Park et al., 2009).

Decrease pH of the medium showed the release of organic acids by Microorganisms solvent phosphate (Whitelaw, 2000; Maliha et al., 2004) through direct oxidation that occurs on the outside of the cytoplasmic membrane (Zaidi et al., 2009b). Organic acids are products of microbial metabolism, mostly by oxidative respiration or fermentation of organic carbon source (Atlas and Bartha 1997; Trolove et al., 2003; Omar, 1998). Synthesis and release of organic acids from microorganisms phosphate solvent acidify the cell into the surrounding environment and the environment, which in turn causes the release of mineral ions P by substitution of H + cations bound to phosphate (Park et al., 2009; Goldstein,

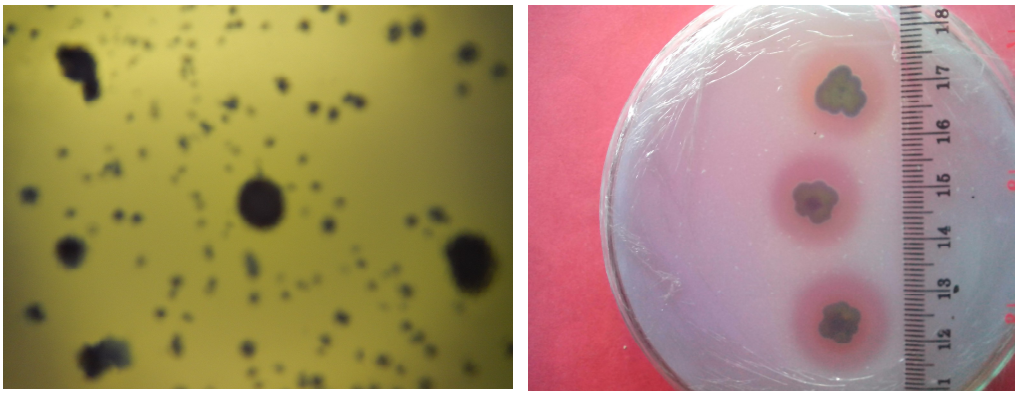
1994; Jorquera et al., 2008). The resulting acid phosphate solubilization microorganisms solvent soluble P is gluconic acid (Di-Simine et al., 1998; Bar-Yosef et al., 1999), oxalic acid, citric acid (Kim et al., 1997), lactic acid, tartaric acid, aspartic acid (Venkateswarlu et al., 1984). However, acidification does not seem to be the only mechanism of solubility, as the ability to reduce the pH in some cases did not correlate with the ability to dissolve minerals P (Subba Rao, 1982).

The ability of phosphate solubilization is marked by the formation of transparent halos around the colony bacteria. The best capability in solubilizing phosphate was isolate HB3 (14.2376 mg l^{-1}) and isolate HB18 also was able to solubilize phosphate but resulted in lower concentration of soluble phosphate (4.457 mg l^{-1}).

Table.1 The Ability of phosphate solubilization isolates

Code Isolates	Diameter Colony (cm)	Diameter Halo Zone (cm)	Index Pelarutan (IP)	Eficiency Pelarutan (EP)	pH	Contration Phosphate Solubilization (mg P l^{-1})
HB1	0.65	0.95	0.46	146.15	5.46	12.433
HB3	0.65	2.00	2.08	307.69	5.32	14.237
HB4	0.75	2.00	1.67	266.67	5.25	10.498
HB5	0.70	0.80	0.14	114.29	5.39	7.494
HB6	0.75	1.05	0.40	140.00	5.24	12.132
HB7	1.20	1.25	0.04	104.17	5.26	11.564
HB8	0.90	1.50	0.17	116.67	5.45	13.210
HB9	0.75	0.85	0.13	113.33	5.22	12.492
HB10	0.65	0.95	0.46	146.15	5.23	11.685
HB11	0.70	1.15	0.64	164.29	5.31	12.515
HB12	0.60	1.00	0.67	166.67	5.20	11.830
HB15	0.50	0.90	0.80	180.00	5.71	10.889
HB16	0.50	0.80	0.60	160.00	5.09	11.857
HB17	0.75	1.05	0.40	140.00	5.27	12.662
HB18	0.65	1.00	0.54	153.85	4.67	4.457
HB19	0.80	0.95	0.19	118.75	5.29	11.742
HB20	0.70	1.20	0.71	171.43	5.31	12.302
HB21	0.80	1.05	0.31	131.25	5.16	12.641
HB22	0.70	1.00	0.43	142.86	5.58	11.076
HB23	0.50	0.65	0.30	130.00	5.72	11.873
HB24	1.20	1.50	0.25	125.00	5.11	12.684
HB27	0.70	1.00	0.43	142.86	5.06	11.375
HB28	0.30	0.50	0.67	166.67	4.72	4.882
HB29	0.80	1.40	0.75	175.00	5.54	11.238
HB30	0.50	1.00	1.00	200.00	5.31	10.935
HB31	0.60	0.90	0.50	150.00	5.19	12.372
HB32	0.45	0.80	0.78	177.78	5.21	12.911
HB33	0.65	0.95	0.46	146.15	4.34	12.175
HB34	0.45	0.70	0.56	155.56	5.23	12.973
HB35	0.50	0.75	0.50	150.00	5.40	12.635
HB36	1.00	1.20	0.20	120.00	5.37	12.417
HB37	0.65	0.95	0.46	146.15	5.47	11.238
HB38	0.75	0.95	0.27	126.67	5.20	5.102
HB39	0.50	0.70	0.40	140.00	5.20	8.691
HB40	0.50	0.70	0.40	140.00	5.16	9.289
HB42	1.00	1.50	0.50	150.00	5.33	11.904

Figure.1 Phosphate solubilization test : (a) isolate non forming halo zone (b) isolate HB3 forming of transparent halos around the colony



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