UNIVERSITI TEKNOLOGI MARA

ISOLATION, CHARACTERIZATION AND EFFECTS OF ZINC-SOLUBILIZING BACTERIA ON GROWTH PROMOTION AND NUTRIENT CONTENTS IN RICE

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ABSTRACT

Rice is one of the major crops after oil palm and rubber in Malaysia and more familiar in peninsular where lowland areas has become the focal point for cultivation activities. However, zinc deficiencies have become an issue and widespread in all rice grower country. Chemical zinc fertilizer used by the rice grower to encounter this issue become unavailable for plant uptake after two weeks of the application. The alternative way to solve this zinc issue is through biological method by using bacteria that cable in solubilizing zinc thus make it available to plant. This study was designed to isolate zinc-solubilizing bacteria (ZSB), identify its species and evaluate its potential as plant growth-promoters. Mineral salts medium (MSM) with the addition of insoluble zinc was used to confirm the bacterial zinc solubilization ability followed by nitrogen fixing ability, phosphate solubilization ability, potassium solubilization ability, hydrolyzing enzyme production and siderophores production. Out of 28 bacterial strains isolated from rice soil, ten were classified as ZSB. Isolate SR R-10 was the best ZSB as it obtained a significantly highest solubilization efficiency for zinc oxide (558.33%), zinc carbonate (419.33%). Isolate SR R-10 also records the highest phosphate solubilization (146.67%), while isolate SR R-12 was the best potassium solubilizer, recorded at 225% solubilization efficiency. Eight ZSB isolates were found to be able to fix atmospheric nitrogen, and seven were positive in siderophores and hydrolyzing enzyme production. Regarding phytohormone production, five selected ZSB isolates (SR N-2, SR N-3, SR R-2, SR R-10 and SR R-12) evaluated from its biochemical properties were tested. Isolate SR R-10 produced the highest indole compounds (6.140 μg mL^{-1}) followed by SR R-2 and SR N-3 which 5.865 and 5.167 μg mL^{-1} of indoles compound, respectively. Based on the characterization, two best ZSB isolates were SR R-10 and SR R-12. A 16S rRNA molecular identification result revealed that these two ZSB isolates, SR R-10 and SR R-12, are identified as *Acinetobacter nosocomialis* (ON834324) and *Acinetobacter seifertii* (ON954519), respectively. *In vivo* experiment with four treatments and four replications was conducted to determine the effect of ZSB inoculation on the growth of rice plant at 40 days after sowing and *Acinetobacter nosocomialis* (SR R-10) with *Acinetobacter seifertii* (SR R-12) were used as the inoculants. The treatments were rice plant: 1) without ZSB inoculation, 2) inoculated with SR R-10, 3) inoculated with SR R-12 and 4) inoculated with SR R-10 + SR R-12. Rice height, root length, biomass, leaf greenness and number of leaves were significantly positive affected by the ZSB inoculation with rice inoculated with SR R-10 has the highest reading of plant height (63.467 cm), root length (19.933 cm), biomass (4.670 g) and SPAD reading (32.667). Analysis of nutrient contents in rice leaves showed that the concentrations of nitrogen, phosphorus, potassium, and zinc were significantly increased by the ZSB inoculation while iron concentrations in the leaves was significantly reduced by the ZSB inoculation. Soil pH was also significantly reduced as the lowest pH was recorded at 5.793 (SR R-10) compared to 5.933 (non-inoculated). The reduction in soil pH was good for rice growth as some of the macro and micronutrients solubilized at low soil pH attributed to the production of organic acids by the bacterial isolates. While for available zinc in soil, only two inoculation treatments (SR R-10 and SR R-10 + SR R-12) were significant increased as they recorded 5.373 and 6.171 mg kg^{-1} Zn, respectively. From the results, the inoculation of ZSB has been successful in enhancing the growth of rice plants and potentially used as a biofertilizer to minimize chemical fertilizer usage.

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CHAPTER ONE
INTRODUCTION

1.1 Research Background

In Malaysia, rice plant is one the major crops after oil palm and rubber and it is a major staple food for its local. The good crop yield is depending on the adequate availability of both macronutrients and micronutrients. Zinc deficiency has been reported to be involved in reducing rice yields. In soils, high soil pH, low organic matter content, excessive use of phosphate fertilizer, less textured soil, and use of synthetic or chemical fertilizer to correct zinc deficiency instead of organic fertilizer are the conditions that favour zinc inaccessible to plants, leading to zinc unavailability after a week of application (Sunitha et al., 2014).

Farmers typically have a high dependency on chemical fertilizers in agricultural practices to compensate for the lack of critical nutrients, thereby achieving high yields. The excessive and unreasonable use of chemical fertilizers will contaminate the groundwater as well as create an unhealthy environment via greenhouse gas emissions, which exacerbates global warming and climate change (Shakeel et al., 2015). Zinc is unavailable micronutrient that can be reverted by bacterial strains which capable of solubilizing it to the available form. The beneficial bacteria also help to boost crop production via nitrogen fixation, phosphate-solubilizing activity, and phytohormone production. Thus, this research will therefore concentrate on the impact of zinc-solubilizing bacteria (ZSB) isolated from a rice area and how the plant-soil microbe interactions can influence the growth and nutrient content in rice plant. Laboratory studies were conducted to characterize the ZSB as plant growth regulator while pot studies were conducted to observe the effect of ZSB inoculation on the growth of rice plant.

1.2 Problem statement

Zinc deficiency is one of the major challenges for rice cultivation in Malaysia and has been reported that most of the rice soil series in Kedah and Kelantan contained