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EFFECT OF PHAGES ON SOME PHOSPHATE SOLUBLE BACTERIA IN THE SOIL

By

ALAA MOHAMED FATHI EL-SAYED BARAKAT
B.Sc. of Agriculture (Biothenology), Fac. of Agric., Ain Shams Univ., 2011
M.Sc. of Microbiology (Virology), Fac. of Agric., Ain Shams Univ., 2017

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Approval Sheet

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ABSTRACT

Alaa Mohamed Fathi El-Sayed Barakat: Effect of phages on some phosphate soluble bacteria in the soil. Unpublished Ph.D Thesis, Department of Agricultural Microbiology, Faculty of Agriculture, Ain Shams University, 2022.

Phosphate-solubilizing microorganisms play an important role in supplementing phosphorus to the plants, allowing a sustainable use of phosphate fertilizers. Microorganisms are involved in a range of process that affect the transformation of soil phosphorus (P) and thus are integral component of the soil 'P' cycle. Several mechanisms like lowering of soil pH by acid production, ion chelation, and exchange reactions in the growth environment have been reported to play a role in phosphate solubilization by Phosphate- solubilizing bacteria (PSB). It is slowly emerging as important organisms for the soil improvement.

In this study four bacterial viruses (phages) specific for *Bacillus velezensis* were isolated on the basis of the differences between the plaque morphology. The phages were signed as Bv_1- Bv_2- Bv_3 and Bv_4 . The isolated phages were propagated by the enrichment liquid method which gave particles with concentration of 10^{11} PFU/ml. Phages were purified and concentrated by differential centrifugation. Electron microscopy of the 2% urinyal acetate negatively stained showed that phages were belonging to family Siphoviridiae.

Bacillus velezensis phages infect many strains belong to its species, which means that they have a broad host range. Physical properties of *B. velezensis* phages revealed that, they different in their stability to TIP, DEP and *LIV*, pH degrees, Osmotic shock and effect of different solvents.

Sodium dodecyl sulfate polyacrylamide gel electrophoresis (SDS-PAGE) technique was performed to determine the properties of viral proteins. The purity and quantity of isolated DNAs were determined spectrophotometrically. Data showed that concentration of Bv_1 DNA was 0.75 µg/µL , Bv_2 DNA and Bv_3 DNA were 0.60 µg/µL and finally Bv_4 DNA 0.55 µg/µL. The analysis of genetic material of *B. velezensis* phages was determined based on both Inter Simple Sequence Repeat (ISSR) PCR technique and restriction enzymes. Data showed different amplifications patterns with all phages .

Finally, study the effect of *B. velezensis* phages on *B. velezensis* as solubilized phosphate bacteria and it's reflectance on sunflower was investigated in the pot experiment. The results revealed that : adding the rock phosphate to the soil led to increase plant height, stem diameter, number of leaves, chlorophyll A&B, caroteins, prolein, head diameter, seed weight and the oil percentage .Inoculation of the sandy soil with the bacteria increased solubilized phosphate .The virus deceased the total count of bacteria .Inoculation of the sandy soil with the bacteria increased plant height, steam diameter, number of leaves, chlorophyll A&B, caroteins, prolein, head diameter, seed weight and the oil percentage .The infection of bacteria with the virus reduced the parameters mentioned before .

Keywords: Bacteriophages, Solubilizing phosphorus, Phage stability, Electron microscopey, SDS-PAGE, Restriction enzymes and ISSR – PCR.

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CONTENTS

LIST OF TABLES

LIST OF FIGURES