

NUTRIENT ENHANCEMENT OF FLOODED
SOIL BY SYMBIOTIC SOIL FUNGI

NOR HAZWANI BINTI AZIZ

Doctor of Philosophy

UNIVERSITI MALAYSIA PAHANG



SUPERVISOR'S DECLARATION

We hereby declare that we have checked this thesis, and, in our opinion, this thesis is adequate in terms of scope and quality for the award of the degree of Doctor of Philosophy.

ASSOC. PROF. IR. DR. NORAZWINA BT. ZAINOL
DEPUTY DEAN
ACADEMIC & STUDENT AFFAIR
COLLEGE OF ENGINEERING
UNIVERSITI MALAYSIA PAHANG
LEBUHRAYA TUN RAZAK, 26300 KUANTAN
TEL : +609-549 2690 FAX : +609-549 2689

(Supervisor's Signature)

Full Name : IR. DR. NORAZWINA BINTI ZAINOL
Position : ASSOCIATE PROFESSOR
Date : 7 MAY 2021

DR. SITI HATIHAH BINTI MORTAN
SENIOR LECTURER
DEPARTMENT OF CHEMICAL ENGINEERING
COLLEGE OF ENGINEERING
UNIVERSITI MALAYSIA PAHANG
LEBUHRAYA TUN RAZAK
26300 GAMBANG, KUANTAN, PAHANG
TEL : +609-549 2824 FAX : +609-549 2689

(Co-supervisor's Signature)

Full Name : DR. SITI HATIHAH BINTI MORTAN
Position : SENIOR LECTURER
Date : 7 MAY 2021



STUDENT'S DECLARATION

I hereby declare that the work in this thesis is based on my original work except for quotations and citations which have been duly acknowledged. I also declare that it has not been previously or concurrently submitted for any other degree at Universiti Malaysia Pahang or any other institutions.

A handwritten signature in black ink, appearing to be 'NOR HAZWANI BINTI AZIZ', written over a horizontal line.

(Student's Signature)

Full Name : NOR HAZWANI BINTI AZIZ

ID Number : PKB15001

Date : 7 MAY 2021

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NOR HAZWANI BINTI AZIZ

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ABSTRAK

Fenomena banjir telah diklasifikasikan sebagai salah satu ancaman utama bagi Asia dan Rantau Pasifik, dan bahkan kepada kebanyakan bahagian di dunia ini. Walaupun bencana banjir bukan merupakan fenomena yang berbahaya, namun bencana ini menunjukkan peningkatan jumlah kekerapan yang ketara dan telah dikenal pasti sebagai peristiwa yang paling kerap berlaku. Antara kesan buruk banjir adalah ia menyebabkan tanah menjadi tandus di mana tanah tidak dapat menyokong pertumbuhan tanaman kerana kehilangan nutrien dan mikroorganisma hidup yang bermanfaat dalam membantu pengambilan air dan nutrien. Oleh itu, satu kaedah biologi telah dicadangkan untuk memulihkan kesuburan tanah dengan memanfaatkan keupayaan kulat tanah dalam membantu pengambilan nutrien dari tanah dengan membentuk interaksi simbiotik dengan tanaman perumah (*Allium cepa*). Sehubungan dengan itu, kajian semasa ini berusaha untuk menyaring faktor-faktor yang mempengaruhi pemulihan tanah banjir dan mengoptimalkan keadaan untuk pemulihan nutrien secara optimum oleh kulat tanah. Selain itu, kajian ini bertujuan untuk memencilkan dan mengenal pasti populasi kulat tanah dari tanah yang dirawat, dan untuk melakukan kajian kinetik ke atas pengambilan nutrien oleh kulat tanah melalui model Michaelis-Menten, serta untuk menentukan mekanisme pembebasan nutrien oleh tanah kulat. Dalam kajian semasa ini, tanah subur yang mengandungi kulat secara semula jadi telah disampelkan dari tanah yang subur, manakala tanah banjir telah disimulasikan melalui kaedah rumah hijau. Pertumbuhan tanaman perumah telah dikaji terlebih dahulu dalam tiga keadaan tanah iaitu tanah yang subur, tanah yang dibanjiri dan tanah yang di rawat (campuran tanah subur dan tanah banjir). Kepekatan nutrien (nitrogen, fosforus dan kalium) telah diukur dengan menggunakan spektrofotometer. Faktor-faktor yang mempengaruhi pemulihan tanah telah dipilih dan disaring menggunakan rekabentuk pecahan faktorial sementara pengoptimuman pemulihan tanah banjir telah dilakukan dengan menggunakan kaedah gerak balas permukaan. Kulat tanah telah dipencilkan dan dikenalpasti menggunakan Sistem Pengenalpastian Biolog Gen III Microplate. Kajian kinetik telah dijalankan dan analisis enzimatik dilakukan untuk menyelidiki mekanisme pembebasan nutrien oleh kulat tanah. Penyaringan faktor-faktor yang mempengaruhi pemulihan tanah membuktikan bahawa empat faktor termasuk kandungan air, bekalan cahaya, kedalaman tanah dan nisbah tanah mempunyai kesan yang signifikan terhadap pemulihan nutrien. Kandungan air tanah pada 28 mL dan pada 5 cm kedalaman tanah merupakan keadaan optimum dalam meningkatkan kepekatan nutrien di dalam tanah. Pada keadaan ini, pengambilan nutrien oleh kulat tanah dikatakan berada pada kadar maksimum di mana kepekatan nutrien meningkat dua kali ganda daripada kepekatan awal yang direkodkan. Pemalar kinetik V_{max} dan K_m yang diperolehi adalah berbeza untuk nitrogen, fosforus dan kalium dan dilaporkan sebagai 6.28mg/L.d dan 82.17 mg/L untuk nitrogen, 9.80 mg /L.d dan 60.96 mg/L.d untuk fosforus dan 0.07mg/L.d dan 4.55 mg/L.d untuk kalium masing-masing. Dua spesies kulat telah dikenal pasti dengan kebarangkalian lebih daripada 90% iaitu *Aspergillus aculeatus* dan *Paecilomyces lilacinus*. Penemuan kajian ini menunjukkan bahawa inokulasi tanah banjir dengan tanah subur yang mengandungi kulat tanah yang bermanfaat dapat meningkatkan kepekatan nutrien dengan ketara dan dapat mengurangkan penggunaan baja berasaskan kimia untuk membantu mencapai pertanian lestari.

ABSTRACT

Flooding was classified as one of the major threats to Asia and the Pacific region, and in fact, to numerous parts of the world. While the impacts are not the deadliest, this sort of common catastrophes exhibited a sharp increment in number and was identified as the most frequent event. The worsening effect of flooding is that it left behind the barren land where the soil could not support plant growth due to the leaching of nutrients and the losses of beneficial living microorganisms that facilitate the uptake of water and nutrients. Therefore, a biological method was proposed to restore soil fertility by exploiting the ability of soil fungi in promoting the uptake of nutrients from the soil by forming a symbiotic interaction with the host plants (*Allium cepa*). In light of this, this current study attempts to screen the factors affecting the recovery of flooded soil and optimize the condition for optimum recovery of nutrients by soil fungi. Also, this study aimed to isolate and identify the soil fungi population from the treated soil and to perform the kinetic study of nutrient uptake by soil fungi through the Michaelis-Menten model, as well as to determine the mechanism of nutrient released by soil fungi. In this current study, the fertile soil containing naturally occurring fungi was collected from fertile land, and flooded soil was simulated under the greenhouse. The host plant's growth was preliminarily studied in three soil conditions, namely fertile soil, flooded soil, and the treated soil (a mixture of fertile and flooded soil). The concentration of nutrients (nitrogen, phosphorus, and potassium) was measured by using a spectrophotometer. The factors affecting the soil recovery were selected and screened by fractional factorial design, while optimization of flooded soil recovery was conducted by employing response surface methodology. Soil fungi were isolated and identified by the Biolog Gen III MicroPlate Identification system. A kinetic study was carried out, and the enzymatic assay was performed to investigate the mechanism of nutrients released by soil fungi. The screening of factors affecting the soil recovery presented that four factors, including water content, light supply, soil depth, and soil ratio to have significant effects on the recovery of nutrients. Soil water content at 28 mL and soil depth of 5 cm was found to be optimum in increasing the concentration of nutrients in the soil. At this condition, the nutrient uptake by soil fungi was said to be at their maximum rate, where the concentration of nutrients was doubled from the recorded initial concentration. The kinetic constant of V_{max} and K_m obtained were varied for nitrogen, phosphorus, and potassium and were reported as 6.28 mg/L.d and 82.17 mg/L for nitrogen, 9.80 mg/L, and 60.96 mg/L.d for phosphorus and 0.07 mg/L and 4.55 mg/L.d for potassium, respectively. Two fungi species were identified with a probability of more than 90%, namely *Aspergillus aculeatus* and *Paecilomyces lilacinus*. The finding of this study suggests that inoculating flooded soil with fertile soil containing beneficial soil fungi could significantly increase the nutrient concentration and hence could reduce the application of chemical-based fertilizers in order to achieve sustainable farming.

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