BIOPROSPECTING FOR POTENTIAL USEFUL MICROORGANISMS FROM MWAKIRUNGE DUMPSITE IN MOMBASA COUNTY, KENYA

BERYLE ATIENO OKOTH

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DECLARATIONS

This thesis is my original work and has not been presented for a degree award in any other University.

Signature……. Date…….

Beryle Atieno Okoth

MSB/0013/2018

This thesis has been submitted with our approval as University Supervisors

Signature……. Date…….

Dr. Huxley Mae Makonde

Department of Pure and Applied Sciences

Technical University of Mombasa

Signature……. Date…….

Dr. Cromwell Mwiti Kibiti

Department of Pure and Applied Sciences

Technical University of Mombasa

Signature……. Date…….

Dr. Carren Okeri Bosire

Department of Pure and Applied Sciences

Technical University of Mombasa

DEDICATION

This thesis is dedicated to my mom Judith Okoth, my dear husband Erick, my kids Clancy, Whitney and Jabali, my sisters Diolence and Caroline, and my brother Braine.

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LIST OF ABBREVIATIONS

APC	Air pollution control
BPA	Bisphenol A
BLAST	Basic Local Alignment Tool
CMC	Carboxymethyl cellulose
DNA	Deoxyribonucleic acid
DDT	Dichlorodiphenyltrichloroethane
EPA	Environmental protection agency
EDTA	Ethyl diamine tetraacetic acid
GHGs	Greenhouse gases
KW	Kitchen waste
KMFRI	Kenya Marine and Fisheries Research Institute
LB	Luria Bertani
MSW	Municipal solid waste
MSWM	Municipal solid waste management
NA	Nutrient agar
PET	Polyethene Terephthalate
PVC	Polyvinyl chloride
PS	Polystyrene
PE	Polyethylene
PP	Polypropylene
PVC	Polyvinyl chloride

PUR	Polyurethanes
PAHs	Polycyclic hydrocarbons
PBDEs	Polybrominated diphenyl ethers
PCBs	Polychlorinated biphenyls
PDA	Potato dextrose agar
PCR	Polymerase chain reaction
RNA	Ribonucleic acid
SWM	Solid waste management
SWG	Solid waste generation
SA	Starch agar
SCA	Starch casein agar
TCA	Tricarboxylic cycle
TE	Tris EDTA

ABSTRACT

Accumulation of solid waste as a result of urbanization, industrialization and the increasing human population is one of the biggest problems affecting the globe today. The conventional methods used for disposal of solid waste create even more environmental problems; hence, there is need for sustainable alternatives of mitigating solid waste pollution. The aim of this study was to isolate and identify potential useful microorganisms that are able to degrade and utilize diverse organic and inorganic materials from Mwakirunge dumpsite-Mombasa. A total of 16 soil samples were collected using a randomized block design at Mwakirunge dumpsite. The samples were inoculated in an enriched basal media containing mixed municipal solid waste and incubated at 37°C for 21 days. A total of 20 fungal and 46 bacterial isolates were recovered. Microbial identification was done using standard morphological, biochemical and molecular approaches. Pure genomic DNA was isolated from the pure recovered microbial isolates using organic DNA isolation methods. PCR amplification was conducted using universal prokaryotic primers (27F and 1492R) for bacteria and (ITSI and ITS4) for fungal isolates. Sequencing of the purified PCR products was done at Inqaba Biotech (S.A). Evolutionary analysis of the sequences obtained was done using BLASTn algorithm. Evolutionary analysis of the 16S rRNA gene sequences grouped the bacterial isolates into three (3) phyla; Actinobacteria, Firmicutes and Proteobacteria that included members affiliated to the genera *Bacilli, Pseudomonas, Brevibacilli, Microbacterium, Ochrobactrum, Paenibacillus*, *Staphylococcus, Isoptericola* and *Streptomyces*. Phylogenetic analysis of the ITS gene sequences grouped fungal isolates into the phylum Ascomycota with members from the genus *Aspergillus.* The ability of the isolated microbes to secrete useful extracellular enzymes was tested on media supplemented with tween 20, tween 80, carboxy methyl cellulase (CMC), starch and gelatin and the results showed a significant level of enzyme production by the isolates ($p<0.05$). *Bacillus cereus* (MZ571899) exhibited the highest esterase activity; *Streptomyces thermocarboxydus* (MZ5718820) exhibited the highest lipase activity, *Bacillus subtilis* (MZ571887) exhibited the highest amylase activity, *Bacillus licheniformis (*MZ571888*)* exhibited the highest cellulase activity while *Pseudomonas stutzeri* (MZ571900) exhibited the highest gelatinase activity. This study confirms that diverse soil microorganisms from the dumpsite have potential of waste degradation. These microbes together with their enzymes can be further studied to improve their biodegradation potential by genetic engineering. The potential useful isolates and enzymes screened from this study can also be produced in large quantities for industrial and other biotechnological applications. Three (3) nearly novel bacterial isolates B4S2 b (MZ571886), B3S1 (MZ571907) and B3S4 B (MZ571915) and one fungal isolate B2S2 a1(MZ569413) were isolated which could further be characterized using a polyphasic approach.