Botany Department
Faculty of Women
Ain Shams University

BIOLOGICAL PULPING OF SOME AGRICULTURAL WASTES

Thesis
Submitted in Partial Fulfillment
of the Requirements for the Degree of
Master of Science in Microbiology

By

Douaa Hussein Abdel-Aziz Ibrahim
B.Sc. (Microbiology-Chemistry), Faculty of Women for Arts-Science and Education, Ain Shams University, 2007

Botany Department
Faculty of Women for Arts - Science and Education
Ain Shams University
2014

BIOLOGICAL PULPING OF SOME AGRICULTURAL WASTES

Thesis
Submitted in Partial Fulfillment
of the Requirements for the Degree of
Master of Science in Microbiology

By

Douaa Hussein Abdel-Aziz Ibrahim
B.Sc. (Microbiology- Chemistry), Faculty of Women for Arts- Science and Education, Ain Shams University, 2007

Under the Supervision of

Prof. Dr. Zeinab M. H. Kheiralla
Botany Dept.
Faculty of Women for Arts-
Science and Education,
Ain Shams Univ.

Prof. Dr. Said M. Badr El-Din
Agricultural Microbiology Dept.
National Research Centre

Prof. Dr. Saad M. Abdel Malek
Agricultural Microbiology Dept.
National Research Centre

Botany Department
Faculty of Women for Arts - Science and Education
Ain Shams University
2014

Botany Department
Faculty of Women
Ain Shams University

Approval sheet

BIOLOGICAL PULPING OF SOME AGRICULTURAL WASTES

By
Douaa Hussein Abdel-Aziz Ibrahim
B.Sc. (Microbiology- Chemistry), Faculty of Women for Arts- Science and Education, Ain Shams University, 2007

Supervisor	**Approved**
Prof. Dr. Zeinab M. H. Kheiralla Botany Dept. Faculty of Women for Arts - Science and Education, Ain Shams Univ.	------------------
Prof. Dr. Said M. Badr El-Din Agricultural Microbiology Dept. National Research Centre	-------------------
Prof. Dr. Saad M. Abdel Malek Agricultural Microbiology Dept. National Research Centre	--------------------

BIOLOGICAL PULPING OF SOME AGRICULTURAL WASTES

Thesis Submitted in Partial Fulfillment of the Requirements for the Degree of Master of Science in Microbiology

By

Douaa Hussein Abdel-Aziz Ibrahim

B.Sc. (Microbiology- Chemistry), Faculty of Women for Arts- Science and Education, Ain Shams University, 2007

Under the Supervision of

Prof. Dr. Zeinab M. H. Kheiralla
Botany Dept.
Faculty of Women for Arts- Science and Education,
Ain Shams Univ.

Prof. Dr. Said M. Badr El-Din
Agricultural Microbiology Dept.
National Research Centre.

Prof. Dr. Saad M. Abdel Malek
Agricultural Microbiology Dept.
National Research Centre.

Arbitration Committee

Prof. Dr. Said M. Badr El-Din, Professor of Microbiology, National Research Centre.
Prof. Dr. Zeinab M. H. Kheiralla- Professor of Microbiology, Faculty of Women, Ain Shams University.
Prof. Dr. Mohamed Ibrahim Ali- Professor of Microbiology- Cairo university.
Prof. Dr. Mohamed Elsayed Osman- Professor of Microbiology-Helwan university.

Botany Department
Faculty of Women for Arts - Science and Education
Ain Shams University- 2014

This thesis has not been submitted for any degree at this or any other university

Signed

Douaa Hussein Abd-El Aziz

ACKNOWLEDGMENT

A special word of thank goes to **Dr. Zeinab Kheiralla**, Professor of Microbiology, Faculty of Women for Arts- Science and Education, Ain Shams University, for her encouragement and her interest in this work. Also a great thanks to **Dr. Said M. Badr El-Din**, Professor of Agriculture Microbiology, National Research Centre, for suggesting the topic of this thesis, earnest supervision, patience, valuable advises, and for offering every possible help.

I would like to express my great appreciation for **Dr. Saad M. Abdel Malek,** Professor of Agriculture Microbiology, National Research Centre, for his valuable advices and supervision.

I want to thank all my professors in the Botany Department, Faculty of Women for Arts- Science and Education and all members of the Department. Thanks are also extended to all staff members (my Professors and my colleagues) in the Agriculture Microbiology Department, the National Research Centre, for the warm feeling of the one family spirit, and for everyone gave me a hand of help.

Dedication

*I would like to dedicate this thesis to my wonderful family (**my beloved father Hussein, my beloved mother Souad, my great husband Ahmed and my dearest sister Ghada**). All words of thanks and gratitudes will not be enough for all what they have done for me, and for their continuous help, support and encouragement.*

Douaa Hussein Abd-El Aziz

BIOLOGICAL PULPING OF SOME AGRICULTURAL WASTES

By

Douaa Hussein Abdel-Aziz Ibrahim

ABSTRACT

The present work aims to evaluate the fungal pretreatments for use in biopulping processes of agricultural wastes (rice straw). Therefore, Sixty two fungal isolates were screened for lignin peroxidase production. The most potent isolates for the lignin peroxidase production were identified as *Phanerochaete chrysosporium* and *Pleurotus ostreatus* using DNA sequence of Internal Transcribed Spacer (ITS) region. The optimization cultural conditions for the optimum production of lignin peroxidase were investigated. Of the media used, Sabouraud broth medium showed the highest production of lignin peroxidase with both fungal isolates. Lignin peroxidase production has been optimized for these isolates using the following conditions; pH 5.0, temperature 30 ^{0}C, agitation at 100 rpm and 4 days of incubation. Fungal discs have been found to be the most suitable inoculums than mycelia suspension. Urea as a sole N-source and fructose as a sole C-sources were more suitable for lignin peroxidase production at concentration of 0.30 and 3.0 %, respectively. The pretreatment of rice straw with *P. chrysosporium*, *Pl. ostreatus* or lignin peroxidase for use in biopulping process was studied. Pretreatment of rice straw with *P. chryosporium* for 25 days resulted in a substantial decrease in pulp yield (by 9.1 %) and kappa number (by 25.6 %). Losses of pulp yield and kappa number were considerably low with lignin peroxidase treatment (3.7 and 14.1 %, respectively). However, the pretreatment of rice straw with *Pl. ostreatus* isolate caused moderate pulp yield losses (5.8 %) and preferential lignin degradation (as kappa number losses by 34.6 %). This indicated that the isolate of *Pl. ostreatus* might be superior to isolate of *P. chrysosporium* and lignin peroxidase for use in biopulping process or other processes in which preferential lignin degradation is desired.

Key words:
Biopulping, *Phanerochaete chrysosporium*, *Pleurotus ostreatus*, Lignin peroxidase, rice straw.

CONTENTS

List of Tables

List of Figures

List Of Abbreviations

Abbreviation	Word
Bp	Base pair
C-source	Carbon source
CTMP	combination thermo mechanical process
DO	Dissolved oxygen
ITS	Internal transcribed spacer
KDa	KiloDalton
Lac	Laccase
LDPs	Lignin degrading peroxidase enzymes
LiP	Lignin peroxidase
MEB	malt extract broth
MnP	Manganese peroxidase
MSB	mineral salt broth
P. chrysosporium	*Phanerochaete chrysosporium*
PCR	Polymerase Chain Reaction
PDA	Potato dextrose medium
pI	Isoelectric point

List Of Abbreviations

Pl. ostreatus	*Pleurotus ostreatus*
Rpm	Round per minute
TMP	thermomechanical pulp
VA	Veratryl alcohol
w/v	Weight per volume