

**EFFECTS OF AZOLLA AND UREA ON GROWTH
PARAMETERS AND YIELD OF RICE**

A Dissertation

**Submitted for the Partial Fulfillment of the
Requirements for the M.Sc. in Botany**

By

Madhu Shudan Thapa Magar

Roll No. 655

Batch No. 060/062

T.U. Regd. No. 5-1-22-107-98

**Central Department of Botany
Tribhuvan University, Kirtipur
Kathmandu, Nepal**

2007

Prof. Dr. B.N. Prasad

Ph.D., F.B.S.

Office:
Professor (Plant Biotechnology)
Central Department of Botany
Tribhuvan University, Kirtipur
Kathmandu, Nepal
Tel: 977-1-4331322 (O)
Fax: 977-1-4332636



Mailing Address:
Residence
Professor's Quarter A-1
Tribhuvan University,
Kirtipur, Kathmandu, Nepal
Tel: (977-1) 4330582
Fax: (977-1) 4332636
E-mail: brajnandan06@yahoo.com

Ref. No.

21st December 2006

CERTIFICATE

This is to certify that Mr. Madhu Shudan Thapa Magar has completed the dissertation work entitled, "**Effects of Azolla and Urea on Growth Parameters and yield of Rice**" under my supervision. To the best of my knowledge this work has not been submitted for any other academic degree else where. I recommend this dissertation for the partial fulfillment of M.Sc. Botany with specialization on Biotechnology Degree in Tribhuvan University.

Prof. Dr. Braj Nandan Prasad

(Supervisor)

Biotechnology Unit

Central Department of Botany

Tribhuvan University, Kirtipur



TRIBHUVAN UNIVERSITY
INSTITUTE OF SCIENCE AND TECHNOLOGY
CENTRAL DEPARTMENT OF BOTANY

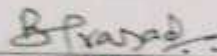
Ref. No.

Kirtipur, Kathmandu
Nepal

LETTER OF APPROVAL

The dissertation paper submitted by Madhu Shudan Thapa Magar entitled "Effects of *Azolla* and Urea on growth parameters and yield of rice" has been accepted as a partial fulfillment of M.Sc. in Botany.

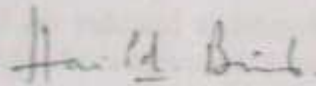
EXPERT COMMITTEE



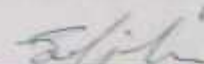
Prof. Dr. B.N. Prasad
(supervisor)
Central Department of Botany
Tribhuvan University



Prof. Dr. P.K. Jha
(Head)
Central Department of Botany
Tribhuvan University



Dr. H. P. Bimb
(External examiner)
Bio-Technology Unit
NARC, khumaltar, Lalitpur



Prof. Dr. S. D. Joshi
(Internal Examiner)
Central Department of Botany
Tribhuvan University

Date of examination: 29th March 2007

ACKNOWLEDGEMENTS

First of all I would like to express my sincere gratitude to my Supervisor Prof. Dr. B.N. Prasad, Central Department of Botany, T.U. for his valuable guidance and continuous suggestions through out the investigation period.

I am also thankful to Prof. Dr. P.K. Jha, Head of the Department, Central Department of Botany, T.U. for providing me the necessary facilities during the period of investigation.

My heartfelt thanks also goes to respected teacher Bharat Babu Shrestha, Lecturer, Central Department of Botany, T.U. for his kind help and suggestions during the analysis of soil samples in T.U. Laboratory.

I am thankful to all respected teachers of Central Department of Botany, T.U. for their suggestions and helps during academic period.

I am also thankful to Mr. Harikrishna Upreti (Agricultural Botany Division, NARC) for providing rice seeds required for research work.

My heartfelt thanks goes to friend Dhan Raj Kandel for bringing *Azolla pinnata* from Bishazari Lake, Chitwan.

I am indebted to Miss Sushma Gurung and Mrs. Nutan Sapkota for their valuable suggestions during research work.

My heartfelt thanks goes to my friends Purbendra Nath Yogi, Yagya Prasad Poudel, Nabin Bhattarai, Shreeti Pradhan, Saraswati Aryal, Binod Bastola, Dharma Raj Koirala, Dinesh Baral, Rajendra Acharya, Basanta Raj Pokhrel and all others who directly or indirectly helped on my research work.

I am thankful to Nepal Academy of Science and Technology (NAST) and Cornell Nepal Study Program (CNSP) for providing me financial support.

Finally, I am grateful to my family for helping me in all the possible ways to complete my research work properly.

ABSTRACT

The present work was carried out aiming to determine the effect of different level of urea along with different methods of *Azolla* application on Nitrogen and Organic matter content of soil, Chlorophyll content of rice and finally to assess its impact on the yield and yield components of rice. The work was performed both in field and pot and conducted in the year 2005.

Experimental results showed that growing of four crops of *Azolla* with incorporation thrice (first before transplanting, second and third after transplanting) was found more effective and have grain yield increment up to 47.13% and straw yield up to 40.97%. Three crops of *Azolla* with twice incorporation after transplanting plus 20 kg N ha⁻¹ have grain yield and straw yield increment up to 36.12% and 31.04% respectively which was almost equal to yield produced by NPK (80:40:30 kg ha⁻¹); N in split dose.

Increased number of crops of *Azolla* and incorporated frequently, was found helpful in increasing nitrogen and organic matter content of soil, chlorophyll content of rice and finally increase the yield of rice, which could reduce the amount of urea or may even substitute the urea from rice field. The present study also revealed that *A. caroliniana* was more effective biofertilizer since *A. caroliniana* exhibited higher Heterocyst frequency, lower Doubling time, higher Relative Growth Rate, chlorophyll content and Amino nitrogen than *A. pinnata*.

CONTENTS

	Page
CERTIFICATE	
LETTER OF APPROVAL	
ACKNOWLEDGEMENT	
LIST OF TABLES	
LIST OF FIGURES	
ABBREVIATIONS AND SYMBOLS	
ABSTRACT	
CHAPTER I : INTRODUCTION	1-5
1.1 Background	1
1.2 <i>Azolla</i>	4
CHAPTER II : OBJECTIVES	6-7
2.1 Objectives	6
2.2 Justification of the study	6
CHAPTER III : LITERATURE REVIEW	8-17
3.1 <i>Azolla</i> as biofertilizer	8
3.2 Chemical fertilizer	16
CHAPTER IV : MATERIALS AND METHODS	18-31
4.1 Study Area	18
4.1.1 Location and topography	18
4.1.2 Climate	18
4.1.3 Soil	20
4.2 Materials	20
4.2.1 Plant materials	20
4.2.2 <i>Azolla</i>	21
4.2.3 Chemical fertilizer	21
4.3 Methods	22
4.3.1 Field experiment	22

4.3.2 Pot experiment	24
4.3.3 Measurement of heterocyst frequency in <i>Anabaena azollae</i> inhabiting in <i>Azolla caroliniana</i> and <i>A. pinnata</i>	24
4.3.4 Measurement of Doubling time (Dt) and Relative Growth Rate (RGR) of <i>Azolla caroliniana</i> and <i>A. pinnata</i>	25
4.3.5 Estimation of chlorophyll in <i>Azolla caroliniana</i> and <i>A. pinnata</i>	26
4.3.6 Measurement of amino N ₂ in <i>Azolla caroliniana</i> and <i>A. pinnata</i>	27
4.3.7 Chemical analysis of soil sample	27
4.3.7.1 Determination of P ^H	27
4.3.7.2 Estimation of total nitrogen	28
4.3.7.3 Determination of organic matter	29
4.3.8 Estimation of chlorophyll in rice leaf	30
4.3.9 Measurement of yield and yield components	31
4.3.10 Statistical analysis	31
CHAPTER V : RESULTS	32-50
5.1 Results on Heterocyst frequency in <i>Anabaena azollae</i> inhabiting in <i>Azolla caroliniana</i> and <i>A. pinnata</i>	32
5.2 Results on Doubling time (Dt) and Relative Growth Rate (RGR) of <i>Azolla caroliniana</i> and <i>A. pinnata</i>	33
5.3 Results on chlorophyll content in <i>Azolla caroliniana</i> and <i>A. pinnata</i>	34
5.4 Results on amino nitrogen in <i>Azolla caroliniana</i> and <i>A. pinnata</i>	35

5.5 Results on effect of <i>Azolla</i> and Urea on chlorophyll content in rice leaf	37
5.6 Results on the effect of <i>Azolla</i> and Urea on the rice yield and yield components	39
5.7 Results on the effect of <i>Azolla</i> and Urea on the Nitrogen content of soil	48
5.8 Results on the effect of <i>Azolla</i> and Urea on organic matter content of soil	49
CHAPTER VI : DISCUSSIONS	51-56
6.1 Discussion on the Heterocyst frequency in <i>Anabaena azollae</i> inhabiting in <i>Azolla caroliniana</i> and <i>A. pinnata</i>	51
6.2 Discussion on Doubling time (Dt) and Relative Growth Rate (RGR) of <i>Azolla caroliniana</i> and <i>A. pinnata</i>	51
6.3 Discussion on chlorophyll and Amino nitrogen content in <i>Azolla caroliniana</i> and <i>A. pinnata</i>	52
6.4 Discussion on the effect of <i>Azolla</i> and Urea on chlorophyll content in rice leaves	52
6.5 Discussion on the effect of <i>Azolla</i> and Urea on the rice yield and yield components	53
6.6 Discussion on the effect of <i>Azolla</i> and Urea on the nitrogen content of the soil	55
6.7 Discussion on the effect of <i>Azolla</i> and Urea on organic matter content of soil	56
CHAPTER VII : CONCLUSION	57
CHAPTER VIII: RECOMMENDATIONS	58
CHAPTER IX: REFERENCES	59-65
PHOTO PLATES	
APPENDICES	

LIST OF TABLES

	Page
Table 1: Chemical formula and nutrient content of chemical fertilizers used in the experiment.	22
Table 2: Treatments in the field experiments	23
Table 3: Heterocyst frequency in <i>Anabaena azollae</i> of <i>A. caroliniana</i>	32
Table 4: Heterocyst frequency in <i>Anabaena azollae</i> of <i>A. pinnata</i>	33
Table 5: Dt and RGR in <i>A. caroliniana</i> and <i>A. pinnata</i>	34
Table 6: Absorbance reading and chlorophyll content in <i>A. caroliniana</i> and <i>A. pinnata</i>	35
Table 7: Absorbance reading of different Glycine concentration and extract of <i>A. caroliniana</i> and <i>A. pinnata</i> .	36

LIST OF FIGURE

	Page
Fig 1: Graphical Representation of Climatic data of Lalitpur (2005)	19
Fig 2: Graphical Representation of Climatic data of Kathmandu (2005)	20
Fig.3: Determination of amino acid from standard calibration curve.	36
Fig.4: Graphical representation of chlorophyll content in rice leaf of field at different DAT	38
Fig.5: Graphical representation of chlorophyll content in rice leaf of pot at different DAT	38
Fig.6: Graphical representation of plant height (cm) in the field	40
Fig.7: Graphical representation of plant height (cm) in the pot	40
Fig.8: Graphical representation of no. of panicles/hill in the field	41
Fig.9: Graphical representation of no. of panicles/hill in the pot	41
Fig.10: Graphical representation of no. of 1° branches/panicle in the field	42
Fig.11: Graphical representation of no. of 1° branches/panicle in the pot	42
Fig.12: Graphical representation of no. of filled grains/panicle in the field	43
Fig.13: Graphical representation of no. of filled grains/panicle in the pot	43
Fig.14: Graphical representation of % filled grain in the field	44
Fig.15: Graphical representation of % filled grain in the pot	44

Fig.16: Graphical representation of wt. of 1000 grains (gm) in the field	45
Fig.17: Graphical representation of wt. of 1000 grains (gm) in the pot	45
Fig.18: Graphical representation of grain yield $t\ ha^{-1}$ in the field	46
Fig.19: Graphical representation of grain yield (gm)/pot in the pot	46
Fig.20: Graphical representation of straw yield $t\ ha^{-1}$ in the field	47
Fig.21: Graphical representation of straw yield(gm)/pot in the pot	47
Fig.22: Graphical representation of Nitrogen content in soil in the field	48
Fig.23: Graphical representation of Nitrogen content in soil in the pot	49
Fig.24: Graphical representation of Organic matter content in soil in the field	50
Fig.25: Graphical representation of Organic matter content in soil in the pot.	50

ACRONYMS AND ABBREVIATIONS

ANOVA	Analysis of Variance
%	Percent
APP	Agricultural Prospective Plans
ARA	Acetylene Reducing Activity
BNF	Biological nitrogen fixation
(°C)	Degree Celsius
CBS	Central Bureau of Statistics
Chl.	Chlorophyll
DAT	Days after transplanting
Dt.	Doubling time
<i>et al.</i>	et alebi(and others)
FAO	Food and Agricultural Organization
FYM	Farm yard manure
fwl	Fresh weight of leaf tissue
g	gram
GDP	Gross Domestic Product
ha	hectare
IAAS	Institute of Agriculture and Animal Science
IRRI	International Rice Research Institute
K	Potassium
Kg	Kilogram
MoAC	Ministry of Agriculture and Cooperatives
Mt	Metric tone
MPE	Ministry of Population and Environment
N	Nitrogen
NARC	Nepal Agriculture Research Council
NAST	National Academy of Science and Technology
nm	nano meter

OD	Optical Density
OM	Organic Matter
P	Phosphorus
p ^H	Negative logarithm of Hydrogen ion concentration
RGR	Relative Growth Rate
SPSS	Statistical Package for Social Sciences
t ha ⁻¹	tons per hectare
T.U.	Tribhuvan University
viz.	videlicet(namely)
Vol.	Volume
wt.	weight