UNIVERSITAS
GADJAH MADA
Diversity of Lignocellulose-Degrading Bacteria from the Gastro-Intestinal Tract of Bawean Deer (Axis kuhlii)
Arief Muammar, Prof. Dr. Endah Retnaningrum, M.Eng.; Assoc. Prof. Dr. Clemens Karl Peterbauer; Prof. Dr. Budi Se
Universitas Gadjah Mada, 2026 | Diunduh dari http://etd.repository.ugm.ac.id/

TABLE OF CONTENTS

COVER PAGE i
TITLE PAGE ii
DEAN'S APPROVAL PAGE iii
DISSERTATION SUPERVISORY COMMITTEE APPROVAL PAGE iv
DISSERTATION APPROVAL PAGE v
DECLARATION OF ORIGINALITY AND NON-PLAGIARISM vi
ACKNOWLEDGMENT vii
TABLE OF CONTENTS viii
LIST OF TABLE x
LIST OF FIGURES xi
LIST OF APPENDICES xii
ABSTRACT xiii
INTISARI xiv
I. INTRODUCTION
A. Background 1
B. Research questions 4
C. Aims and benefits of research 5
D. Originality of the research 6
E. Scope of the research 9
II. LITERATURE REVIEW
A. Lignocellulose 10
B. Enzymes play a role in lignocellulose degradation 11
C. Lignocellulose-degrading enzyme database 13
D. Source of lignocellulose-degrading enzyme in lignocellulolytic microorganisms 15
E. Analysis of diversity of structure and function of microorganisms with culture-independent methods (metagenomics approach) 19
III. THEORETICAL BASIS AND HYPOTHESIS
A. Theoretical basis 21
B. Hypothesis 24
IV. METHODS
A. Period and area of study 25
B. Material 26
C. Equipment 28
D. Research design 28
E. Procedure 28
F. Data analysis 36
V. RESULTS
A. Variation in DNA yield and quality across different collection, preservation, and extraction methods 38
B. Differences in gut bacterial diversity and abundance between deer from distinct habitats 40
C. Variation in lignocellulolytic gene diversity between wild and captive deer, though overall functional gene group composition remains similar 45
D. Active lignocellulolytic genes using an independent culture approach 50

UNIVERSITAS
GADJAH MADA
Diversity of Lignocellulose-Degrading Bacteria from the Gastro-Intestinal Tract of Bawean Deer (Axis kuhlii)
Arief Muammar, Prof. Dr. Endah Retnaningrum, M.Eng.; Assoc. Prof. Dr. Clemens Karl Peterbauer; Prof. Dr. Budi Se
Universitas Gadjah Mada, 2026 | Diunduh dari http://etd.repository.ugm.ac.id/

VI. GENERAL DISCUSSION 55
VII. CONCLUSION AND SUGGESTION
A. Conclusion 65
B. Suggestion 67
SUMMARY 69
RINGKASAN 72
REFERENCES 75
APPENDICES 87

UNIVERSITAS
GADJAH MADA
Diversity of Lignocellulose-Degrading Bacteria from the Gastro-Intestinal Tract of Bawean Deer (Axis kuhlii)
Arief Muammar, Prof. Dr. Endah Retnaningrum, M.Eng.; Assoc. Prof. Dr. Clemens Karl Peterbauer; Prof. Dr. Budi Se
Universitas Gadjah Mada, 2026 | Diunduh dari http://etd.repository.ugm.ac.id/

LIST OF TABLE

Table1. Studies on intestinal and fecal microbiota of cervidae using *16S rRNA* gene sequencing …… 7

UNIVERSITAS
GADJAH MADA
Diversity of Lignocellulose-Degrading Bacteria from the Gastro-Intestinal Tract of Bawean Deer (Axis kuhlii)
Arief Muammar, Prof. Dr. Endah Retnaningrum, M.Eng.; Assoc. Prof. Dr. Clemens Karl Peterbauer; Prof. Dr. Budi Se
Universitas Gadjah Mada, 2026 | Diunduh dari http://etd.repository.ugm.ac.id/

LIST OF FIGURES

Figure 1. Three types of cellulase enzymes and how they degrade cellulose 12
Figure 2. Bawean deer and its habitat 17
Figure 3. Nature reserve and wildlife sanctuary area Bawean island 25
Figure 4. Bawean island map 29
Figure 5. Comparison between swab and bolus samples in DNA quality and PCR amplification rate 38
Figure 6. Comparison between different preservatives in DNA quality and PCR amplification rate 39
Figure 7. Total reads from twelve stool samples 40
Figure 8. Relative abundance of bacterial communities from fecal deer at phylum and species level 41
Figure 9. Principle Coordinate Analysis of fecal bacterial communities in three different species of deer 43
Figure 10. Heatmap of relative abundances of functional prediction 45
Figure 11. Abundance of Carbohydrate Enzyme (CAZyme) in Bawean deer samples 46
Figure 12. Abundance of Glycoside Hydrolases in Bawean deer samples 47
Figure 13. Abundance of Carbohydrate Esterases in Bawean deer samples 48
Figure 14. Abundance of Glycosyltransferases in Bawean deer samples 48
Figure 15. Abundance of Polysaccharide Lyases in Bawean deer samples 49
Figure 16. Abundance of Carbohydrate-binding Modules in Bawean deer samples 49
Figure 17. Abundance of *endoglucanase* and β- *glucosidase* from 9 stool sample of *A. kuhlii* 50
Figure 18. Endoglucanase CMC assay and β-glucosidase 4MUGlu Assay 51
Figure 19. SDS PAGE of endoglucanase 52
Figure 20. SDS PAGE and Western Blot of β-glucosidase 52
Figure 21. SDS PAGE of cellobiohydrolase 53
Figure 22. SDS PAGE of aryl-alcohol dehydrogenase 54

UNIVERSITAS
GADJAH MADA
Diversity of Lignocellulose-Degrading Bacteria from the Gastro-Intestinal Tract of Bawean Deer (Axis kuhlii)
Arief Muammar, Prof. Dr. Endah Retnaningrum, M.Eng.; Assoc. Prof. Dr. Clemens Karl Peterbauer; Prof. Dr. Budi Se
Universitas Gadjah Mada, 2026 | Diunduh dari http://etd.repository.ugm.ac.id/

LIST OF APPENDICES

Appendix 1. Scheme of metagenomic strategies for the identification of novel cellulases from environmental samples …… 87
Appendix 2. Phylogenetic tree inferred from the eight sequences resulting in active enzymes and the ten best BLAST hits for each one ……... 88
Appendix 3. Electrophoresis results of amplified cellulases by Multiplex PCR, HIFI Assembly cellulases to pPICZαA, and RCA (a) *endoglucanase* (b) β-*glucosidase* …….……. 89
Appendix 4. Screening of colonies to enable hydrolase xylan, PH acid/alkaline and thermostability…..… 90
Appendix 5. Cellulase genes selection for Multiplex PCR ..… 91
Appendix 6. The list of primers of *endoglucanase* ad β-*glucosidase* for HIFI Assembly to pPICZαA ….. 93
Appendix 7. The map of pPICZαA, pET21d and pBE-S plasmid …… 95
Appendix 8. Approval of access of *Sumber Daya Genetik* (SDG) …….. 97
Appendix 9. Cotutelle doctoral degree agreement ……... 101