



TABLE OF CONTENTS

THESIS COVER	i
LEGALIZATION SHEET	ii
FOREWORDS	iv
TABLE OF CONTENTS	vi
LIST OF TABLES	viii
LIST OF FIGURES	ix
LIST OF APPENDIXES	x
ABSTRACT	xi
CHAPTER I INTRODUCTION	1
1.1 Background	1
1.2 Problem statement	5
1.3 Objectives	5
1.4 Benefit of research.....	6
CHAPTER II LITERATURE STUDY	7
2.1 Oyster mushrooms.....	7
2.2 Tryptophan and Its Derivatives	10
2.3 Ultrasound-Assisted Extraction.....	16
2.4 Method Validation.....	21
2.5 Design Experimental	25
2.6 Response Surface Methodology	27
2.7 Research Hypothesis	27
CHAPTER III RESEARCH METHODS	29
3.1 Time and Place of Research	29
3.2 Research Material and Instruments	29
3.2.1 <i>Pleurotus spp.</i> Material	29
3.2.2 Chemicals and Reagent	30
3.3 Research Implementation	31
3.3.1 Samples preparation	31
3.3.2 Ultrasound-assisted extraction (UAE)	31
3.3.3 Identification and quantification of tryptophan and derivatives ...	31
3.3.4 Experimental design and statistical analysis	32
3.3.5 Determination of optimum extraction time.....	34



3.3.6 Stability assessment of tryptophan and derivatives	35
3.3.7 Method validation	35
CHAPTER IV RESULTS AND DISCUSSIONS	37
4.1 Performance evaluation of the UHPLC-PDA method	37
4.2 Identification of tryptophan and its derivatives in oyster mushroom extract.....	38
4.3 Screening of extraction parameters	40
4.3.1 Effect of solvent type on extraction recovery	40
4.3.2 Effect of solvent composition on extraction recovery	42
4.4 Multifactorial analysis using Box-Behnken design.....	42
4.5 Optimization of ultrasound-assisted extraction conditions	45
4.6 Evaluation of optimal extraction time	48
4.7 Stability assessment of tryptophan and its derivatives under optimized conditions	49
4.8 Validation of the UAE method.....	50
4.9 Application to real samples: Tryptophan and derivative profiles in oyster mushrooms	51
CHAPTER V CONCLUSION AND SUGGESTION	54
5.1 Conclusion.....	54
5.2 Suggestion	55
REFERENCES.....	56



UNIVERSITAS
GADJAH MADA

**OPTIMIZATION OF ULTRASOUND-ASSISTED EXTRACTION OF TRYPTOPHAN AND ITS
DERIVATIVES FROM OYSTER
MUSHROOM (*Pleurotus spp.*) THROUGH A RESPONSE SURFACE METHODOLOGY**

Anita Nurmulya Bahari, Dr. Widiastuti Setyaningsih, S.T.P., M.Sc.

Universitas Gadjah Mada, 2025 | Diunduh dari <http://etd.repository.ugm.ac.id/>

LIST OF TABLES

Table 1. Content of tryptophan and its derivatives in mushroom	13
Table 2. UAE method and its variables on tryptophan and its derivatives	20
Table 3. Data elements normally required for each of method validation.....	22
Table 4. Accepted recovery as a function of analyte concentration	23
Table 5. The standard of precision (CV%) valur from AOAC	23
Table 6. Independent factors and levels	33
Table 7. Peak area responses of tryptophan and its derivatives.....	33
Table 8. UHPLC-PDA performance for the analysis	37
Table 9. The concentration of tryptophan and its derivatives	52



UNIVERSITAS
GADJAH MADA

**OPTIMIZATION OF ULTRASOUND-ASSISTED EXTRACTION OF TRYPTOPHAN AND ITS
DERIVATIVES FROM OYSTER
MUSHROOM (*Pleurotus spp.*) THROUGH A RESPONSE SURFACE METHODOLOGY**

Anita Nurmulya Bahari, Dr. Widiastuti Setyaningsih, S.T.P., M.Sc.

Universitas Gadjah Mada, 2025 | Diunduh dari <http://etd.repository.ugm.ac.id/>

LIST OF FIGURES

Figures 1. Oyster mushrooms	10
Figures 2. Metabolic pathway of tryptophan	12
Figures 3. Chemical structure of tryptophan.....	14
Figures 4. The main pathway for the biosynthesis of aromatic compounds	15
Figures 5. Cavitation bubble collapse and plant material releasing.....	17
Figures 6. Oyster mushrooms studied.....	30
Figures 7. Chromatogram of tryptophan and its derivatives.....	39
Figures 8. Effect of type of solvent and solvent composition.....	41
Figures 9. Pareto chart of factors affecting the extraction conditions.....	43
Figures 10. Response surface plots	46
Figures 11. Effect of extraction time.....	48
Figures 12. Effect of extraction time on the stability.....	50



UNIVERSITAS
GADJAH MADA

**OPTIMIZATION OF ULTRASOUND-ASSISTED EXTRACTION OF TRYPTOPHAN AND ITS
DERIVATIVES FROM OYSTER
MUSHROOM (*Pleurotus spp.*) THROUGH A RESPONSE SURFACE METHODOLOGY**

Anita Nurmulya Bahari, Dr. Widiastuti Setyaningsih, S.T.P., M.Sc.

Universitas Gadjah Mada, 2025 | Diunduh dari <http://etd.repository.ugm.ac.id/>

LIST OF APPENDIXES

Appendix 1	Calibration curve	73
Appendix 2	Design of experiment of BBD.....	80
Appendix 3	ANOVA of Response Area	81
Appendix 4	Assesment of extraction time.	83
Appendix 5	Assesment of extraction time on stability	84
Appendix 6	Accuration and precision	84
Appendix 7	Identification proof of sample.	85