TABLE OF CONTENTS

Contents		Page No.
Certificate		i
Declaration	by the Candidate & Certificate by the Supervisor	ii
Copyright T	ransfer Certificate	iii
Dedication		iv
Acknowledge	ements	v-vii
Table of Contents		viii-xi
List of Figures		xii-xv
List of Table	s	xvi-xvii
List of Symb	ols/Abbreviation	xviii-xx
Preface		xxi-xxiv
CHAPTER 1	: INTRODUCTION	1
1.1 Microb	vial Fuel cell for Azo dyes treatment	10
1.2 Dye de	gradation mechanism in microbial fuel cell (MFC)	12
1.3 Effects	of operational parameters on dye removal efficiency in MFC	13
1.4 Problem	m statement	16
1.5 Signifi	cance of research	17
CHAPTER 2	2: LITERATURE REVIEWS	18
2.1 Dyes and their history		18
2.1.1	Classification of dyes	19
2.1.2	Potential threats of Azo dyes to the environment and their	29
	treatment technology	
2.1.3	Current treatment technology for dyes degradation	33
2.2 Microbial fuel cell and its history		43
2.2.1	Working principle of MFCs	50
2.2.2	Metabolism in MFCs	53
2.2.3	Current research on MFCs	55
2.2.4	Types of MFCs	58
2.2.5	Parameter affecting the performance of MFCs	62

2.2.6	Limitation of MFCs	65
2.2.7	Research Gap	65
2.2.8	Research Objective	66
CHAPTER 3	: MATERIALS & METHODS	67
3.1 Dye	legrading bacteria isolation, and their study for dye	67
decolo	orizing capacity	
3.1.1	Sample selection for isolation of bacteria	67
3.1.2	Dye selection	67
3.1.3	Media used for the growth of Culture	71
3.1.4	Screening and identification of the isolated organism	72
3.1.5	Study for dye decolorizing capacity	73
3.2 MFC setup construction		74
3.2.1	MFC 1	75
3.2.2	MFC 2	76
3.3 Analy	tical Procedure	77
3.3.1	Assessment of dye decolorization in batch study and in MFC	77
3.3.2	COD Removal efficiency	78
3.3.3	Dry biomass estimation	78
3.3.4	CO ₂ measurement	79
3.3.5	CFU estimations	79
3.3.6	Fourier-transform infrared spectroscopy (FTIR) analysis	79
3.3.7	Identification of metabolites using Liquid chromatography and	80
	mass spectroscopy analysis (LCMS)	
3.3.8	SEM analysis	80
3.3.9	Electrochemical monitoring	80
3.4 Kineti	c Study	81
3.5 Statist	ical Modeling for the analysis of dye decolorization &	82
current density though Response surface methodology (RSM)		
3.6 Phytotoxicity Studies		83

RESULTS & DISCUSSIONS		
CHAPTER 4.		84
4.1 Dve degrading bacteria is	olation & screening and their batch	84
study for dye decolorizing c	anacity	01
4.1.1 Screening and Isolatic	apacity	84
4.1.2 The Identification of I	coloted Microorganisms	0 1 0 <i>5</i>
4.1.2 The Identification of I	solated Microorganisms	85
4.1.3 Dye Decolorization S	tudies	88
4.1.4 Concluding remarks		94
CHAPTER 5:		95
5.1 A comparative study of a	bio fuel cell with two different proton	95
exchange membrane for th	e production of electricity from waste	
water		
5.1.1 Experiment design		95
5.1.2 Results & discussion	s	95
5.1.3 Concluding remarks		100
CHAPTER 6:		101
6.1 Biodegradation of Reactive	Orange 16 Dye in Microbial Fuel Cell:	101
An Innovative Way to Mi	nimize Waste Along with Electricity	
Production		
6.1.1 Experiment design		101
6.1.2 Results & discussions		101
6.1.3 Concluding remarks		112
CHAPTER 7:		113
7.1 Biodegradation of reactive	red 120 in microbial fuel cell by	113
Staphylococcus equoruma RA	P1: Statistical modelling and process	
optimization		
7.1.1 Experiment design		113
7.1.2 Results & discussions		113
7.1.3 Concluding remarks		137
CHAPTER 8:		138
8.1 A comparative study on the j	performance of Microbial Fuel Cell for	138

the treatment of Reactive Orange 16 dye using mixed and pure	
bacterial species and its optimization using response surface	
methodology	
8.1.1 Experiment design	138
8.1.2 Results & discussions	138
8.1.3 Concluding remarks	162
CHAPTER 9:	163
9.1 CONCLUSIONS	
9.2 FUTURE RESEARCH PROSPECTIVE	
REFERENCES	
APPENDIX	
LIST OF PUBLICATIONS	
CONFERENCES	

LIDU UL LIGULUD

Figure No.	Figure Captions	Page No.
Figure 1.1	Sources of dye wastewater	1
Figure 1.2	Wastewater discharge into the river	3
Figure 1.3	Mechanism of waste remediation and electricity production	6
Figure 1.4	Dye remediation in MFC	7
Figure 1.5	Mechanism of waste degradation in MFC	13
Figure 2.1	Effects of dye wastewater	30
Figure 2.2	Dye treatment technology	34
Figure 2.3	Mechanism of electron transport to the anode surface	51
Figure 2.4	Electron transport mechanism from bacterial metabolism to	52
	the surface of the anode	
Figure 2.5	Metabolism of a substrate (waste) in MFC	54
Figure 2.6	Schematics of single-chambered MFC	59
Figure 2.7	Schematics of double-chambered MFC	60
Figure 2.8	Schematics of Up-flow MFC	61
Figure 3.1	Calibration plot for model dyes (a) Reactive orange 16 (b)	70
	Reactive red 120	
Figure 3.2	MFC 1	76
Figure 3.3	MFC 2	77
Figure 4.1	Isolated strains	84
Figure 4.2 (a)	SEM images of Acinetobacter pitii at 20 K X on the surface	86
	of cellulose filter 0.2 microns, 4.2	
Figure 4.2 (b)	SEM images of cellulose filter 0.2 microns with growth	86
	media without bacteria at 20KX	
Figure 4.3	Phylogenic tree of the bacterial isolate (Acinetobacter pitii)	86
Figure 4.4 (a)	SEM images of <i>Staphylococcus equorum</i> ta at 20 K X on	87
	the surface of cellulose filter 0.2 microns	
Figure 4.4 (b)	SEM images of cellulose filter 0.2 microns with growth	87
	media without bacteria at 20KX	
Figure 4.5	Phylogenic tree of the bacterial isolate (Staphylococcus	88
	equorum RAP1)	

Figure 4.6	Study on isolated strains for (a) Staphylococcus eqrum and	89
	(b) Acinetobacter pitti degradation of RO16 dye and RR120	
	Dye in batch and MFC for 100 ppm initial concentration of	
	dyes	
Figure 4.7	Study on isolated strains for (a) Staphylococcus eqrum and	91
	(b) Acinetobacter pitti degradation of RO16 dye and RR120	
	Dye in batch and MFC for 100 ppm initial concentration of	
	dyes at 72 h	
Figure 4.8	Study on isolated strains for (a) Staphylococcus eqrum and	93
	(b) Acinetobacter pitti degradation of RO16 dye and	
	RR120 Dye in batch and MFC for 100 ppm initial	
	concentration of dyes at 72 h.	
Figure 5.1	Comparison of voltage produced by Nafion as separator and	96
	agar salt bridge as a separator	
Figure 5.2	Comparison of current density produced by Nafion as	97
	separator and agar salt bridge as a separator	
Figure 5.3	Polarization curve for Nafion and agar salt bridge as PEM	99
Figure 6.1(a)	Variation of voltage and current over time and	102
Figure 6.1(b)	Variation of voltage and Colony-forming unit with time	102
Figure 6.2	Relation between power density and COD removal over	103
	time	
Figure 6.3	Variation of % color removal and bacterial growth with	104
	dye concentration	
Figure 6.4	Relation between COD Removal Vs Dye Concentration	107
Figure 6.5	Relation between CO2 production and Dye concentration	109
Figure 6.6	Monod Equation fitted till 400 ppm of dye (obtained	110
	experimental value)	
Figure 7.1(a)	voltage variation with time at different pH for 100 ppm dye	115
Figure 7.1 (b)	voltage variation with time at pH 7.0 for different	115
	concentration of dye	
Figure 7.1 (c)	power density variation with time at different pH for 100	116
	ppm dye	

Figure 7.1 (d)	power density variation with time at pH 7.0 for different	116
	concentration of dye	
Figure 7.2 (a)	Degradation of dye at different pH at concentration range of	118
	100 to 300 ppm	
Figure 7.2 (b)	% Degradation of dye at different pH at concentration range	118
	of 100 to 300 ppm	
Figure 7.3	COD removal efficiency	119
Figure 7.4	Actual vs Predicted value of RR120 dye	121
Figure 7.5 (a)	2D Contour plot for the degradation of RR120 dye as a	124
	function of the concentration of dye and pH	
Figure 7.5 (b)	3D Response surface plot for the degradation of RR120 dye	124
	as a function of the concentration of dye and pH	
Figure 7.5 (c)	2D Contour plot for the degradation of RR120 dye as a	125
	function of time and pH	
Figure 7.5 (d)	3D Response surface plot for the degradation of RR120 dye	125
	as a function of time and pH	
Figure 7.5 (e)	2D Contour plot for the degradation of RR120 dye as a	126
	function of time and concentration of dye	
Figure 7.5 (f)	3D Response surface plot for the degradation of RR120 dye	126
	as a function of time and concentration of dye	
Figure 7.6	Predicted Vs actual value of current density	127
Figure 7.7 (a)	2D Contour plot for current density as a function of	129
	concentration and pH	
Figure 7.7 (b)	3D Response surface plot for current density as a function	129
	of concentration and pH	
Figure 7.7 (c)	2D Contour plot for current density as a function of time	130
	and pH	
Figure 7.7 (d)	3D Response surface plot for current density as a function	130
	of time and pH	
Figure 7.7 (e)	2D Contour plot for current density as a function of time	131
	and concentration	
Figure 7.7 (f)	3D Response surface plot for current density as a function	131
	of time and concentration	

Figure 7.8	FTIR spectra of RR120 dye (Control) and (b) FTIR spectra	133
	of degraded metabolites	
Figure 7.9	Proposed mechanism of biodegradation of RR120 dye by Staphylococcus equorum RAP1	135
Figure 7.10	Phytotoxicity effect of treated dye samples using	136
	Staphylococcus equrumin MFC	
Figure 8.1 (a)	Voltage variation along with the time of mixed and isolated	140
	species	
Figure 8.1 (b)	Effect of CFU/ml on Voltage.	140
Figure8.2	COD removal efficiency	142
Figure 8.3	Variation of color removal efficiency with time	143
Figure 8.4	CO_2 variation with time	144
		144
Figure 8.5	predicted vs actual data of dye degraded	147
Figure 8.6 (a, b)	Dye degradation as function of pH and concentration	148
Figure 8.6 (c, d)	Dye degradation as function of pH and time	149
Figure 8.6 (e, f)	Dye degradation as function of time and concentration	150
Figure 8.7	Predicted vs actual data of current density	152
Figure 8.8 (a,b)	current density as function of pH and concentration,	153
Figure 8.8 (c,d)	current density as function of pH and time	154
Figure 8.8 (e,f)	current density as function of time and concentration	155
Figure 8.9 (a)	FTIR spectrums of control reactive orange 16	158
Figure 8.9 (b)	decolorized reactive orange 16	158
Figure 8.10	Proposed biodegradation pathway for reactive orange 16	160
	degradation by Acinetobacter pitii	
Figure 8.11	Phytotoxicity study	161

List of Tables

Table No.	Table Captions	Page No.
Table 1.1	Electricity production using Dyes	11
Table 2.1	Classification of dyes based on chromophore group	21
Table 2.2	Examples of dyes based on affinity towards material	24
Table 2.3	Examples Based on application methods	28
Table 2.4	Characteristics of textile industry effluent	31
Table 2.5	International standard of dye effluent discharge into the	32
	environment	
Table 2.6	Advantages and disadvantages of various physical and chemical	38
	methods	
Table 2.7	Work done by other researchers in MFC	44
Table 3.1	Waste water characteristics used for MFC	67
Table 3.2	Physicochemical Characteristics of Dyes	68
Table 3.3	Medium specific to Geobacter sp. (For1 L).	71
Table 3.4	Experimental Design for the degradation of RR120	83
Table 3.5	Experimental Design for the degradation of RO 16	83
Table 4.1	Decolorization efficiency of all the isolated strains for RO 16 and	85
	RR 120 dyes for 100 ppm for 24 h.	
Table 6.1	Electricity production using dyes	105
Table 6.2	Haldane kinetic parameter for different dyes	111
Table 7.1	Design table for Face Centred Central Composite design for	119
	MFC	
Table 7.2	Analysis of variance (ANOVA) for the degradation of RR120 by <i>Staphylococcus equorum</i> RAP1 terms of concentration of dye	122
	degraded	
Table 7.3	Analysis of variance (ANOVA) for the degradation of RR120 by	128
	Staphylococcus equorum RAP1 terms of current density	
Table 7.4	Optimized numerical solutions for degradation of dye and current	132
	density	
Table 8.1	Design table for Face Centred Central Composite design for MFC	144

Table 8.2	Analysis of variance (ANOVA) for the degradation of RO 16 by Acinetobacter pitii terms of Concentration of dye degraded	146
Table 8.3	Analysis of variance (ANOVA) for the degradation of RO 16 by Acinetobacter pitii in terms of Current density	151
Table 8.4	Optimized numerical solutions for degradation of dye and current	156