

# **Proof of principle of the first application of a hydroxyl-functionalized ionic liquid for the reactive extraction of carboxylic acids**

by

**EDWARD TATENDA MARONDEDZE**

Submitted in fulfilment of the academic

requirements for the degree of

Doctor of Philosophy in the

School of Chemistry and Physics,

University of KwaZulu-Natal,

Westville Campus

Durban

February 2023

## Abstract

The production of value-added chemicals through fermentation has been practised for decades. The productivity of fermentation systems is hindered by end-product inhibition. A compromise always must be drawn with respect to the environmental and economic sustainability of the separation and recovery technologies. Drawbacks in the industrial applicability of fermentation to produce alcohols and carboxylic acids have led to intensive research towards extractive fermentation processes. Reactive extraction is a comparatively new technology that can be applied for extractive fermentation. The reactive extraction principle is based on the occurrence of a reversible chemical reaction between the solute and an extractant, which facilitates the distribution of the solute between two immiscible phases. The availability of cheap, effective, biocompatible, and environmentally sustainable extractants remain a challenge.

Ionic liquids (ILs) have been identified as designer solvents, and therefore this study focuses on the preparation, characterization, and application of a customized IL in the reactive extraction of carboxylic acids from aqueous solution. The application of 1-(6-hydroxyhexyl)-3-methylimidazolium bis(trifluoromethanesulfonyl)imide, (IL-OH) as a reactive extractant for carboxylic acids from aqueous solution was investigated. It is postulated that during the reactive extraction of carboxylic acids with IL-OH, the reactive extraction is driven by an acid-catalysed esterification reaction, which in turn produces 1-(hexyl-4-oxopentanoate)-3-methylimidazolium bis(trifluoromethanesulfonyl)imide (IL-Ester).

The behaviour of the IL-OH and IL-Ester in the presence of an aqueous environment was investigated to determine phase miscibility. The effect of the initial pH of the aqueous phase as well as the initial amount of acid catalyst (methanesulfonic acid) present on the miscibility of IL-OH and water were investigated. The aqueous solubility of IL-OH in the pH range 1.0 – 7.5 was nearly constant around  $2.7 \text{ g L}^{-1}$ . As the initial amount of methanesulfonic acid was increased within the range 1 – 12 wt.%, the solubility of IL-OH in water slightly fluctuates around  $2.4 \text{ g L}^{-1}$ . On the other hand, approximately 56 - 62  $\text{mg g}^{-1}$  of water dissolved in the IL, irrespective of the pH of the aqueous phase or the initial amount of methanesulfonic acid present. A similar trend was observed with respect to IL-Ester.

The reactive extraction of carboxylic acids with IL-OH was investigated using fumaric and levulinic acids as model carboxylic acids. The initial concentration of fumaric and levulinic acid was in the range 0.003 – 0.034 and 0.059 – 0.323 M, respectively. The distribution coefficient for fumaric and levulinic acid were in the ranges 2.7 – 3.9 and 11.3 – 29.9, respectively. Similarly, the extraction efficiencies for fumaric acid and levulinic acid were in the ranges 73.2 – 79.9% and 91.9 – 96.8%, respectively.

The extracted carboxylic acid was recovered from the IL phase by means of an acid-catalysed transesterification reaction. The effects of reactants' molar ratio (1:3 and 1:10), reaction temperature (85 – 115 °C), and catalyst loading (0 – 5 wt.%) on the transesterification of IL-Ester with butanol were investigated. An optimal conversion (92.29%) of IL-Ester to butyl levulinate and IL-OH was achieved at 85 °C, using an IL-Ester-to-butanol molar ratio of 1:10 in the presence of 1 wt.% methanesulfonic acid as a catalyst. An isolation protocol for the recovery of butyl levulinate from the transesterification mixture was developed. At least 95% of the excess butanol was recovered from the reaction mixture by means of vacuum distillation.

The regenerated IL-OH was characterized by means of FTIR spectroscopy, <sup>1</sup>H NMR, and <sup>19</sup>F NMR spectrometry. Furthermore, the composition of the regenerated IL-OH was determined with respect to both the cation and anion. The cations in the regenerated IL-OH were identified and quantified by means of HPLC. The anion was identified and quantified by means of <sup>19</sup>F NMR spectrometry. It was found that the sum of IL cations determined from HPLC analysis was consistent and the amount of the [NTf<sub>2</sub>]<sup>−</sup> determined from <sup>19</sup>F NMR.

The environmental fate of chemical compounds can be estimated based on the octanol-water partition coefficient (K<sub>ow</sub>) of the compounds. K<sub>ow</sub> values for IL-OH and IL-Ester were measured experimentally, and were found to be 0.0194 and 0.2059, respectively. It can be concluded that presence of IL-Ester and IL-OH in the environment present low risk of bioaccumulation of the respective species.

Data derived from this study will serve as a basis for the design of IL-based reactive extraction systems, incorporating minor modifications, adjustments, and optimizations where necessary.

## **Preface**

The experimental work described in this thesis was carried out at the University of KwaZulu-Natal in the College of Agriculture, Engineering and Science (CAES) at the Department of Chemical Engineering, Howard College Campus, and in the School of Chemistry and Physics, Westville Campus, Durban, from April 2019 to January 2023, under the supervision of Professors Annegret Stark and Bice Martincigh.

These studies represent original work by the author and have not otherwise been submitted in any form for any degree or diploma to any tertiary institution. Where use has been made of the work of others it is duly acknowledged in the text.

Name: Edward Tatenda Marondedze

Signed:



Date: 7 February 2023

## Declaration 1 - Plagiarism

I, Edward Tatenda Marondedze, declare that

1. The research reported in this thesis, except where otherwise indicated, is my original research.
2. This thesis has not been submitted for any degree or examination at any other university.
3. This thesis does not contain other person's data, pictures, graphs, or other information, unless specifically acknowledged as being sourced from other persons.
4. This thesis does not contain other person's writing, unless specifically acknowledged as being sourced from other researchers. Where other written sources have been quoted then:
  - a. Their words have been re-written, but the general information attributed to them has been referenced.
  - b. Where their exact words have been used, then their writing has been placed in italics and inside quotation marks and referenced.
5. This thesis does not contain text, graphics or tables copied and pasted from the Internet, unless specifically acknowledged, and the source being detailed in the thesis and references sections.

Name: Edward Tatenda Marondedze

Signed:

A black rectangular box used to redact the signature of the author.

Date: 7 February 2023

## **Declaration 2 – Conferences, presentations, and publications**

1. **E.T. Marondedze**, B.S. Martincigh and A. Stark, *The fate of a hydrophobic ionic liquid in an aqueous environment*, College of Agriculture, Engineering and Science (CAES) Research and Innovation Day, UKZN, Durban 10 – 11 December 2020. (**Virtual Poster**).

## Acknowledgements

I will always honour the golden opportunity that got me working under the mentorship of Professors Annegret Stark and Bice Martincigh. Over the years I have learnt a lot from them, as well as through them. Most of all, they have allowed me to learn through myself. My supervisors headed two very diverse research groups which eventually grew to be my campus community.

*Grazie Prof BSM per la sua guida.*

*Vielen dank für ihre anleitung Prof AS.*

The priceless technical support I received from Preyothan Nayager, Thobekile Mofokeng and Xoli Hadebe will forever be appreciated.

To Josiane Ayingeneye, thank you for the technical and moral support.

I am grateful for all the support I received from my family, Ephraim, Rosemary, and Oswald Marondedze thank you for being there.

Ellen Mutsakatira, thank you, I really appreciate the support.

Obert Mupomoki, Principal Mdolo, Dr Phakamile Ndlovu, and Dr Paul Zwawanda you helped me in so many ways, more than you can imagine.

I appreciate the financial support received through the SMRI/NRF SARChI Research Chair for Sugarcane Biorefining at the University of KwaZulu-Natal.

The financial assistance of the National Research Foundation (NRF) toward this research is hereby acknowledged. Opinions expressed and conclusions arrived at, are those of the author and are not necessarily to be attributed to the NRF.

## **Dedication**

For my parents.

I thought it would be easy!



## Table of contents

Abstract.....	i
Preface .....	iii
Declaration 1 - Plagiarism .....	iv
Declaration 2 – Conferences, presentations, and publications .....	v
Acknowledgements .....	vi
Dedication.....	vii
Table of contents .....	viii
List of figures .....	xiii
List of tables .....	xvii
List of schemes .....	xix
List of abbreviations .....	xx
1 Introduction .....	1
1.1 Recovery of carboxylic acids from aqueous solution .....	1
1.2 Reactive extraction of carboxylic acids from aqueous solutions.....	3
1.2.1 Categorization of extraction concepts .....	3
1.2.2 Limitations in the reactive extraction of carboxylic acids.....	7
1.2.3 Application of ionic liquids in the extraction of carboxylic acids.....	7
1.3 Hypothesis .....	11
1.4 Aims and objectives.....	12
1.5 Statement of novelty .....	13
1.6 Thesis structure .....	15
2 Literature review.....	16
2.1 The biorefinery concept .....	16
2.2 Product portfolio for biorefineries .....	17

2.3	Production of carboxylic acids via fermentation .....	19
2.4	Carboxylic acid market value .....	20
2.5	Recovery of carboxylic acids from aqueous solution .....	21
2.6	ILs as designer solvents .....	24
2.6.1	Classification and preparation of ILs .....	24
2.6.2	Examples of cation-functionalized ILs.....	27
2.6.3	Environmental impact of ILs .....	29
2.7	Consideration of solubility aspects of reactive extractants.....	31
2.8	Reactive extraction of carboxylic acids via chemical reaction.....	34
2.9	The esterification reaction .....	35
2.10	Regeneration of the reactive extractant.....	37
2.10.1	Temperature.....	37
2.10.2	Diluent .....	38
2.11	Transesterification reaction .....	39
2.12	Chapter summary .....	40
3	Experimental.....	42
3.1	Materials and instruments .....	42
3.2	Preparation of ILs .....	43
3.2.1	Alkylation reaction .....	44
3.2.2	Ion-exchange reaction.....	45
3.2.3	Esterification reaction.....	46
3.3	Instrumental methods.....	47
3.3.1	HPLC method.....	47
3.3.1.1	Calibration .....	47
3.3.1.2	Quality assurance.....	48
3.3.2	GC method.....	48

3.3.2.1	GC calibration .....	49
3.3.2.2	Quality assurance.....	49
3.3.3	Karl-Fischer titration .....	50
3.3.4	Nuclear magnetic resonance spectrometry .....	50
3.3.5	FTIR spectroscopy.....	50
3.4	Mutual solubility studies.....	51
3.4.1	Effect of pH on mutual solubility .....	51
3.4.2	Effect of methanesulfonic acid concentration on mutual solubility .....	51
3.5	Octanol-water partition coefficient measurements .....	52
3.6	Extraction of carboxylic acids using IL-OH.....	52
3.6.1	Preparation of levulinic acid solutions .....	53
3.6.2	Preparation of fumaric acid solutions.....	53
3.6.3	Physical extraction.....	53
3.6.4	Reactive extraction .....	53
3.7	Transesterification of IL-Ester with butanol.....	54
3.7.1	Transesterification procedure .....	54
3.7.1.1	Effect of reactant molar ratio on the transesterification reaction .....	54
3.7.1.2	Effect of reaction temperature on transesterification .....	55
3.7.1.3	Effect of catalyst loading on the transesterification reaction .....	55
3.7.2	Sample handling and analysis.....	56
3.7.3	Purification of the reaction mixture.....	56
3.8	Chapter summary .....	57
4	Results and Discussion .....	58
4.1	Preparation of ILs .....	58
4.2	HPLC method development.....	59
4.3	GC-FID method development .....	61

4.4	Mutual solubility of IL-OH and IL-Ester with water.....	63
4.4.1	Effect of pH on mutual solubility .....	63
4.4.2	Effect of acid catalyst concentration on the aqueous solubility of IL-OH.....	67
4.5	Octanol-water partitioning .....	69
4.6	Extraction of carboxylic acids from aqueous solution using IL-OH .....	73
4.7	Transesterification of IL-Ester with butanol, and regeneration of IL-OH.....	81
4.7.1	Factors affecting the transesterification of IL-Ester with butanol.....	81
4.7.1.1	Effect of reactant molar ratio on the transesterification reaction .....	81
4.7.1.2	Effect of temperature on transesterification reaction .....	82
4.7.1.3	Effect of catalyst loading on the transesterification reaction .....	82
4.7.2	Product separation and regeneration of IL-OH .....	83
4.7.2.1	Removal of butanol by distillation .....	83
4.7.2.2	Recovery of butyl levulinate .....	85
4.7.2.3	Regenerated IL-OH .....	86
4.8	Comparison with common extractants .....	90
4.9	Process configuration and preliminary mass balances for reactive extraction of carboxylic acids .....	91
4.10	Chapter summary .....	98
5	Conclusions, outlook, and future work.....	100
5.1	Conclusions.....	100
5.1.1	Application of IL-OH as a reactive extractant.....	100
5.1.2	Isolation of extracted carboxylic acid and regeneration of IL-OH.....	101
5.1.3	Environmental impact of IL-OH and IL-Ester .....	102
5.1.4	Process design and configuration .....	102
5.2	Outlook .....	103
5.3	Future work.....	103

References .....	105
Appendix A: HPLC data .....	118
Appendix B: GC data .....	123
Appendix C: Extraction data .....	125
Appendix D: Group contribution data .....	127
Appendix E: NMR spectra .....	131

## List of figures

Figure 1.1: Examples of inert extractants.....	4
Figure 1.2: Chemical structure of tributyl phosphate.....	4
Figure 1.3: Examples of tertiary amines used for the extraction of carboxylic acids from aqueous solution.....	5
Figure 1.4: Chemical structure of Cyphos IL-104.....	8
Figure 1.5: Examples of functionalized IL cations, where R, FG, and n are the alkyl/aryl substituents, the specific functional group, and the chain length, respectively.....	11
Figure 2.1: Examples of value-added chemicals derived from biorefinery processes...	18
Figure 2.2: Examples of value-added chemicals derived from levulinic acid. ....	19
Figure 2.3: Chemical structures of some of the cations used in the design of ILs, where R <sub>i</sub> represents any alkyl or aryl substituent.....	26
Figure 2.4: Examples of anions used in the design of ILs.....	27
Figure 4.1: <sup>19</sup> F NMR spectrum of IL-Ester and CF <sub>3</sub> CO <sub>2</sub> Na in acetonitrile-d <sub>3</sub> , with CF <sub>3</sub> CO <sub>2</sub> Na acting as the internal standard.....	59
Figure 4.2: Sample chromatogram for the determination of IL-Ester and IL-OH by means of HPLC. IL-Ester and IL-OH were eluted from the column at 2.339 and 3.354 minutes, respectively. The solvent front eluted at t = 1.0 minutes.....	60
Figure 4.3: GC-FID sample chromatogram showing the elution of butanol, ethyl levulinate and butyl levulinate from a Zebron ZB-WAXPLUS column at 4.035, 8.5884, and 10.334 minutes, respectively. ....	62
Figure 4.4: Effect of the initial pH of the aqueous phase on the amount of water dissolved in the IL-OH at 25 °C (n = 3). ....	64
Figure 4.5: Effect of the initial pH of the aqueous phase on the solubility of IL-OH in water at 25 °C. ....	65
Figure 4.6: Effect of the initial pH of the aqueous phase on the amount of water dissolved in the IL-Ester at 25 °C (n = 3).....	66
Figure 4.7: Effect of the initial pH of the aqueous phase on the solubility of IL-Ester in water at 25 °C. ....	66
Figure 4.8: Solubility of water in IL-OH as a function of initial methanesulfonic acid concentration in the aqueous phase at 25 °C (n = 3). ....	67

Figure 4.9: Effect of the initial amount of acid catalyst on the aqueous solubility of IL-OH at 25 °C. ....	68
Figure 4.10: Equilibrium concentrations of IL-OH and IL-Ester in the octanol and aqueous phases at 25 °C. ....	70
Figure 4.11: Effect of initial IL concentration on $K_{ow}$ at 25 °C. ....	72
Figure 4.12: Solubility of water in the IL phase during the extraction of fumaric acid. ....	74
Figure 4.13: Solubility of water in the IL phase during the extraction of levulinic acid. ....	74
Figure 4.14: Effect of initial concentration of fumaric acid on the aqueous solubility of IL-OH during the extraction of fumaric acid with IL-OH at 25 °C. ....	75
Figure 4.15: Effect of initial concentration of levulinic acid on the aqueous solubility of IL-OH during the extraction of levulinic acid with IL-OH at 25 °C. ....	76
Figure 4.16: Sample HPLC chromatogram showing the elution of levulinic acid and IL-OH at 1.498 and 3.005 minutes, respectively. The analysed sample was taken from the aqueous phase after the physical extraction of levulinic acid using IL-OH at 25 °C. ....	77
Figure 4.17: Effect of water loading on the esterification of IL-OH with levulinic acid in the presence of methanesulfonic acid as a catalyst (1 wt.% with respect to IL-OH) at 70 °C using a 1:1 molar ratio of IL-OH to levulinic acid. ....	78
Figure 4.18: Effect of increasing the initial concentration of fumaric acid on the initial molar ratio of fumaric acid with respect to IL-OH at 25 °C. The reactive extraction experiments were conducted in the presence of 1.0 wt.% methanesulfonic acid as a catalyst. ....	79
Figure 4.19: Effect of increasing the initial concentration of levulinic acid on the molar ratio of levulinic acid with respect to IL-OH at 25 °C. The reactive extraction experiments were conducted in the presence of 1.0 wt.% methanesulfonic acid as a catalyst. ....	79
Figure 4.20: Extraction equilibria of fumaric acid with IL-OH at 25 °C. Experiments were conducted using a 1:1 v/v phase ratio. The reactive extraction was conducted in the presence of 1.0 wt.% methanesulfonic acid as a catalyst. ....	80
Figure 4.21: Extraction equilibria of levulinic acid with IL-OH at 25 °C. Experiments were conducted using a 1:1 v/v phase ratio. The reactive extraction was conducted in the presence of 1.0 wt.% methanesulfonic acid as a catalyst. ....	80

Figure 4.22: Effect of reactant IL-Ester: butanol molar ratio on the transesterification reaction at 85 °C and 1.0 wt.% methanesulfonic acid as a catalyst.....	81
Figure 4.23: Effect of temperature on the transesterification of IL-Ester with butanol in the presence of 1.0 wt.% methanesulfonic acid catalyst and an IL: alcohol ratio of 1:10. ....	82
Figure 4.24: Effect of methanesulfonic acid catalyst loading on the transesterification of IL-Ester with butanol using an IL: alcohol ratio of 1:10 at 85 °C.....	83
Figure 4.25: GC-FID determination of butanol on a Zebron ZB-WAXPLUS capillary column, in the presence of ethyl levulinate as an internal standard and helium as the carrier gas at a flow rate of 1.27 cm <sup>3</sup> min <sup>-1</sup> . ....	84
Figure 4.26: <sup>1</sup> H NMR spectrum for butyl levulinate recovered by solvent extraction in diethyl ether. 10 mg of sample was dissolved in 0.7 cm <sup>3</sup> of acetonitrile-d <sub>3</sub> .....	85
Figure 4.27: FTIR spectrum for butyl levulinate.....	86
Figure 4.28: Sample HPLC chromatogram for the regenerated IL showing IL-OH with traces of IL-Ester eluting at 3.002 and 2.345 minutes, respectively. ....	86
Figure 4.29: Comparison of <sup>1</sup> H NMR spectra for (a) regenerated IL, (b) IL-OH, and (c) IL-Ester, recorded in acetonitrile-d <sub>3</sub> . ....	87
Figure 4.30: <sup>19</sup> F NMR spectra of (a) regenerated IL mixture, (b) IL-OH and (c) IL-Ester recorded in acetonitrile-d <sub>3</sub> . ....	88
Figure 4.31: <sup>19</sup> F NMR spectrum of regenerated IL in the presence of CF <sub>3</sub> CO <sub>2</sub> Na as an internal standard. The spectrum was recorded in acetonitrile-d <sub>3</sub> .....	88
Figure 4.32: Comparison of FTIR spectra for pure IL-OH, IL-Ester, and regenerated IL. ....	89
Figure 4.33: Idealized set-up for the reactive extraction of carboxylic acids using IL-OH. ....	92
Figure 4.34: Overall mass balance for the reactive extraction of levulinic acid from aqueous solution using IL-OH.....	97
Figure A 1: HPLC calibration curve for determination of IL-OH at 211 nm. ....	118
Figure A 2: Residuals plot for the calibration curve for the determination of IL-OH. ....	118
Figure A 3: HPLC calibration curve for determination of IL-Ester at 211 nm. ....	119
Figure A 4: Residuals plot for the calibration curve for the determination of IL-Ester. ....	119



Figure A 5: HPLC calibration curve for determination of levulinic acid at 210 nm....	119
Figure A 6: Residuals plot for the calibration curve for the determination of levulinic acid. .....	120
Figure A 7: HPLC calibration curve for determination of fumaric acid at 211 nm. ....	120
Figure A 8: Residuals plot for the calibration curve for the determination of fumaric acid. .....	120
Figure A 9: Sample chromatogram for the elution of fumaric acid at 1.908 minutes using the HPLC method. ....	121
Figure A 10: Sample chromatogram for the elution of levulinic acid at 1.384 minutes using the HPLC method. ....	121
Figure A 11: Sample chromatogram for the blank injection during the HPLC analysis. The solvent front was eluted at t = 1.137 minutes.....	121
Figure B 1: GC-FID calibration curve for the determination of butanol in the presence of ethyl levulinate as an internal standard.....	123
Figure B 2: Residuals plot for the calibration curve for the determination of butanol using GC-FID.....	123
Figure B 3: GC-FID calibration curve for the determination of butyl levulinate in the presence of ethyl levulinate as an internal standard. ....	124
Figure B 4: Residuals plot for the calibration curve for the determination of butyl levulinate using GC-FID. ....	124
Figure E 1: $^1\text{H}$ NMR spectrum of the regenerated IL in acetonitrile- $\text{d}_3$ .....	131

## List of tables

Table 1.1: Processes used for the recovery of carboxylic acids from aqueous solution. .	2
Table 1.2: Classes of solvents used in reactive extraction of carboxylic acids. ....	6
Table 1.3: Extraction of carboxylic acids using imidazolium based ILs.....	10
Table 2.1: Classification of conversion pathways used in biorefineries [56]......	17
Table 2.2: Examples of carboxylic acids produced via fermentation.....	20
Table 2.3: Comparison of the selling price of levulinic acid and its esters in South Africa. .....	20
Table 2.4: Solvent selection criteria in reactive extraction of carboxylic acids [6, 79].	23
Table 2.5: Classes of IL mixtures.....	24
Table 2.6: Classification of ILs. ....	25
Table 2.7: Octanol-water partition coefficients of typical organic compounds and ILs.	30
Table 2.8: Solubility of selected amines in water at 25 °C. ....	32
Table 3.1: List of chemicals and reagents. ....	42
Table 3.2: List of instruments.....	43
Table 3.3: Purity of ILs synthesized in this study. ....	47
Table 3.4: Temperature programme for GC method.....	49
Table 3.5: Instrument acquisition parameters for FTIR spectroscopy. ....	51
Table 4.1: Sample preparation for quantitative <sup>19</sup> F NMR analysis. ....	58
Table 4.2: HPLC retention times of various analytes at 211 nm.....	60
Table 4.3: Calibration parameters for the HPLC method.....	61
Table 4.4: GC-FID retention times of butanol, ethyl levulinate and butyl levulinate....	62
Table 4.5: Calibration parameters for the GC-FID method.....	62
Table 4.6: Effect of initial pH of the aqueous phase on the solubility of water in IL-OH. .....	64
Table 4.7: Effect of initial IL-Ester concentration on K <sub>OW</sub> . ....	71
Table 4.8: Effect of initial IL-OH concentration on K <sub>OW</sub> .....	71
Table 4.9: Solubilities of fumaric acid and levulinic acid in water at 25 °C.....	73
Table 4.10: Recovery of butanol as a function of reactants' molar ratio. ....	84
Table 4.11: Composition of the regenerated IL determined by HPLC. ....	89
Table 4.12: Physicochemical properties of IL-OH, IL-Ester, TOA and CYPHOS IL-104. .....	91

Table 4.13: Mass balance data for the reactive extraction of levulinic acid stage. ....	94
Table 4.14: Mass balance data for the transesterification stage. ....	95
Table 4.15: Mass balance data for the vacuum distillation stage. ....	95
Table 4.16: Mass balance data for the solvent recovery of butyl levulinate. ....	96
Table A 1: Analytical precision in the determination of IL-OH. ....	122
Table A 2: Analytical precision in the determination of levulinic acid.....	122
Table A 3: Analytical precision in the determination of fumaric acid. ....	122
Table A 4: Analytical precision in the determination of IL-Ester. ....	122
Table B 1: Analytical precision in the determination of butanol (n = 6).....	124
Table B 2: Analytical precision in the determination of butyl levulinate (n = 6).....	124
Table C 1: Physical extraction of fumaric acid with IL-OH at 25 °C. ....	125
Table C 2: Reactive extraction of fumaric acid with IL-OH at 25 °C.....	125
Table C 3:Physical extraction of levulinic acid with IL-OH at 25 °C.....	125
Table C 4: Reactive extraction of levulinic acid with IL-OH at 25 °C. ....	126
Table C 5: Converting Between pH and hydrogen ion concentration.....	126
Table D 1: Atom type description and contributions to obtain $P_{\text{calc}}$ [172]. ....	127
Table D 2: Prediction of density from molar refraction. ....	129

## List of schemes

Scheme 1.1: Neutralization of carboxylic acids with tertiary amines. ....	5
Scheme 2.1: Synthesis of 1-[(6-methacryloyloxy) hexyl]-3-methylimidazolium bromide .....	28
Scheme 2.2: Generalized chemical equation for the acid-catalysed Fischer esterification. .....	35
Scheme 2.3: Acid-catalysed Fischer esterification reaction mechanism.....	36
Scheme 2.4: Generalized chemical equation for the acid-catalysed transesterification reaction, where R, R <sub>1</sub> and R <sub>2</sub> represent different alkyl groups.....	39
Scheme 2.5: Reaction mechanism for the acid-catalysed transesterification [145]. ....	40
Scheme 3.1: Synthesis of functionalized ILs.....	44
Scheme 3.2: Transesterification of IL-Ester with butanol in the presence of methanesulfonic acid as a catalyst.....	56
Scheme 4.1: Synthesis and application of IL-OH in the reactive extraction of levulinic acid. ....	98

## List of abbreviations

Abbreviation	Description
FA	Fumaric acid
FID	Flame ionizing detector
FTIR	Fourier transform infrared
GC	Gas chromatography
HPLC	High performance liquid chromatography
IL	Ionic liquid
IL-Ester	1-(hexyl-4-oxopentanoate)-3-methylimidazolium bis-(trifluoromethanesulfonyl)imide
IL-OH	1-(6-hydroxyhexyl)-3-methylimidazolium bis(trifluoromethanesulfonyl)imide
K <sub>D</sub>	Distribution coefficient
KF	Karl-Fischer
NMR	Nuclear magnetic resonance
LA	Levulinic acid

# **1 Introduction**

Carboxylic acids are a group of organic compounds appended with a carboxyl functional group,  $\text{-COOH}$ . Like any other organic compounds, the physicochemical properties of carboxylic acids are dependent on their molecular structure. The number and position of carboxyl functional groups, presence of other functionalities, and chain length of the carboxylic acid determine the behaviour of individual carboxylic acids. As a result, carboxylic acids find use in a wide range of applications. Carboxylic acids are central ingredients in the food, pharmaceutical, paint, adhesive, preservative, cosmetic, and agriculture industries amongst others.

While carboxylic acids available on the commercial market are often derived from petroleum resources, the drive towards environmental sustainability favours the development and adoption of biochemical pathways in biorefineries for their production. The feed used in biorefineries cover a wide range of renewable materials. Both first-generation and second-generation feeds have been successfully used to produce carboxylic acids [1, 2].

However, the recovery and isolation of carboxylic acids from the fermentation broth remains a major drawback in biorefineries [1-4]. The fermentation broth is a complex mixture of the solute of interest, by-products, and the fermentation microbes. The conventional recovery of the carboxylic acid from the broth often involves a combination of unit operations which include isolation, recovery, concentration, acidification, and purification of the solute. It is estimated that the downstream separation costs range well above 50% of the overall processing costs [1, 2, 5, 6]. Therefore, there is a need for efficient and cost-effective separation protocols for the recovery of carboxylic acids from aqueous solution.

## **1.1 Recovery of carboxylic acids from aqueous solution**

The recovery of carboxylic acids from aqueous solutions is influenced by the type of the carboxylic acid, the concentration of the acid in solution and the composition of the solution from which the acid is to be extracted [1]. As a result, there is no universal method of recovery of carboxylic acids from fermentation broths. Table 1.1 shows examples of some of the common methods used for the recovery of carboxylic acids from

aqueous solution and briefly mentions the operating principle and the limitations of each method.

Table 1.1: Processes used for the recovery of carboxylic acids from aqueous solution.

Process	Concept	Limitations	Ref
Distillation	Recovery of the carboxylic acid from aqueous solution based on differences in volatility from the other components in the matrix.	Carboxylic acids have low volatility and tend to occur at very low concentrations in the broths. Therefore, distillation becomes energy intensive and can possibly result in undesirable product transformations.	[7]
Precipitation	Formation of the insoluble calcium salt of the carboxylic acid by using either calcium carbonate ( $\text{CaCO}_3$ ) or calcium hydroxide ( $\text{Ca(OH)}_2$ ), with the subsequent reacidification of the salt with sulfuric acid, $\text{H}_2\text{SO}_4$ .	The use of $\text{H}_2\text{SO}_4$ to recover the carboxylic acid from the calcium salt leads to the production of an excess of calcium sulfate ( $\text{CaSO}_4$ ) as a solid waste.	[1, 3]
Ion-exchange	Selective adsorption of the undissociated carboxylic acid molecules on ionic resins.	The recovery of the adsorbed acid from the resin requires additional chemicals. In addition, ion-exchange resins are costly.	[7, 8]
Solvent extraction	Distribution of the carboxylic acid between two immiscible phases based on preferential solubility of the solute in the extractant solvent.	Limited transfer of the carboxylic acid from the aqueous phase due to low activity coefficients of the organic acids in water. Requires the use of volatile and toxic solvents.	[9]
Reactive extraction	Recovery of solute from aqueous phase into an immiscible extractant by means of a chemical reaction between the targeted solute and the extractant.	Unavailability of compatible extractants with high performance characteristics. Solvent regeneration is difficult.	[10, 11]

It is evident that there is a need for innovation with regards to separation technologies in biorefineries. Efficient and cost-effective methods of separation in biorefineries will heighten the competitive advantage of bio-based carboxylic acids on the market.

## **1.2 Reactive extraction of carboxylic acids from aqueous solutions**

In general, the applicability of continuous distillation as a method of separation and recovery of substances from solution is limited by technical and economic feasibility. For example, heat sensitive solutes and azeotropic mixtures can be very difficult to purify. In the case of carboxylic acids, high boiling points hamper separation by distillation. As a result, solvent extraction is often employed to compensate for such limitations. The energy consumption associated with solvent extraction followed by distillation to remove the solvent is often lower than direct distillation.

Reactive extraction increases extraction yields with relatively low amounts of extractant by way of enhancing the capacity of the solvent. Furthermore, the chemical reaction enhances the selectivity of the solvent towards the solute. Hence, the application of reactive extraction in the recovery of carboxylic acids from dilute aqueous solutions is a promising technique. Furthermore, reactive extraction can be conducted simultaneously with the reaction to the desired product, which allows for the *in situ* removal of the accumulating product thereby reducing chances of end-product inhibition during fermentation. In the absence of toxic metabolites, such as fermentation products, the productivity of the microbes is significantly enhanced.

Solvent properties are critical to the performance of a reactive extractant. Viscosity, biocompatibility, capacity, and availability are some of the key considerations in the selection criteria for reactive extractants.

### **1.2.1 Categorization of extraction concepts**

The mechanism behind the extraction of carboxylic acids from aqueous solution is dependent on the behaviour of the acid proton towards the solvent (extractant). As a result, solvents used for the extraction of carboxylic acids from aqueous solutions are classified into three distinct categories as described by Kertes and King [3]. The first category of extractants is comprised of inert hydrocarbons, oxygen bearing solvents, as well as substituted hydrocarbons as shown in Figure 1.1. The extractability of the



carboxylic acids by these solvents is dependent on the physical partitioning of the acid between the aqueous phase and the organic solvent.

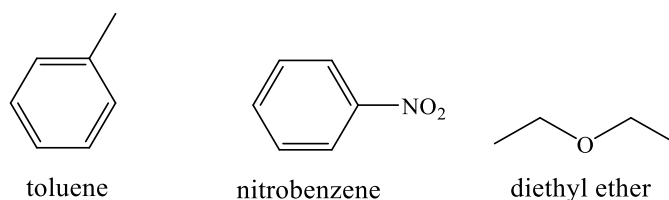


Figure 1.1: Examples of inert extractants.

The second category of extractants is based on organophosphorus solvents such as tributyl phosphate (TBP). These solvents have a phosphorous-bonded oxygen, which is central to the solvation of the carboxylic acids [3]. The chemical structure of TBP is presented in Figure 1.2.

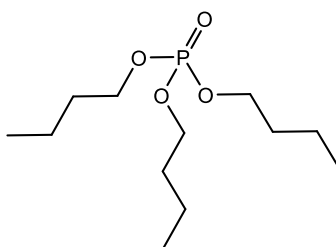


Figure 1.2: Chemical structure of tributyl phosphate.

The extractability of the carboxylic acid from the aqueous phase is dependent on the extent of solvation of the carboxylic acid molecules. For example, Shakya et al. [12] and Firdous and Ahmad [13] reported the use of tributyl phosphate in the recovery of acetic and valeric acid, respectively.

The third category of extractants is based on the extraction due to proton transfer, ion-pair formation and hydrogen bonding between the carboxylic acid and the solvent molecules. Long-chain tertiary amines are usually the preferred choice of solvent. Canari and Eyal [14] investigated the extraction of carboxylic acids by amine-based extractants such as tris(2-ethylhexyl) amine (TEHA), and trioctylamine (TOA). The chemical structures of TEHA and TOA are shown in Figure 1.3. Their study concluded that if the corresponding anion of the extracted carboxylic acid is a weaker base than the tertiary amine used for the extraction, i.e.,  $pK_a(\text{amine}) > pK_a(\text{acid})$ , a protonated amine is formed

due to ion-pair formation. This is an equilibrium reaction, and the extent of proton transfer depends on the difference in the  $pK_a$  values. However, if  $pK_a(\text{amine}) < pK_a(\text{acid})$ , then the contribution of ion-pairing is insignificant, such that the extraction of the carboxylic acid is dependent on the extent of hydrogen bonding between the carboxylic acid and the tertiary amine.

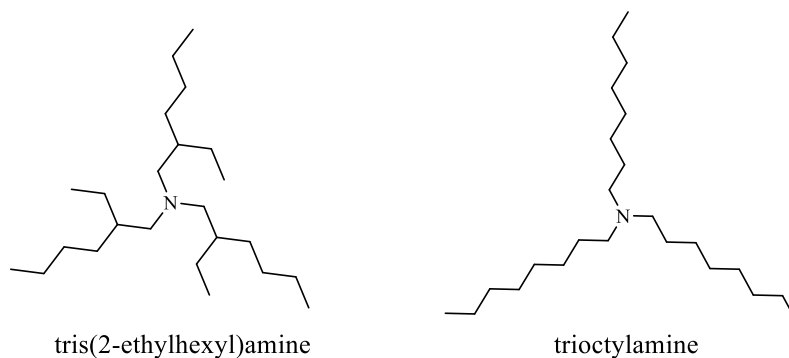
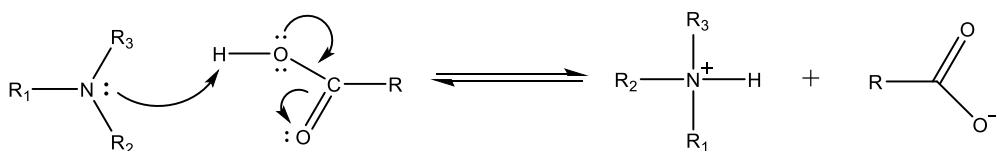


Figure 1.3: Examples of tertiary amines used for the extraction of carboxylic acids from aqueous solution.

The extraction of carboxylic acids by solvents belonging to the first two categories is driven by physical phenomena. However, the extraction of carboxylic acids from aqueous solution by tertiary amines is considered as reactive extraction [3, 15-18]. The extraction mechanism is based on neutralization of the carboxylic acid by the amine. It must be emphasized that just like ammonia, amines are weak bases, and therefore they react with acids to form alkylammonium salts as shown in Scheme 1.1. Therefore, the extraction of carboxylic acids from aqueous solutions with tertiary amines ( $R_3N$ ) is driven by the reversible neutralization equilibrium, with the resulting salt being preferentially soluble in the organic phase.



Scheme 1.1: Neutralization of carboxylic acids with tertiary amines.

Recent reports indicate the successful use of tertiary amines as reactive extractants for carboxylic acids from aqueous solution [15, 19-21]. Kertes and King investigated the

extractability of carboxylic acids with long-chain tertiary aliphatic amines [3]. The extractability of the carboxylic acids was linked to the formation of alkylammonium salts, where there is ion-pair association between the alkylammonium and carboxylate ions.

The high viscosities of tertiary amines tend to complicate dispersion during application, thereby reducing mass transfer efficiency. However, physicochemical properties such as density, viscosity, and surface tension of the organic phase can be modified through use of diluents [22, 23]. Furthermore, diluents tend to stabilize the alkylammonium salt [11].

The diluents used in the extraction of carboxylic acids from aqueous solutions are classified based on polarity [24]. Active diluents are polar solvents, they promote the solvation of the acid-amine complex into the organic phase. Inactive diluents are non-polar solvents with a characteristic poor solvation of the acid-amine complex formed during the extraction. The different classes of diluents are shown in Table 1.2.

Table 1.2: Classes of solvents used in reactive extraction of carboxylic acids.

<b>Active diluents</b>	<b>Inactive diluents</b>
Chlorinated solvents	Alkanes
Alcohols	Benzene
Ketones	Alkyl substituted aromatic solvents
Halogenated aromatic solvents	

Interestingly, a fourth category of extractants can be conceptualised, where the extractant features chemical moieties that undergo reaction with the compound to be extracted, forming more stable covalent bonds than those of the above-mentioned reactive extractions by neutralization. At present, this category has not received much attention in the literature, and to the best of the author's knowledge, the application of this fourth category of extraction has not been reported. Although in principle also a type of reactive extraction, the extent of the reaction is not dependent on the difference in the  $pK_a$  values of the extractant and the corresponding anion of the carboxylic acid. The extent of the reaction is dependent on the equilibrium yield, which can be influenced if, according to Le Chatelier, the co-product of the reaction is removed. In this study, a hydroxyl-functionalized extractant is used, which lends itself ideally for an esterification reaction with the carboxylic acid. As will be shown in this thesis, the co-product of the

esterification, water, is partially removed, because the esterification occurs in a second liquid phase. The carboxylic acid is recovered either as such, or as an alternative ester, by either adding water (ester hydrolysis) or an alcohol (transesterification).

The main advantages of this type of reactive extraction over any of the above-mentioned alternative separation and recovery methods include high capacity, ease of scale-up, high selectivity, reduced downstream processing, adaptability to operating conditions, and reusability of the extractant [6]. In this study, whenever the term “reactive extraction” is used, reference is made to the fourth category of extraction concepts, although certain aspects may also pertain to the third category.

### **1.2.2 Limitations in the reactive extraction of carboxylic acids**

The application of amine-based reactive extraction in the recovery of carboxylic acids from aqueous solution is yet to be adopted at an industrial scale. The development of an efficient extractant system for extractive fermentation is still an area of active research. In 2018, Djas and Henczka reviewed the use of organic solvents and supercritical fluids for the reactive extraction of carboxylic acids from aqueous solution [21]. The review concluded that the use of organic solvents as reactive extractants is mostly limited by the following factors:

- toxicity of the solvents (amines),
- unavailability of biocompatible solvents with high extraction capacity,
- lack of environmentally sustainable solvents, and
- difficulties in the regeneration of the organic phase.

### **1.2.3 Application of ionic liquids in the extraction of carboxylic acids**

The discovery of ionic liquids (ILs) dates back about one century, with the earliest reports from 1914, where Paul von Walden reported ethylammonium nitrate  $[\text{EtNH}_3][\text{NO}_3]$  as a molten salt with a melting point of 12 °C [25]. The definition of ILs has evolved over the years as scientists improved their understanding of the concept [25-28]. In principle, ILs are salts that are comprised of organic cations paired to inorganic anions and the salts have a melting point lower than 100 °C. ILs should not be mistaken for ionic solutions, where a salt is dissolved in a molecular solvent.

The wide liquidus range, negligible vapour pressure, and high thermal stability are some of the remarkable physicochemical properties of ILs [27]. The most investigated groups of ILs include the 1-alkyl-3-methylimidazolium and 1-alkylpyridinium cations. Inserting functionalities such as hydroxyl, amine, ether, ester, carboxyl, among others, within the IL allows for the preparation of functionalized ILs.

The application of ILs in the extraction of carboxylic acids has been reported [29, 30], and in certain cases, the term reactive extraction is used incorrectly, and the extraction mechanism does not involve a chemical reaction in the true sense. For example, Marták and Schlosser [31] proposed a detailed mechanism for the extraction of butyric acid with phosphonium-based ILs such as trihexyl(tetradecyl)phosphonium bis(2,4,4-trimethylpentyl) phosphinate (Cyphos IL-104) in dodecane. As described in Section 1.2.1, Cyphos IL-104 falls in the second category of extractants, and its chemical structure is shown in Figure 1.4. Their investigation highlighted that the extraction of butyric acid occurs via the simultaneous occurrence of multiple mechanisms. Firstly, the butyric acid extracted due to the interactions between butyric acid and dodecane, and this was identified as physical extraction.

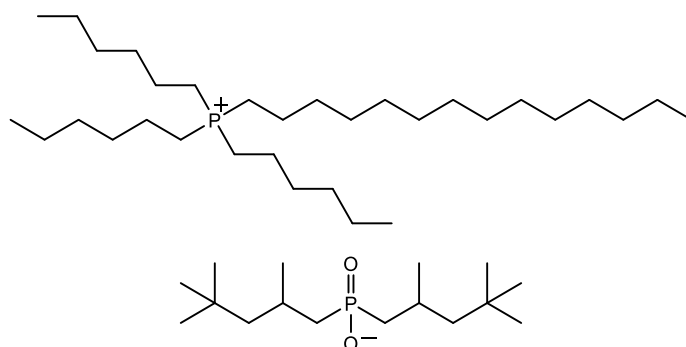


Figure 1.4: Chemical structure of Cyphos IL-104.

Secondly, the solvation of butyric acid molecules with Cyphos IL-104 was considered as competitive reactive extraction. Thirdly, the interactions between the extracted butyric acid and the free butyric acid were considered as non-competitive reactive extraction. The extractability of the carboxylic acid from the aqueous phase into the IL phase is attributed to electrostatic and hydrogen bonding between the IL and the carboxylic acid.

Hence, it is evident that the extractability of butyric acid with Cyphos IL-104 is not driven by a chemical reaction but electrostatic interactions.

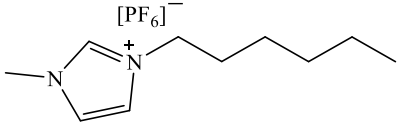
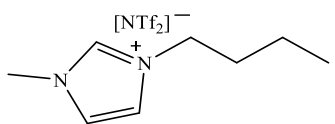
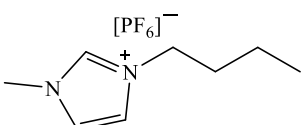
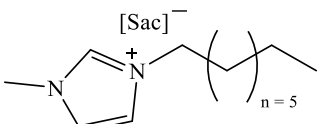
Marták and Schlosser [32] also investigated the extraction of lactic acid from aqueous solution using phosphonium-based ILs, such as Cyphos IL-101, Cyphos IL-104, Cyphos IL-105, and Cyanex 272. At 25 °C, the  $pK_a$  of lactic acid is 3.86 and at low pH values of the aqueous phase, only undissociated lactic acid is present. It was observed that the distribution of lactic acid into the solvent phase decreased significantly with increasing pH, suggesting that the extraction mechanism is driven by hydrogen bond coordination.

Bai et al. [33] investigated the extraction of methacrylic acid from aqueous solution using a series of imidazolium-based ILs with the same cation but different anions, namely  $[BF_4]^-$ ,  $[NTf_2]^-$ , and  $[PF_6]^-$ . It was concluded that the extraction efficiency of the ILs depended on the formation hydrogen bonding between methacrylic acid and the ILs, respectively. The interactions between methacrylic acid and the ILs were systematically studied via FTIR spectroscopy. It was observed that methacrylic acid tends to dimerize and therefore has distinct vibrational band due to the surface deformation of  $OH\cdots O$ . However, in the presence of  $[HMIM][BF_4]$ , the intensity of the marked band decreased with increasing mole ratio of  $[HMIM][BF_4]$  in the system. Increasing the ratio of  $[HMIM][BF_4]$  in the system resulted in a bathochromic shift with respect to in-plane vibrations of  $O=C-O$  and  $O=C-C$ . This shows evidence of hydrogen bonding between  $[HMIM][BF_4]$  and methacrylic acid. Similar interactions are to be expected between 1-alkyl-3-methylimidazolium-based ILs and carboxylic acids. This is because the ILs usually incorporate anions such as  $[BF_4]^-$ ,  $[NTf_2]^-$ , and  $[PF_6]^-$ , which are hydrogen bond acceptors. On the other hand, the carboxyl group of carboxylic acids is a good hydrogen bond donor, thereby promoting hydrogen bond coordination between ILs and the carboxylic acids. As pointed out above, the extraction of carboxylic acids via this mechanism is often incorrectly reported as reactive extraction in literature [33-36].

The application of ILs in the extraction of carboxylic acids from aqueous solution has also been extended to using ILs as diluents [37]. In a study by Ayan et al. [36], 1-hexyl-3-methylimidazolium hexafluorophosphate ( $[HMIM][PF_6]$ ) and 1-hexyl-3-methylimidazolium bis(trifluoromethanesulfonyl)imide ( $[HMIM][NTf_2]$ ) were used as diluents for tributyl phosphate (TBP) in the extraction of propionic acid. In this case, the

extraction mechanism can still be explained based on the solvent categories discussed in Section 1.2.1, and the occurrence of a chemical reaction between the carboxylic acid and the solvent has not been confirmed. Examples of imidazolium-based ILs that have been previously used in the recovery of carboxylic acids from aqueous solution are presented in Table 1.3.

Table 1.3: Extraction of carboxylic acids using imidazolium based ILs.

IL	Structure	Acid extracted	Ref
1-hexyl-3-methylimidazolium hexafluorophosphate		Propionic acid	[36]
1-butyl-3-methylimidazolium bis(trifluoromethanesulfonyl) imide		Levulinic acid	[38]
1-butyl-3-methylimidazolium hexafluorophosphate		Glycolic acid	[39]
1-octyl-3-methylimidazolium saccharinate		Lactic acid	[40]

The introduction of additional functionality in an IL can induce additional properties such as catalytic activity of the IL [41-43]. The ease of preparation of functionalized ILs further enhances the use of ILs as designer solvents [26, 41].

For example, Zeng et al. [44] reported the use of 1-butyl-3-methylimidazolium hydrogen sulfate ([BMIM][HSO<sub>4</sub>]) as a dual solvent and catalyst for the esterification of isobutyric acid with isobutanol to form isobutyl isobutyrate. The Brønsted acidity of [BMIM][HSO<sub>4</sub>] due to the [HSO<sub>4</sub>]<sup>-</sup> anion eliminated the need for a mineral acid catalyst during the esterification reaction. Furthermore, the insolubility of esters in the isobutyl isobutyrate in [BMIM][HSO<sub>4</sub>] allows for a simple recovery of products after the esterification reaction.

The application of ammonium-based ILs as bifunctional media to improve the separation of heavy and light rare earth elements from a nitrate and chloride medium, respectively, are further examples [45].

Functionalization of the anion has been shown to be rather complex and sophisticated compared with functionalizing the cation [46]. This can be attributed to the nature of the chemical reactions involved in the synthetic pathways. Generic examples of functionalized cations are shown in Figure 1.5.

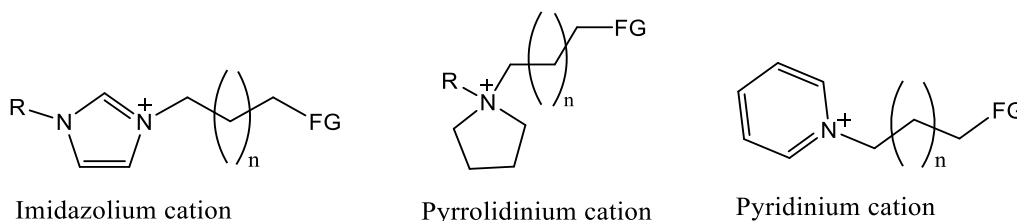


Figure 1.5: Examples of functionalized IL cations, where R, FG, and n are the alkyl/aryl substituents, the specific functional group, and the chain length, respectively.

As stated above, the application of ILs functionalized with a reactive moiety in the reactive extraction of carboxylic acids is yet to be reported. It can be imagined that the functionality of ILs can be customized to incorporate reactive functional groups such as -OH, -COOH, and -NH<sub>2</sub>, capable of reacting with carboxylic acids. In addition, the IL can be designed to eliminate the need for diluents.

### 1.3 Hypothesis

Integrated process units in biorefineries enhance the valorization of biomass through sustainable production of biochemicals. High costs in the downstream recovery and purification of carboxylic acids from solution hinder biochemical production of carboxylic acids at an industrial scale. It is hypothesized that the recovery of carboxylic acids via reactive extraction will enhance productivity while simultaneously reducing the number of unit operations. Reactive extraction is a cost-effective separation technique that leads to process intensification.

The use of conventional molecular solvents as reactive extractants (of the third category) in extractive fermentation of carboxylic acids has been reported [47-49]. However, molecular solvents are highly viscous, corrosive, toxic and volatile in nature. Furthermore, the use additional organic solvents as diluents to alter the physicochemical properties of the organic phase is unavoidable. The toxicity of the organic solvent towards the microorganisms in the fermentation broth is still a major concern. The degree



of toxicity is entirely dependent on the combination of the microorganism and the solvent system. The toxicity of the organic phase can be attributed to either extractant or diluent, and sometimes it is a combination of both.

The discovery of ILs has offered almost unlimited alternatives in solvent engineering. The design and synthesis of functionalized ILs has further increased the specificity, versatility, and robustness of ILs. Therefore, it is possible to design and synthesize an IL which can satisfy the solvent selection criteria for reactive extraction without the need for additives to enhance and optimize its performance.

The foundation of this study lies in the idea that functionalizing the cation of a hydrophobic imidazolium IL with a hydroxyl moiety allows the IL to react with a carboxylic acid in an esterification reaction thereby extracting the carboxylic acid from aqueous solution. Thus, the reactive extraction of the carboxylic acid from aqueous solution will specifically be driven by the esterification reaction. A transesterification reaction will ensure the recovery and isolation of the carboxylic acid from the ester while simultaneously regenerating the IL. The carboxylic acid will be isolated in the form of a value-added ester of choice.

## **1.4 Aims and objectives**

The current study aims to demonstrate a proof of concept for the successful use of a hydroxyl-functionalized (abbreviated IL-OH herein) ionic liquid as a reactive extractant for carboxylic acids from aqueous solution. To explore the aim of this study, the following objectives were defined:

- Preparation and characterization of hydroxyl- (IL-OH) and ester- (IL-Ester) functionalized ILs,
- Investigation of phase behaviour of IL-OH and IL-Ester ILs in the presence of aqueous media,
- Extraction of levulinic acid and fumaric acid from aqueous solution with pure IL-OH,
- Study the acid-catalysed transesterification of IL-Ester with butanol and recovery of IL-OH,

- Development of a conceptual framework and model for the optimal reactive extraction of carboxylic acids from aqueous solution using IL-OH.

## 1.5 Statement of novelty

The application of tertiary amines for the reactive extraction of carboxylic acids has been an active area of research lately. The advent of ILs as an alternative to molecular solvents offered new insights and opportunities in solvent development and engineering. As a result, the applicability of ILs as reactive extractants for carboxylic acids has been investigated in this thesis. The “reactive” aspect of the reactive extraction reported in the literature, for both tertiary amines and ILs is questionable. In principle, the current state of the art of reactive extraction is based on electrostatic interactions between the solute and the extractant, i.e., ion-pair formation and hydrogen bonding [50]. The extractability of carboxylic acids from aqueous solutions with these known solvents is driven by solubilization, diffusion, and neutralization [34]. The application of an extractant which reacts with the carboxylic acids, in a chemical reaction other than neutralization, is yet to be reported.

Secondly, most of the research on the application of ILs in the reactive extraction of carboxylic acids from aqueous solutions focuses on establishing performance parameters such as distribution coefficients and efficiencies. The mechanism behind the extraction of carboxylic acids using ILs is either misinterpreted or not considered. Furthermore, the mutual solubility between the hydrophobic ILs and the aqueous phase is usually neglected. As a result, the performance of ILs from both techno-economic and environmental perspectives is misrepresented.

This study presents an experimental approach with respect to the preparation and application of a hydroxyl-functionalized IL for the reactive extraction of carboxylic acids from aqueous solutions. The hydroxyl-functionalized IL was previously (in the author’s Master of Science dissertation) synthesized through the alkylation of 1-methylimidazole with 6-chlorohexanol to form 1-(6-hydroxyhexyl)-3-methylimidazolium chloride. The hydrophobic character was induced into the IL through an ion-exchange reaction with lithium bis(trifluoromethanesulfonyl)imide, resulting in the formation of 1-(6-hydroxyhexyl)-3-methylimidazolium bis(trifluoromethanesulfonyl)imide (IL-OH) [51].

Additionally, the density, viscosity, thermal stability, and refractive index of binary [IL-OH + IL-Ester] mixtures were investigated across the entire mole fraction range [51]. As an extension of this, in the course of the current work, a group contribution method has been applied to the data which allows the determination of the density of IL-OH, IL-ester as well as their mixtures, in dependence of temperature from refractive index data.

One of the key outcomes of the previous work was that the density difference between both, IL-OH and IL-Ester, and water is greater than  $0.4 \text{ g cm}^{-3}$  and both ILs are hydrophobic and immiscible with water. These are important aspects for their application in the reactive extraction: the large density difference between the IL and aqueous phases will allow for ease of phase separation during the application of IL-OH in aqueous environments and reduces the likelihood of forming emulsions. Phase separation should hence readily occur.

The targeted application of IL-OH is in the reactive extraction of carboxylic acids from aqueous solution. During application, IL-OH will be exposed to large volumes of water, therefore mutual solubility of water and IL-OH was investigated as a function of operating conditions such as initial pH and catalyst loading. This provided evidence of the extent to which the miscibility of IL-OH and water is influenced by operating conditions.

The application of IL-OH for the reactive extraction of carboxylic acids from aqueous solution was investigated using model aqueous solutions of fumaric or levulinic acid. In principle, the extraction of the carboxylic acids can be based on two extraction mechanisms, physical and chemical extraction. The extractability of fumaric or levulinic acid was investigated under physical and chemical extraction conditions. The operating conditions for physical extraction were chosen such that the occurrence of a chemical reaction between IL-OH and carboxylic acids was not possible. The presence of an acid catalyst during chemical extraction led to the formation of IL-Ester, providing evidence that the extractability of the carboxylic acids could be enhanced by a chemical reaction.

It was assumed that if all the IL phase is chemically bound to levulinic acid, then the IL phase is IL-Ester. Hence, there is need to strip the extracted carboxylic acid from IL-Ester for further downstream processing, simultaneously regenerating IL-OH for reuse. The stripping method is largely influenced by process characteristics. The presence of

esters in the IL phase allowed for the use of a transesterification reaction to regenerate IL-OH and simultaneously release a value-added derivative of the carboxylic acid. The acid-catalysed transesterification of IL-Ester with butanol produced IL-OH and butyl levulinate, a value-added derivative of levulinic acid. A forward bias was created by using a large excess of butanol during the transesterification reaction. A two-stage separation protocol was developed to isolate butyl levulinate from the IL phase. This was achieved by initially recovering the excess butanol using vacuum distillation followed by solvent extraction of butyl levulinate from the IL phase using diethyl ether as a solvent.

Finally, a possible process configuration for the reactive extraction of carboxylic acids using IL-OH was conceptualized. The model incorporates both the extraction and back-extraction stages. A mass balance was developed for the overall process based on the proposed model. Furthermore, the mass balance calculations validated the reliability and consistency of the measured data.

## **1.6 Thesis structure**

This thesis is structured into five chapters. The contents of each chapter are as follows:

Chapter 1: the chapter presents an introduction which highlights an overview of the current research, outlines the significance of the study, and thereby establishes the hypothesis of the study.

Chapter 2: the literature review presents a detailed overview of the existing literature in relation to the current study. The chapter presents an evolution of the research question and explores the theoretical framework on which the research methodology is built.

Chapter 3: the methodology chapter presents the experimental approach taken to address the research questions.

Chapter 4: this chapter reports and discusses the findings of the study. Observations and inferences made herein are assessed, evaluated, and interpreted against the aim and objectives of the study.

Chapter 5: this chapter is a comprehensive assessment of the entire study, presenting conclusions, recommendations, and future outlook.

## **2 Literature review**

This chapter presents a detailed review on the importance and development of reactive extraction as a method of separation and recovery for carboxylic acids from aqueous solution. The theory of ILs, with special emphasis on their importance to the current study, is also discussed.

### **2.1 The biorefinery concept**

Industrial production of chemicals and materials has evolved significantly over the past decades. While the growth of the petrochemical industry characterized the 20<sup>th</sup> century, the need to drive towards an environmentally and economically sustainable production of materials was recognized at the start of the 21<sup>st</sup> century [52, 53]. Furthermore, conventional petrochemical routes are limited by the supply and availability of petroleum resources in nature. On the other hand, biomass is an alternative and renewable feedstock that can replace petroleum feedstocks.

The utilization of biomass as a feedstock has an economic value that can positively impact on the profit margins in industrial manufacturing [54]. Thus, the biorefinery concept aiming to enhance the processing and conversion of biomass materials into a wide variety of value-added products and energy was developed [53, 54]. The ultimate vision of the biorefinery concept is to transform the global economy into an environmentally and economically sustainable bio-based economy [55].

Biomass feedstocks vary in terms of composition and chemistry. As a result, biorefineries are classified based on feedstocks, platforms, processes, and products involved [54]. For example, the syngas platform is used for the Fischer-Tropsch production of biodiesel, while the C6 sugar platform is used to produce bioethanol. Biorefineries utilize an integrated system of pathways and processes to produce value-added products. Table 2.1 summarizes the classification of biomass conversion pathways as discussed by Cherubini et al. [56].

Table 2.1: Classification of conversion pathways used in biorefineries [56].

Type	Description	Examples
Mechanical	<ul style="list-style-type: none"> <li>- Physical separation and size reduction and separation of feedstock streams.</li> <li>- Maintains the structural integrity of the substrate.</li> </ul>	<ul style="list-style-type: none"> <li>- Pre-treatment,</li> <li>- Milling</li> <li>- Pressing</li> <li>- Separation</li> </ul>
Biochemical	<ul style="list-style-type: none"> <li>- Microbial conversion of substrates into products.</li> <li>- Requires mild operating conditions.</li> </ul>	<ul style="list-style-type: none"> <li>- Anaerobic Digestion</li> <li>- Fermentation</li> <li>- Enzymatic conversion</li> </ul>
Chemical	<ul style="list-style-type: none"> <li>- Chemical conversion of the substrate via a series of chemical reactions under controlled temperature and pressure.</li> </ul>	<ul style="list-style-type: none"> <li>- Hydrolysis,</li> <li>- Hydrogenation</li> <li>- Transesterification</li> <li>- Oxidation</li> </ul>
Thermochemical	<ul style="list-style-type: none"> <li>- The substrate is exposed to extreme temperature and pressure conditions to achieve conversion.</li> <li>- Both catalytic and non-catalytic.</li> </ul>	<ul style="list-style-type: none"> <li>- Pyrolysis</li> <li>- Gasification</li> <li>- Hydrothermal upgrading</li> </ul>

The successful operation of a biorefinery is entirely dependent on the efficiency of unit operations. The enhanced design and integration of unit operations promote productivity as well as expand the product range and reduce waste within the biorefinery [55].

## 2.2 Product portfolio for biorefineries

The significant difference between biomass and petroleum feedstocks is in the heteroatom substitution, with a higher substitution being found in biomass [57]. This means biomass has an excess of functionalities that can be exploited and manipulated through different pathways to yield a wide range of value-added products. Biochemicals can be incorporated into commodity, platform and specialty chemicals based on their properties and uses. Figure 2.1 shows examples of value-added biochemicals [52, 58-61].

The transformation of biomass into many value-added chemicals can be achieved via fermentation pathways. In addition, several valuable co-products are produced together with the main product during fermentation [4]. For example, the anaerobic fermentation

of sugars to produce succinic acid tends to co-produce ethanol, acetic acid, and lactic acid. Nevertheless, compared with chemical conversion, fermentation is highly selective. Hence, fermentation is an attractive alternative due to its low emissions and high selectivity, and fermentation is both environmentally and economically sustainable.

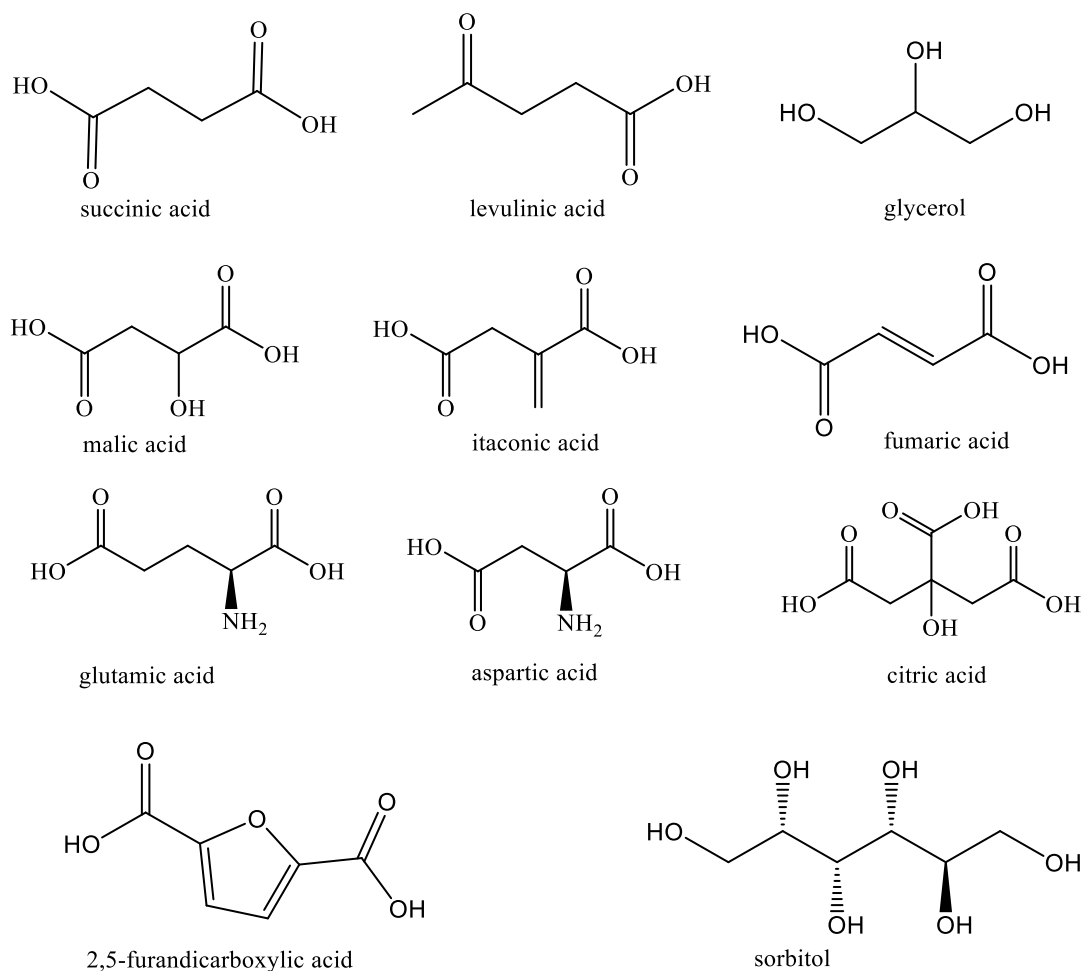


Figure 2.1: Examples of value-added chemicals derived from biorefinery processes.

The value-added chemicals derived from biorefining processes find use in a versatile range of applications. Most importantly, these chemicals are classified as platform chemicals, i.e., they are used as the substrate to produce other value-added materials and chemicals. This further extends the product portfolio of biorefining processes. For example, levulinic acid and its derivatives find use in various applications and industries such as fuels and fuel additives, solvents, polymers, and pharmaceuticals among others as shown in Figure 2.2.

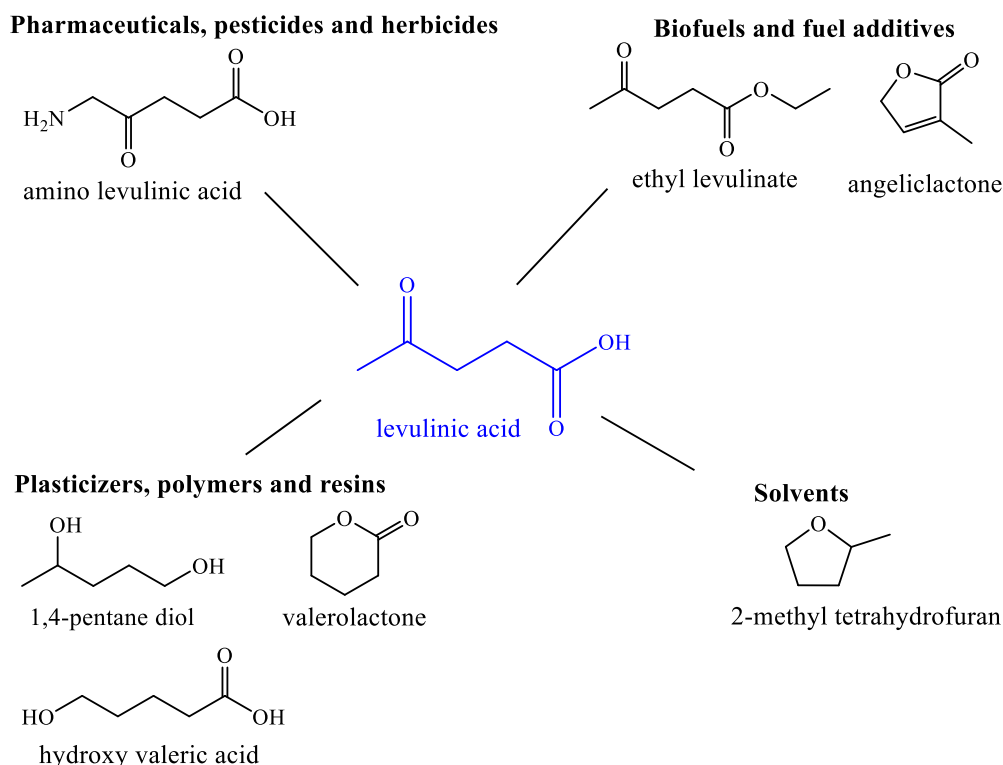


Figure 2.2: Examples of value-added chemicals derived from levulinic acid.

## 2.3 Production of carboxylic acids via fermentation

A wide range of carboxylic acids have been successfully produced from different substrates via fermentation. However, most fermentation routes are limited by end-product inhibition [62, 63]. This means that the optimum productivity of the microbes is restricted to a certain concentration level by the accumulating product. The productivity of most fermentation systems is very low since end-product inhibition is triggered at very low product concentrations. Table 2.2 shows selected examples of carboxylic acids produced via fermentation.

The productivity and efficiency of the system can be improved by continuous removal of the accumulating product, e.g., by extractive fermentation. For example, *in situ* extractive fermentation eliminates the inhibitory effects of the accumulating product during fermentation [4, 64].

Garrett et al. [64] reported that the productivity of *bacillus coagulans* tends to increase 1.4-fold when Amberlite IRA-67, an ion-exchange resin, is used for *in situ* product recovery in the fermentative production of lactic acid.



Table 2.2: Examples of carboxylic acids produced via fermentation.

Microorganism	Product	Productivity/g L <sup>-1</sup>	Ref
<i>Propionibacterium acidipropionici</i>	Propionic acid	0.42	[4]
<i>Bacillus coagulans</i>	Lactic acid	3.69	[4]
<i>Clostridium tyrobutyricum</i>	Butyric acid	21.8	[65]
<i>Gracilaria tenuitipitata</i>	Levulinic acid	3.66	[66, 67]
<i>Rhizopus arrhizus</i>	Fumaric acid	4.25	[68]

## 2.4 Carboxylic acid market value

Organic acids are in great demand, and as a result, their market value is rising. It is predicted that the global market for organic acids will reach USD\$36.9 by 2026 [69]. Carboxylic acids find application in various industries such as the food, pharmaceutical, polymer, cosmetic, and fuel industries. The versatile application of carboxylic acids has led to an increase in demand and therefore investment in the market. For example, the fumaric acid market is projected to grow by 4.8% CAGR during the period 2023-2028, reaching a market value of USD 960.69 million [70]. Similarly, the global levulinic acid market was valued at approximately USD\$27.25 million in 2022, with a predicted compound annual growth rate (CAGR) of 9 – 14% between 2022 and 2030 [67, 71]. On the other hand, levulinic acid esters are value-added derivatives of levulinic acid and they offer a similarly extensive range of applications [72, 73]. The major drive towards the use of levulinic acid esters is due to their low toxicity, stable flashpoint, and excellent low-temperature fluidity and lubricity. A comparison of the selling price of levulinic acid and its esters on the South African market is presented in Table 2.3. It is important to note that small-scale chemical supplier prices are indicative of the overall market value at a global level.

Table 2.3: Comparison of the selling price of levulinic acid and its esters in South Africa.

Product	Purity/%	Price per kg/R	Supplier	Ref.
Levulinic acid	98	2 053.10	Merck	[74]
Ethyl levulinate	99	4 655.00	Merck	[75]
Butyl levulinate	98	7 162.35	TCI Chemicals	[76]

Thus, considering the market value of butyl levulinate, butyl levulinate was selected for the purpose of this study.

## **2.5 Recovery of carboxylic acids from aqueous solution**

A typical fermentation broth is usually a complex matrix of nutrients, microbes, and various carboxylic acids at concentrations in the range between 0 – 10 wt.%. Carboxylic acids have low activity coefficients in the aqueous phase due to the high affinity of the carboxyl group for water. Carboxylic acids tend to exhibit low volatilities and therefore, conventional separation and recovery processes, such as distillation, are less ideal for the recovery of carboxylic acids from aqueous solutions.

The conventional recovery of carboxylic acids, such as citric acid and lactic acid, from macro-fermentation processes is largely based on the precipitation of calcium salts of the carboxylic acids [3]. In this case, calcium hydroxide,  $\text{Ca(OH)}_2$ , is added to the fermentation broth to precipitate the respective calcium carboxylate, followed by filtration. The carboxylic acid is then recovered from the calcium salt through the addition of concentrated sulfuric acid and the preferential precipitation of calcium sulfate,  $\text{CaSO}_4$ . Further treatment on the recovered carboxylic acid with activated carbon and ion-exchange resins is still required. Thus, this method requires the use of large quantities of chemicals and an excess amount of  $\text{CaSO}_4$  is produced, presenting disposal issues.

The mostly used techniques for the recovery of carboxylic acids from aqueous solution have been presented earlier in Table 1.1. These recovery techniques are often used in combination to improve the recovery yields.

Reactive extraction of carboxylic acids is a separation process that utilizes a chemical reaction to transfer a carboxylic acid from an aqueous phase into a water-immiscible extractant. In principle, the practical application of reactive extraction extends to separation, recovery, or conversion of carboxylic acids from their aqueous solutions. A wide range of value-added carboxylic acid derivatives can be derived from the process. The carboxyl functionality in carboxylic acids lends itself ideally for the application of reaction-enhanced separation techniques. The carboxyl group can participate in reversible chemical reactions, which can be exploited for the design of a reactive extractant. The recovery of carboxylic acids from aqueous solution by reactive extraction is achieved through the reaction between the extractant and acid molecules [6]. The

reactive extraction of carboxylic acids with tertiary amines is based on ion-pair formation or proton transfer between the carboxylic acid and the amine [3, 6, 24]. The product must be more readily soluble in the organic phase than the aqueous phase. Long-chain aliphatic amines are usually used as the reactive extractants. Tertiary amines may be the readily available organic extractants offering high extractability of the carboxylic acids. However, their characteristic high viscosities and the corrosive nature limit their utilization. Such properties may need to be adjusted by means of diluents.

Inyang and Lokhat [47] investigated the reactive extraction of propionic acid with TOA in the presence of 1-decanol as a diluent. They optimized the reactive extraction process by making use of a response surface method to determine the interactive effects of process variables. This study concluded that the extractability of propionic acid increases with increasing initial concentration of propionic acid in the aqueous phase. However, increasing the concentration of TOA in the organic phase resulted in a reduced extraction efficiency. Finally, temperature does not affect the extractability of propionic acid with TOA in the presence of 1-decanol as a diluent.

Datta and co-workers [77] investigated the effect of adding oleyl alcohol and *n*-dodecane as diluents in the reactive extraction of nicotinic acid with tri-*n*-dodecylamine (TDDA). The extractability of nicotinic acid was improved by mixing *n*-dodecane and oleyl alcohol with TDDA and a distribution coefficient of 1.84 was reported.

Inyang and Lokhat [78] highlighted that reactive extraction techniques can improve the downstream processing of carboxylic acids, thereby reducing operational costs. The study recommended further research on reactive extraction, suggesting the use of ILs as reactive extractants.

The choice of the solvent is critical in the performance of a reactive extraction system. Furthermore, the extraction mechanism by the selected solvent must be clearly understood. Finally, the solvent must adhere to a set of parameters commonly referred to as the solvent selection criteria summarized in Table 2.4.

Table 2.4: Solvent selection criteria in reactive extraction of carboxylic acids [6, 79].

Parameter	Explanation
Selectivity	High degree of selectivity favours high separation factors.
Capacity	Solvents with a high capacity require low solvent to feed ratios during application.
Viscosity	Solvents with high viscosities are characterized with reduced mass transfer efficiency. Extra pumping equipment is required to improve pumping and dispersion.
Solubility	A hydrophobic solvent with almost negligible miscibility with water will readily form a biphasic system.
Stability	The thermal and chemical stabilities of the solvent should be high enough to withstand degradation, decomposition and transformation during regeneration and recycling.
Toxicity	The solvent must adapt to biochemical processes without affecting viability and productivity of microbes.
Corrosivity	Highly corrosive solvents are dangerous to handle, and they reduce the lifespan of the reactors.
Recoverability	The solvent should be easily stripped from the solute
Environmental impact	The release of the solvent into the environment should be kept minimal. The solvent should be biocompatible with the environment, posing no risk to living organisms
Availability	Heavy material losses are often incurred during operation therefore, the solvent must be cheap and readily available.

The extractability of a solute during solvent extraction is dependent on partitioning of the solute between two immiscible solvents. The extent of the partitioning is governed by the distribution law, which states that regardless of its concentration, a solute is always distributed in a constant ratio between two immiscible phases [80]. It must be emphasized that the distribution is temperature dependent, and the solute must strictly exist as the same molecular species (i.e., not partially dissociated, as could be the case for carboxylic acids) in both phases. The equilibrium concentrations of the solute in both phases can then be used to calculate the distribution coefficient ( $K_D$ ) by means of Equation 2.1.

$$K_D = \frac{[A]_{org}}{[A]_{aq}} \quad (2.1)$$

where  $[A]_{\text{org}}$  and  $[A]_{\text{aq}}$  refer to the equilibrium concentrations of the solute in the organic and aqueous phases, respectively.

## 2.6 ILs as designer solvents

ILs are a class of low melting point, non-molecular solvents that wholly consist of ions, and they offer unlimited possibilities in solvent engineering. An IL has a wide liquidus range, negligible vapour pressure and high thermal stability [81]. The physicochemical properties of ILs can be easily manipulated through careful selection of anion-cation combinations, making them designer solvents for use in a versatile range of applications. Solvent power, biocompatibility, hydrophobicity, viscosity, density, surface tension, among others, are some of the most sought-out properties that can be customized in individual ILs [82].

ILs are also used as mixtures, which can be classified as binary, ternary, or quaternary depending on the number of ions present in the system. Mixtures of ILs tend to have characteristic physicochemical properties and applications extending beyond the range defined by the individual neat ILs [83-85]. Table 2.5 summarizes the classification of IL mixtures.

Table 2.5: Classes of IL mixtures.

Mixture	Number of ILs	Number of ions	Mixture*
Binary	2	3	$[A][X] + [B][X]$ or $[A][X] + [A][Y]$
Reciprocal binary	2	4	$[A][X] + [B][Y]$
Ternary	3	4	$[A][X] + [A][Y] + [A][Z]$ or $[A][X] + [B][X] + [C][X]$
Reciprocal ternary	3	4	$[A][X] + [A][Y] + [B][Y]$ or $[A][X] + [B][X] + [B][Y]$

\* Where the respective cations in the individual neat ILs are [A], [B] and [C] while [X], [Y] and [Z] are the respective anions.

### 2.6.1 Classification and preparation of ILs

There is extensive literature on the different types of ILs that have been synthesized for different uses and applications. Stark and Seddon [26] discussed the classification of ILs,

and three distinct generations were identified. The first generation of ILs is based on the cations accompanied by haloaluminate (III) ions. ILs in this category exhibit unique physical properties such as viscosity and melting point. However, first-generation ILs have poor stability in both air and aqueous environments, hence are difficult to handle. For example, the addition of aluminium(III) chloride ( $\text{AlCl}_3$ ) to 1-ethyl-3-methylimidazolium chloride ( $(\text{EMIM})\text{Cl}$ ) yields the IL  $[\text{EMIM}][\text{AlCl}_4]$ . The IL has a characteristic Lewis acidity profile. In the presence of moisture, the  $[\text{AlCl}_4]^-$  forms the hydroxoaluminate(III) species, thereby decomposing the IL [26].

In the second-generation category, the ILs have high stability in both air and water. Chemical properties, such as solubility, chemical reactivity, electrochemical window, among others, are enhanced for the second-generation ILs. The third generation is comprised of ILs with customized chemical functionalities. Typical anions used for the design of third generation ILs are derived from alkylsulfates, amino and carboxylic acids. Examples of ILs in the three generations are presented in Table 2.6.

Table 2.6: Classification of ILs.

Generation	Example	Abbreviation
First	1-ethyl-3-methylimidazolium chloride aluminium chloride	$[\text{EMIM}]\text{Cl}-\text{AlCl}_3$
Second	1-ethyl-3-methylimidazolium bis(trifluoromethanesulfonyl)imide	$[\text{EMIM}][\text{NTf}_2]$
Third	1-ethyl-3-methylimidazolium methylsulfate	$[\text{EMIM}][\text{MeSO}_4]$

The synthesis of ILs follows two main steps, namely, the alkylation reaction and the ion-exchange reaction. The alkylation reaction is the key step towards the formation of the desired cation. In the case of amine-derived ILs, the amine is either protonated under an acidic environment or an alkyl halide is used in a quaternization reaction [86]. These reactions are typically conducted under temperature-controlled conditions. The choice of the alkyl halide is critical to the functionalization and customization of the cation.

The anion-exchange reaction is the second step in the preparation of ILs. The desired physicochemical properties of the IL are dependent upon the anion. The anion-exchange is achieved by contacting the halide-based IL with either a Lewis acid or with an inorganic

salt of the corresponding acid of the desired anion. Ion-exchange resins can also be employed for this reaction [87]. In principle, the anion-exchange reaction is driven by the interaction energies between the ions of the IL and those of the salt [88]. In this case the IL cation pairs up with the strongest interacting anion in the system. In certain cases, the anion-exchange is driven by the formation of a second, water-insoluble ionic liquid phase.

The classification of ILs primarily rests on the cation group chosen. Among the various sub-classes of IL, N-heterocyclic cations are the most common. Furthermore, the imidazolium cation is usually the most preferred due to the ease of synthesis and derivatization, as well as redox stability [89, 90]. The chemical structures of some of the cations used in the design and synthesis of ILs are presented in Figure 2.3.

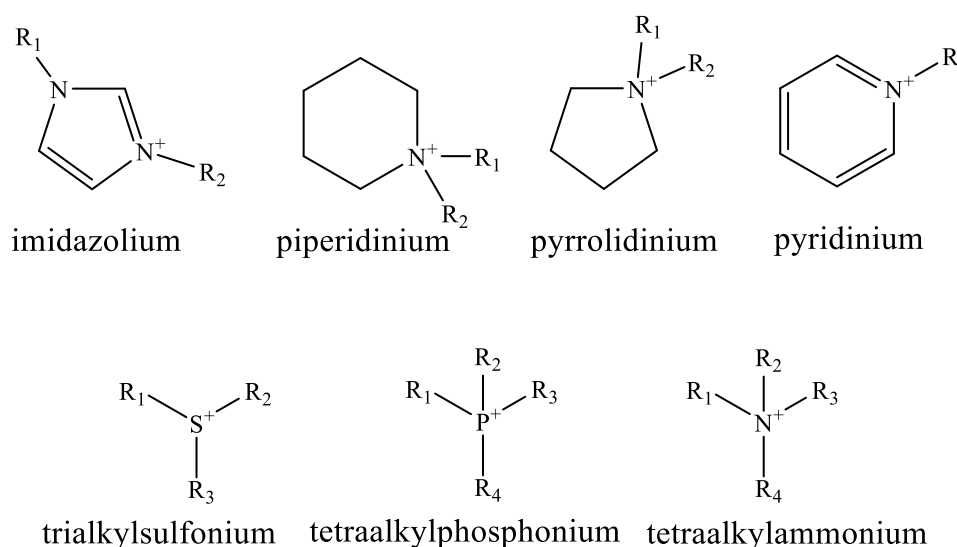


Figure 2.3: Chemical structures of some of the cations used in the design of ILs, where  $R_i$  represents any alkyl or aryl substituent.

Anions used in the design and synthesis of ILs comprise organic to inorganic. Typical anions are shown in Figure 2.4. Apart from halide anions, most of the anions usually have a delocalized negative charge distributed over a group of atoms. The coordination strength of an anion is dependent on the extent of the delocalization of the negative charge. In the case of  $[\text{PF}_6]^-$ ,  $[\text{BF}_4]^-$ , and  $[\text{NTf}_2]^-$  where the charge is delocalized amongst several atoms, the anions become weakly coordinating [91].

Strongly coordinating anions include halide ions, where the negative charge is centred on a single atom. Figure 2.4 shows selected examples of anions used in the design of ILs. Physicochemical properties of ILs, such as density, viscosity, and hydrophobicity among others are largely dependent upon the nature of the anion [92, 93]. Therefore, changing the anion of an IL results in the formation of a new IL with different properties. For example, Jacquemin et al. [94] investigated the effect of anion and alkyl chain on the density of imidazolium-based ILs. The study investigated the densities of 1-butyl-3-methylimidazolium hexafluorophosphate ([BMIM][PF<sub>6</sub>]), 1-butyl-3-methylimidazolium tetrafluoroborate ([BMIM][BF<sub>4</sub>]), 1-butyl-3-methylimidazolium bis-(trifluoromethanesulfonyl)imide ([BMIM][NTf<sub>2</sub>]), and 1-ethyl-3-methylimidazolium bis-(trifluoromethanesulfonyl)imide ([EMIM][NTf<sub>2</sub>]). It was established that the density of the ILs was dependent on the anion as well as the chain length of the alkyl group on the cation. In addition, the density decreased linearly with increasing temperature.

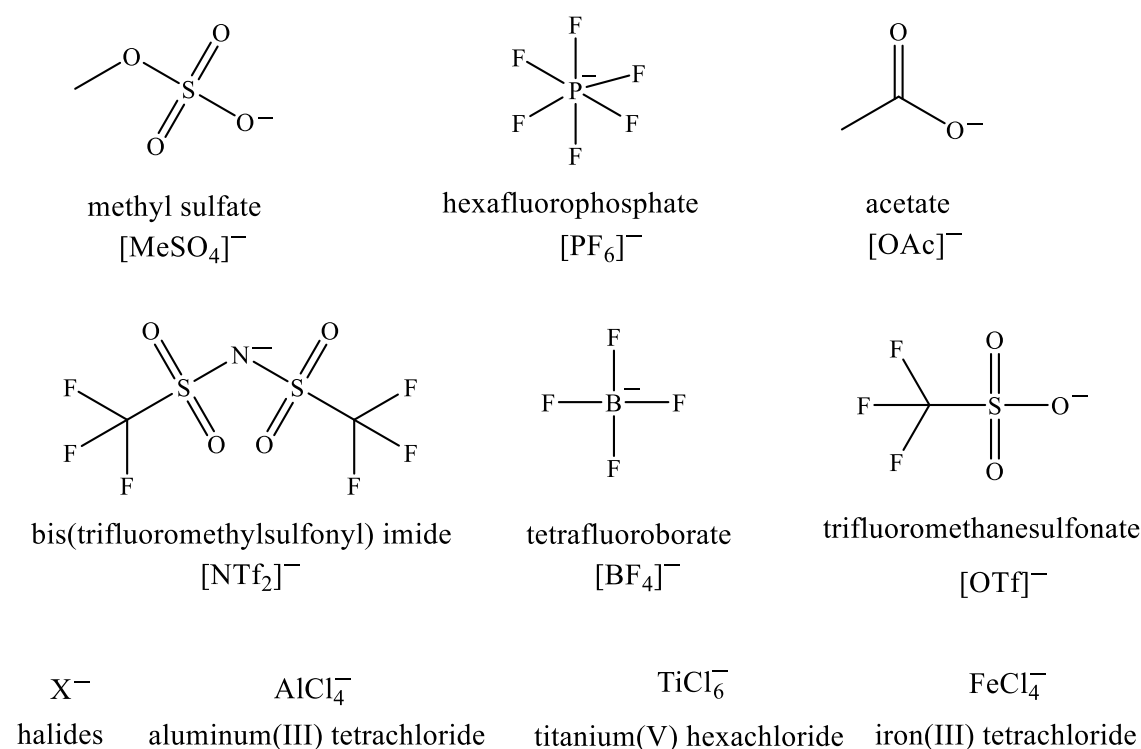


Figure 2.4: Examples of anions used in the design of ILs.

## 2.6.2 Examples of cation-functionalized ILs

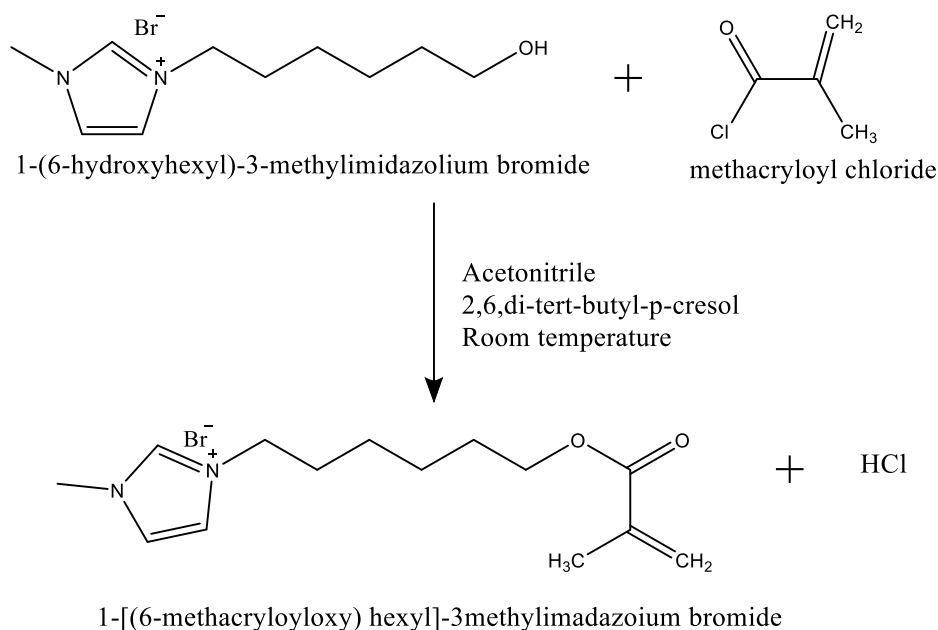
Recent trends have shown widespread interest in the application of ILs. However, most of these applications focus on use of ILs as solvents and catalysts in chemical reactions



and processes. The reactivity of ILs can be manipulated by embedding potentially reactive functional groups in either of the ions, but most preferably the alkyl substituent of the cation [41].

Two examples highlight the applications of cation-functionalized ILs like those developed for this thesis. For example, Sideris et al. [95] reported the microwave-assisted synthesis of hydroxyl-functionalized ILs used for the ionothermal synthesis of lithium iron phosphate,  $\text{LiFePO}_4$ . The study investigated the influence of the length of the hydroxyl-terminated alkyl chain on the particle size and morphology of the resulting  $\text{LiFePO}_4$  crystals. Furthermore, the  $[\text{NTf}_2]^-$  anion was selected as the counter-anion because of the characteristic high thermal stability of  $[\text{NTf}_2]$ -based ILs. Increasing the alkyl chain length improved the recovery yield of  $\text{LiFePO}_4$ .

In 2016, Sardar and co-workers reported the synthesis and characterization of 1-[(6-methacryloyloxy)hexyl]-3-methylimidazolium bromide, a polymerizable IL [96]. The reaction was based on the esterification of a hydroxyl-functionalized IL using an acyl chloride as shown in Scheme 2.1. Similarly, the synthesis of methacrylate-type polymerizable ILs was also reported by Pöhako-Esko et al. [97].



Scheme 2.1: Synthesis of 1-[(6-methacryloyloxy) hexyl]-3-methylimidazolium bromide.

### 2.6.3 Environmental impact of ILs

The negligible vapour pressure of ILs has been a key factor behind the growth in interest for these solvents. Due to their ionic character, volatile emissions of ILs are intrinsically low. However, the release of ILs into the environment is inevitable. For example, the aqueous solubility of ILs as well as spillages can account for the presence of even the most hydrophobic ILs in the environment. Considering the stability of ILs, their presence in the environment is of great concern in the long-term. Therefore, it becomes important to establish toxicological profiles of the respective ILs. The octanol-water partition coefficient ( $K_{ow}$ ) is a useful parameter for this purpose.

The partition coefficient ( $P$ ) is a ratio of the equilibrium concentrations of a solute distributed between two immiscible phases.  $P$  values calculated as the differential solubility of a solute between water and octanol give the  $K_{ow}$  of the respective solute.  $K_{ow}$  is calculated according to Equation 2.2:

$$K_{ow} = \frac{[A]_{org}}{[A]_{aq}} \quad (2.2)$$

where  $[A]_{org}$  and  $[A]_{aq}$  are the equilibrium concentration of the solute in the octanol and aqueous phases, respectively.  $K_{ow}$  values give a measure of the lipophilicity of the solute [98, 99].  $K_{ow}$  can also be presented as  $\log K_{ow}$ . This data is useful in prediction and assessment of the risks of exposure, toxicity and impact of chemical compounds on human health and other living organisms [99, 100].  $K_{ow}$  is critical in applications such as drug design and drug development to predict the permeability profile of active compounds within body cells.  $K_{ow}$  values are used to predict and assess the following:

- a) Soil sorption
- b) Biological uptake
- c) Bioaccumulation and bioconcentration
- d) Toxicity

When determining  $K_{ow}$  of ionizable solutes, pH is a critical parameter. However, the use of buffers tends to modify the observed partitioning. Components of the buffer solution can potentially form ion pairs with the ionized solutes, creating a secondary chemical equilibrium within the system [101]. This ultimately modifies the  $K_{ow}$  value of the

respective solute. Table 2.7 presents log  $K_{ow}$  values of some molecular solvents commonly used for the extraction of carboxylic acids from aqueous solutions.

Table 2.7: Octanol-water partition coefficients of typical organic compounds and ILs.

<b>Solvent</b>	<b>log <math>K_{ow}</math></b>	<b>Ref</b>
Decanol	4.57	[102]
Heptane	4.50	[102]
MIBK	1.30	[103]
TOA	10.5	[104]
[BMIM][PF <sub>6</sub> ]	-1.65	[99, 105]
[OMIM][NTf <sub>2</sub> ]	0.93	[99]
[HMIM][NTf <sub>2</sub> ]	-1.45	[105]
[BMIM]Cl	-2.39	[99]

$K_{ow}$  specifically refers to the distribution of a solute between the octanol and aqueous phases and should be independent of the solute's concentration.

The Stockholm Convention on persistent organic pollutants established a criteria for the identification and classification of toxic organic pollutants [106]. Emphasis is placed on the biodegradability, bioaccumulation and transport properties of the compounds in the environment. A chemical compound is considered to be bioaccumulative if either the bioaccumulation factor is greater than 5000, or if  $\log K_{ow} > 5$ . As can be seen in Table 2.7, typical ILs have very low log  $K_{ow}$  values compared with tertiary amines such as TOA, as well as typical organic solvents.

Even though ILs are considered as “green” solvents, their environmental impact is yet to be fully understood. Aqueous solubility of ILs promotes their release into the environment, increasing chances of ecotoxicity and bioaccumulation. Therefore, it is important to be able to predict the consequences of their release into the environment.

Rybinska et al. [107] developed a quantitative structure property model (QSPR) to estimate the lipophilicity of a set of 335 ILs. The observed log  $K_{ow}$  values were classified according to the Stockholm Convention toxicity criteria as follows:

- a) if  $\log K_{ow} > 5$ , the IL has a higher potential to bioaccumulate.
- b) if  $0 < \log K_{ow} < 5$ , the IL can partially bioaccumulate.

- c) if  $\log K_{ow} < 0$ , the IL is hydrophilic and can be transported in the aqueous system.

There are several methods used to measure the  $\log K_{ow}$  values of chemical compounds. These methods include the shake-flask method, the slow stirring method, the generator column, and the chromatographic method [99, 100, 108]. Ideally; the shake-flask method is the most straightforward approach [108]. In this case, a known amount of solute is dissolved in either water or octanol, followed by shaking the aqueous and organic phases to achieve equilibrium. After phase separation, the concentration of the solute in both phases is established. The equilibrium ratio of the solute concentration is considered for the determination of the  $\log K_{ow}$ . Estimate values can also be predicted using the quantitative structure-activity relationship model (QSAR) [109, 110].

The industrial application of ILs will inevitably generate a significant environmental impact. As a result, the biodegradability of ILs must be understood. The biodegradability of compounds can be classified according to the mechanism and extent of degradation [111]. For example, the loss a specific moiety (for example, the hydrolysis of esters) is considered as primary degradation. On the other hand, if a compound can be broken down completely, then it is ultimately biodegradable. Furthermore, the extent of degradation within a given timeline is also used to establish whether the compound is readily biodegradable. The extent of degradation of ILs has been attributed to the cation more than the anion. This means that the core ring structure, the alkyl substituent, and the nature of functional groups in the alkyl substituent are critical to the degradation mechanism. Imidazolium-based ILs are reported to have low or non-biodegradable characteristics [112]. Furthermore, the presence of functional moieties such as allyl, vinyl, hydroxyl, ether, carboxyl, sulfonate, and nitrile has been reported to have negligible effect on the biodegradability of imidazolium and pyridinium-based ILs.

## **2.7 Consideration of solubility aspects of reactive extractants**

Tertiary amines are the most used solvents for the reactive extraction of carboxylic acids from aqueous solution [11, 113, 114]. The mechanism of the reactive extraction of carboxylic acids by tertiary amines is entirely dependent on the difference in the basicity between the amine and the corresponding carboxylate anion of the carboxylic acid, as highlighted by Canari and Eyal [14]. In principle, the basicity of amines is such that the

basicity of primary amines > secondary amines > tertiary amines. Unfortunately, primary amines have a characteristic high solubility in water, which limits their applicability in the reactive extraction for carboxylic acids from aqueous solution [6]. The extractability of carboxylic acids from aqueous solution using secondary amines is greater compared to tertiary amines. However, the downstream processes following reactive extraction incorporate distillation, where secondary amines tend to react with the extracted carboxylic acids to form amides. The amides are stable and complicate the stripping of the acid-laden organic phase. This makes tertiary amines the only choice for use as a reactive extractant for carboxylic acids. The solubility of amines in water is dependent on the alkyl chain length as seen in Table 2.8.

Table 2.8: Solubility of selected amines in water at 25 °C.

<b>Amine</b>	<b>Class</b>	<b>Solubility in water/g L<sup>-1</sup></b>	<b>Ref</b>
Butylamine	Primary	238.4	[115]
Trimethylamine	Tertiary	> 410	[115]
Triethylamine	Tertiary	282.4	[115]
Tributylamine	Tertiary	0.142	[115]
Trioctylamine	Tertiary	0.000039	[116]

Increasing the chain length tends to reduce the solubility of the amines in water. Additionally, increasing the chain length also increases the dipole moment in the amine thereby increasing basicity. As a result, trioctyl amine (TOA) is usually chosen as the extractant for reactive extraction of carboxylic acids from aqueous solution [11, 113, 114].

Currently, there is lack of evidence from literature sources about the solubility of water in TOA. Understanding the mutual solubility between the extractant and water is important in the design of extraction processes. The activity coefficient of the solute is reduced at high levels of mutual solubility, thereby reducing the distribution of the solute into the organic phase [117].

Tamada and King [19] investigated the solubility of the aqueous phase into the organic phase during the reactive extraction of fumaric acid, maleic acid, lactic acid, malonic acid, acetic acid, or succinic acid with Alamine 336 diluted with either heptane, chloroform, methylene chloride, nitrobenzene, methylisobutylketone (MIBK), or 1-octanol. The concentration of Alamine 336 in the diluents was kept constant for comparison purposes. It was observed that the solubility of water into the organic phase was dependent on the nature of the diluent as well as the carboxylic acid extracted. The solubility of water increased in the order heptane < chloroform < methylene chloride < nitrobenzene < MIBK < 1-octanol.

The release of the solvent into the environment can occur through leaching, spillage, and discharge. Solvents with high solubility in water will readily migrate into the biosphere. This in turn increases the theoretical oxygen demand of the effluent streams [118, 119].

Tamada and King [19] observed that the solubility of water in the organic phase was dependent on the nature of the carboxylic acid, and it followed the order fumaric acid > malonic acid > maleic acid = succinic acid > lactic acid > acetic acid. The trend shows that the solubility of water into the organic phase increases during the extraction of dicarboxylic acids compared to monocarboxylic acids, due to the extra carboxylate group in the dicarboxylic acids, which increases the interaction with water molecules via hydrogen bonding. However, their study did not provide any data with respect to the solubility of the organic phase into the aqueous phase.

While some applications require water-soluble ILs, limited miscibility with water is critical for other applications, including extractions. For example, analytical and separation technologies are dependent on the use of two-phase systems consisting of an IL and an aqueous or molecular solvent [120, 121]. The hydrophobicity of ILs affects the design of IL-based processes [122, 123].

The physicochemical properties of ILs are sensitive to slight changes in the composition of the system. Trace amounts of water can significantly alter the intended function of an IL. For example, during a reaction carried out in an IL, the anion and the reaction intermediates interact with the cation through competitive hydrogen bonding. The presence of water in an IL disrupts the cation's tendency to form hydrogen bonds with the anion, thereby altering the overall reactivity and selectivity [124]. Even in trace

amounts, water can act as an impurity in ILs, altering properties such as viscosity, surface tension and density significantly [125, 126]. This may compromise the applicability of the IL in the chosen application.

Regardless of the degree of hydrophobicity, mutual solubility between the IL and water is critical during the design of IL-based processes. There are three main concerns regarding the miscibility of ILs with water. Firstly, the aqueous phase becomes contaminated and polluted with IL species [127, 128]. This means that the costly ILs are slowly lost due to leaching of the IL phase into the aqueous phase, affecting process economics. Secondly, the presence of water in ILs tends to influence the rates and selectivity of reactions to which they are applied [129]. For example, Seddon and Stark investigated the oxidation of benzyl alcohol in the presence of palladium in 1-butyl-3-methylimidazolium tetrafluoroborate ([BMIM][BF<sub>4</sub>]). The study showed that the selectivity of the conversion to either benzaldehyde or benzoic acid is dependent upon the water content of the IL [130]. Thirdly, any change in physicochemical properties due to mutually dissolved water may negatively affect process parameters such as pumping and energy requirements.

The solubility of the IL in water is dependent on the ease of solvation of the IL cation and anion [131]. The charge density, polarity and structure of the anion collectively influence the solubility of the IL in water. The presence of functional groups such as hydroxyl, amine, and carboxyl, tend to promote hydrogen bonding with water molecules thus increasing the aqueous solubility of the IL. However, experimental data regarding the mutual solubility of ILs and water data is still limited and fragmented across literature sources. The mutual solubility data of hydrophobic ILs and water is critical in the design of IL-based extraction processes.

## **2.8 Reactive extraction of carboxylic acids via chemical reaction**

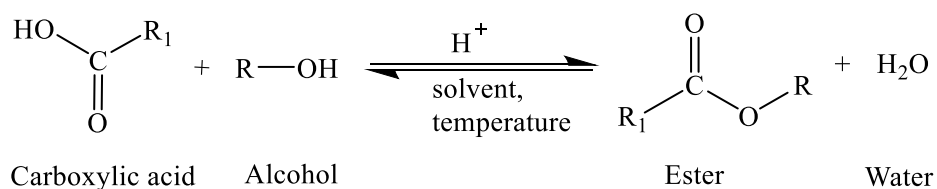
The use of chemical reactions to enhance separation processes is a well-established chemical separation technique [15, 47, 132]. Reactive distillation and reactive extraction are examples of processes during which a chemical reaction occurs simultaneously with a phase transfer of the respective reactants and products. For example, in a study by Toth et al. [133], an emulsion-enhanced biphasic esterification reaction was used for the reactive extraction of acetic acid from aqueous solution using octanol. In the second step,

the free acid was recovered and isolated from the organic phase through the reactive distillation of the organic phase in the presence of 4-dodecylbenzenesulfonic acid as a catalyst.

The chemical nature of IL-OH designed for this study allows the IL to react with carboxylic acids via an acid-catalysed Fischer esterification to form IL-Ester. An acid catalysed transesterification reaction can be employed to strip the carboxylic acid from the IL-Ester, thereby regenerating the IL-OH. IL-OH and IL-Ester are miscible while both are immiscible with water. Furthermore, there is a large density difference ( $\Delta\rho > 0.2 \text{ g cm}^{-3}$ ) between both IL-OH and IL-Ester and water. Therefore, a biphasic state is maintained during the extraction of carboxylic acids with IL-OH.

## 2.9 The esterification reaction

The acid-catalysed Fischer esterification reaction is a valuable reaction in organic chemistry and the industry at large [134]. This reaction offers a mild pathway for the transformation of organic compounds into value-added compounds [135-137]. The esterification reaction is an equilibrium-driven reaction. The accumulation of water as a reaction product creates a backward bias according to Le Chatelier's principle. The generalized chemical equation for the esterification reaction is presented in Scheme 2.2.



Scheme 2.2: Generalized chemical equation for the acid-catalysed Fischer esterification.

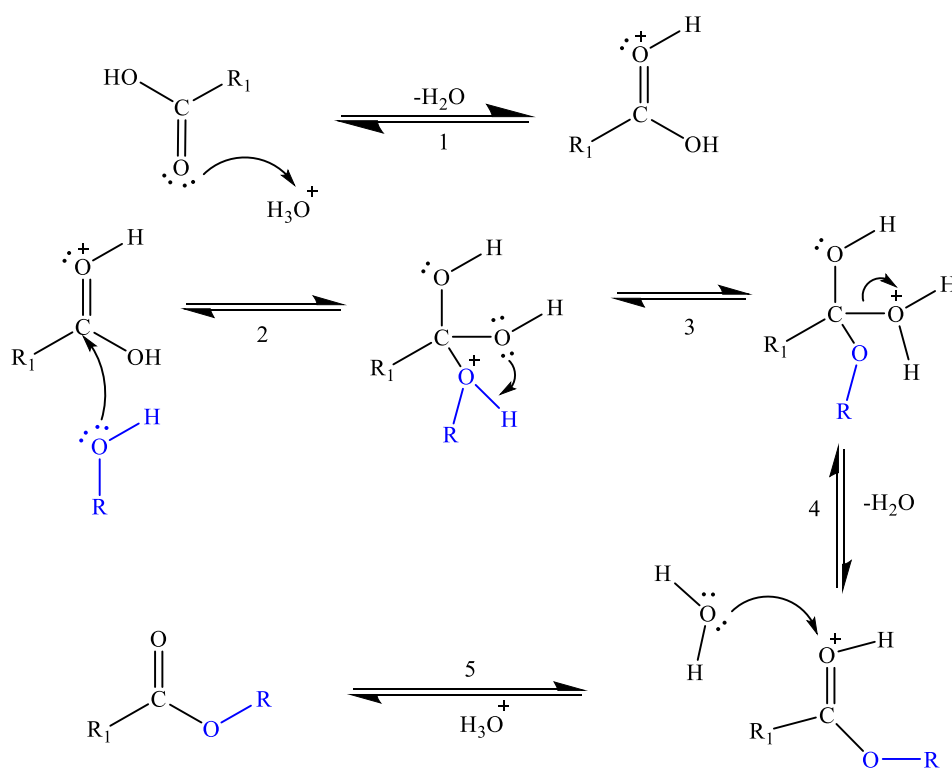
Extractive fermentation driven by esterification ultimately reduces the probability of end-product inhibition of the fermentation, thereby allowing for an increase in the productivity of the system [138, 139]. *In situ* esterification in the fermentation broth will continuously remove the inhibitory product (carboxylic acid) from the fermentation broth and transfer it into an immiscible extractant phase. Hence, it should become possible to increase the substrate concentration in the feed since the product concentration cannot exceed the inhibitory levels. As a result of extractive esterification, the removal of the carboxylic



acids from the broth tends to eliminate secondary reactions due to the carboxyl functionality of the carboxylic acid [140].

The selective removal of products or use of a stoichiometric excess of one of the reactants can influence the forward reaction, promoting higher yields. Scheme 2.3 shows the generalized reaction mechanism for the esterification reaction.

This study proposes the use of a hydrophobic, hydroxyl-functionalized IL as a reactant in the esterification of carboxylic acids. Since the IL is hydrophobic, the water produced during the esterification reaction should be immediately driven out of the IL phase, while the ester is soluble in the IL phase. As a result, a forward bias is created, promoting the forward reaction. The chemical extraction of the carboxylic acid from aqueous solution will be based on the esterification reaction. The extracted carboxylic acid will be in the form of an ester (reactive extraction) as well as physically dissolved [IL + carboxylic acid].



Scheme 2.3: Acid-catalysed Fischer esterification reaction mechanism.

## **2.10 Regeneration of the reactive extractant**

The back-extraction step strips the organic phase of the extracted carboxylic acid. The goal is to separate the carboxylic acid from the extractant, creating a product phase and a product-free solvent phase for reuse and recycling [141]. In principle, altering the extraction conditions in favour of the back-extraction is the key to regeneration of the extractant during reactive extraction-based processes [142]. Temperature and diluent compositions are some of the operating conditions which can influence the back-extraction process.

In a study by Teke and Pott [143], the extractability of lactic acid with 15% TOA in oleyl alcohol was investigated. The partition coefficient of lactic acid was found to be approximately 0.94 and the lactic acid was subsequently recovered from the organic phase via back-extraction with 2 M NaOH or 1 M HCl. In each case, the ratio of organic to aqueous phase was 10:1. The recovery of lactic acid from the organic phase was found to be 95.68 and 77.08% when using NaOH and HCl, respectively.

Back-extraction of carboxylic acids with NaOH produces sodium carboxylates, which are usually a reacidification and purification step to obtain the pure acid. The drawback of this approach is high chemical consumption and production of excess waste. Similarly, the use of HCl results in undesirable interactions between HCl and the solvent. Tertiary amines react with strong acids to form trialkylammonium salts. The complex recovery of the product from the solvent is a clear drawback of amine-based reactive extraction.

### **2.10.1 Temperature**

The reactive extraction of carboxylic acids by tertiary amines is driven by proton transfer, ion-pair formation, and hydrogen bonding. These interactions are exothermic in nature and increasing the temperature tends to destabilize the solute, increasing the entropy of the system. As a result, back-extraction is carried out at a relatively higher temperature than the reactive extraction. Tamada and King [19] discussed the effect of temperature on the extractability of carboxylic acids. During the reactive extraction of carboxylic acids with tertiary amines, the formation of acid-amine interactions reduces the entropy of the system. Therefore, increasing the operating temperature reduces the amount of carboxylic acid extracted into the organic phase. As a result, the extraction equilibrium

will shift in favour of the carboxylic acid distributing readily into the fresh aqueous phase, creating an acid-free organic phase, i.e., the regenerated solvent.

Keshav and Wasewar [141] investigated the back extraction of propionic acid from various TOA-based solvent systems at 363 K. Propionic acid was initially extracted from aqueous solution with TOA dissolved in different diluents, namely 2-octanol, 1-decanol, or methyl-iso-butyl ketone (MIBK). The initial concentrations of propionic acid used for the reactive extraction step were in the range  $0.048 - 0.366 \text{ kmol m}^{-3}$ . The back extraction of propionic acid from the organic phase was achieved by contacting the loaded organic phase with a fresh aqueous phase. It was observed that the amounts of propionic acid recovered from the loaded organic phase constituted 32 – 35%, 25 – 35%, and 66 – 88% of the propionic acid that had been extracted by the [TOA + 2-octanol], [TOA + 1-decanol], and [TOA + MIBK] solvent systems, respectively. Hence, the recovery of propionic acid from the [TOA + MIBK] solvent systems at elevated temperature was significantly higher compared to other solvent systems. However, the solubility of TOA in water at elevated temperature is most likely also increased, causing contamination. In general, it can be stated that few authors quantitatively investigate the phase composition of all components.

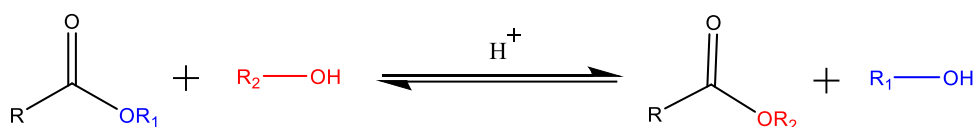
### 2.10.2 Diluent

The choice of a diluent can be adjusted after the extraction to facilitate the back-extraction process [6]. As stated before, the reactive extraction of carboxylic acids using tertiary amines is dependent on the polarity of the solvent system, and polar solvents, such as ketones or alcohols, favour the solvation of the trialkylammonium salt. Hence, reducing the polarity of the system by adding alkanes or alkyl-substituted aromatics, will reduce the distribution of the carboxylic acid into the amine-containing solvent. Under these conditions, the carboxylic acid readily partitions from the organic phase into the fresh aqueous phase, resulting in the formation of a concentrated acid-loaded aqueous phase [19]. For example, Sprakel and Schuur [30] investigated the effect of solvent composition on the reactive extraction of succinic acid from aqueous solution with TOA-based solvent mixtures. The addition of non-polar heptane to the [20% TOA + MIBK] solvent system reduced the extractability of succinic acid, i.e.,  $K_D$  values decreased from 10.2 to 0.46, respectively.

## 2.11 Transesterification reaction

A transesterification reaction is an acid- or base-catalysed reaction during which an ester is converted into a different ester. The transesterification reaction is described by the underlying mechanism, for example, alcoholysis, acidolysis and ester-ester interchange [144]. Alcoholysis occurs when the ester is reacted with an alcohol and the alkoxy groups are exchanged. If the ester is reacted with a carboxylic acid, resulting in the exchange of the carboxyl groups between the acid and the ester, the reaction is termed acidolysis.

The acid-catalysed transesterification of a carboxylic acid ester with an alcohol is presented in Scheme 2.4. In this case, the alcohol is supplied in excess to shift the equilibrium in favour of the forward reaction. The transesterification reaction is used for various industrial transformations such as in the manufacturing of biodiesel, polymers, and plastics, among other products.



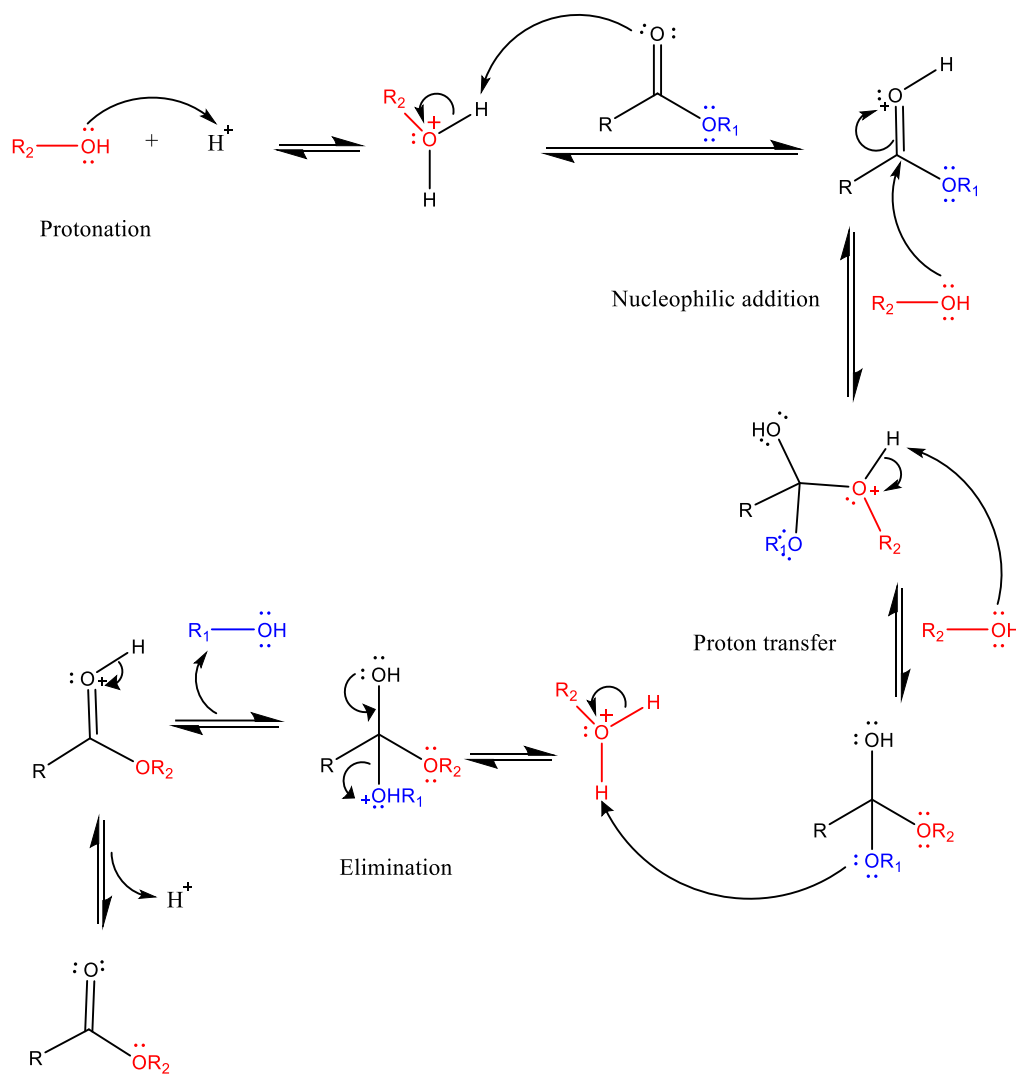
Scheme 2.4: Generalized chemical equation for the acid-catalysed transesterification reaction, where R, R<sub>1</sub> and R<sub>2</sub> represent different alkyl groups.

The acid-catalysed transesterification proceeds under the following mechanism [144]:

- i. protonation of carbonyl group of the ester
- ii. nucleophilic attack of the protonated carbonyl carbon by the alcohol
- iii. proton transfer within the intermediate
- iv. elimination of the leaving group
- v. regeneration of the acid catalyst via deprotonation

A detailed reaction mechanism for the acid-catalysed transesterification of an ester using an alcohol is presented in Scheme 2.4. This study proposes to use of an acid-catalysed transesterification reaction to regenerate the reactive extractant while simultaneously isolating the value-added carboxylic acid derivatives from the reaction mixture. It is anticipated that if there is complete conversion of IL-Ester during the transesterification reaction, then the IL will maintain its physicochemical properties. However, if transesterification reaction fails to achieve full conversion, then a binary mixture of [IL-

OH + IL-Ester] will persist, and the mole fraction of IL-Ester in the mixture will be dependent on the degree of conversion.



Scheme 2.5: Reaction mechanism for the acid-catalysed transesterification [145].

## 2.12 Chapter summary

The biorefinery concept has seen the development of unit operations capable of producing value-added products by making use of biomass as the primary feedstock. This concept is viewed as the ultimate analogue of conventional petroleum refineries. The biorefinery route is environmentally sustainable. A wide range of chemicals can be successfully produced from biorefining.

The product portfolio of biorefineries includes carboxylic acids. However, the productivity of microorganisms responsible for the biochemical conversion of biomass

into carboxylic acids is limited by end-product inhibition. As a result, carboxylic acids tend to occur at very low concentrations in large volumes of aqueous solutions. The separation, recovery, and purification of carboxylic acids from the fermentation broths remains a challenge. The overall cost and energy demand for the conventional separation and recovery methods mitigate the competitive advantage of biomass processing.

Reactive extraction is among the most promising separation methods for the recovery of carboxylic acids from aqueous solution. On the other hand, the advent of ILs has offered a multitude of opportunities in solvent engineering. The versatile nature of ILs continues to allow for the design of customized, task-specific ILs. Therefore, it is possible to design and synthesize an IL with custom properties to satisfy the solvent selection criteria for a reactive extractant while overcoming the disadvantages of amine-based reactive extraction.

The following chapters detail the synthesis and application of a hydroxyl-functionalized IL in the recovery of carboxylic acids from aqueous solution. Furthermore, the behaviour of the IL when exposed to an aqueous environment is also explored, thus addressing the phase behaviour of the IL/water system. Finally, the stripping of the carboxylic acid-loaded IL via an acid-catalysed transesterification is investigated. This allows for the regeneration of the IL as well as the isolation of the carboxylic acid in the form of a value-added derivative. The overall findings will be useful in the design of an IL-based reactive extraction system for extractive fermentation of carboxylic acids.

### 3 Experimental

The chapter identifies the materials, reagents, and chemicals, as well as instruments, used in this study. Additionally, the synthesis and application of a hydroxyl-functionalized IL is described. Emphasis is placed on the phase behaviour of the IL as well as the equilibrium studies on physical and chemical extraction. The analytical methods employed to explore the scope of this study are also discussed herein.

#### 3.1 Materials and instruments

Several chemicals and reagents were used in this study, and they are listed in Table 3.1, together with the names of the suppliers and the percentage purity.

Table 3.1: List of chemicals and reagents.

Name of chemical	Formula	Supplier	Percentage purity
Deionized water	H <sub>2</sub> O		
MilliQ water	H <sub>2</sub> O		Type 1 (ultrapure)
1-Methyl imidazole	CH <sub>3</sub> C <sub>3</sub> H <sub>3</sub> N <sub>2</sub>	Merck	≥ 99
1-Butanol	C <sub>4</sub> H <sub>9</sub> OH	Merck	≥ 99
1-Octanol	C <sub>8</sub> H <sub>17</sub> O H	Sigma-Aldrich	≥ 99
Acetonitrile	CH <sub>3</sub> CN	Sigma-Aldrich	≥ 99.9
Acetonitrile-d <sub>3</sub>	CD <sub>3</sub> CN	Sigma-Aldrich	99.8 atom % D
Hexane	C <sub>6</sub> H <sub>14</sub>	Sigma-Aldrich	95
Diethyl ether	(C <sub>2</sub> H <sub>5</sub> ) <sub>2</sub> O	Sigma-Aldrich	≥ 99.0
Levulinic acid	C <sub>5</sub> H <sub>8</sub> O <sub>3</sub>	Merck	98
Fumaric acid	C <sub>4</sub> H <sub>4</sub> O <sub>4</sub>	Sigma-Aldrich	≥ 99.0
Methanesulfonic acid	CH <sub>3</sub> SO <sub>3</sub> H	Merck	99
Sodium chloride	NaCl	Sigma-Aldrich	5.0 M
Silver nitrate	AgNO <sub>3</sub>	Merck	99.8 – 100.5
6-Chlorohexanol	C <sub>6</sub> H <sub>13</sub> Cl O	Sigma-Aldrich	96
Butyl levulinate	C <sub>9</sub> H <sub>16</sub> O <sub>3</sub>	Aldrich	98

Ethyl levulinate	C <sub>7</sub> H <sub>12</sub> O <sub>3</sub>	Sigma-Aldrich	≥ 98
Sodium trifluoroacetate	CF <sub>3</sub> COO Na	Sigma-Aldrich	98
Helium	He	Afrox	99.999
Hydrogen	H <sub>2</sub>	Afrox	99.999
Potassium dihydrogen phosphate	KH <sub>2</sub> PO <sub>4</sub>	Sigma-Aldrich	≥ 99.0
Lithium bis(trifluoromethanesulfonyl)imide	Li[NTf <sub>2</sub> ]	IOLITEC	99

Instruments used in this study are listed in Table 3.2.

Table 3.2: List of instruments

Instrument	Make	Model
Balance	Optika	B214Ai
pH meter	Thermo Scientific	Orion Star A211
Karl-Fischer titrator	Mettler Toledo	C105 Coulometric KF Titrator
FTIR spectrophotometer	SHIMADZU	IRSpirit
NMR spectrometer	Bruker	Avance Ultrashield 400 MHz
HPLC	SHIMADZU	Prominence LC-20AT + SPD-20A UV-Vis detector
GC-FID	SHIMADZU	GC-2014
Centrifuge	OHAUS	FC5706
Rotary evaporator	Chemtron	Strike 280
Shaker	IKA	HS 501D
Ultrasonic bath	MRC	DC200H
Schlenk line		

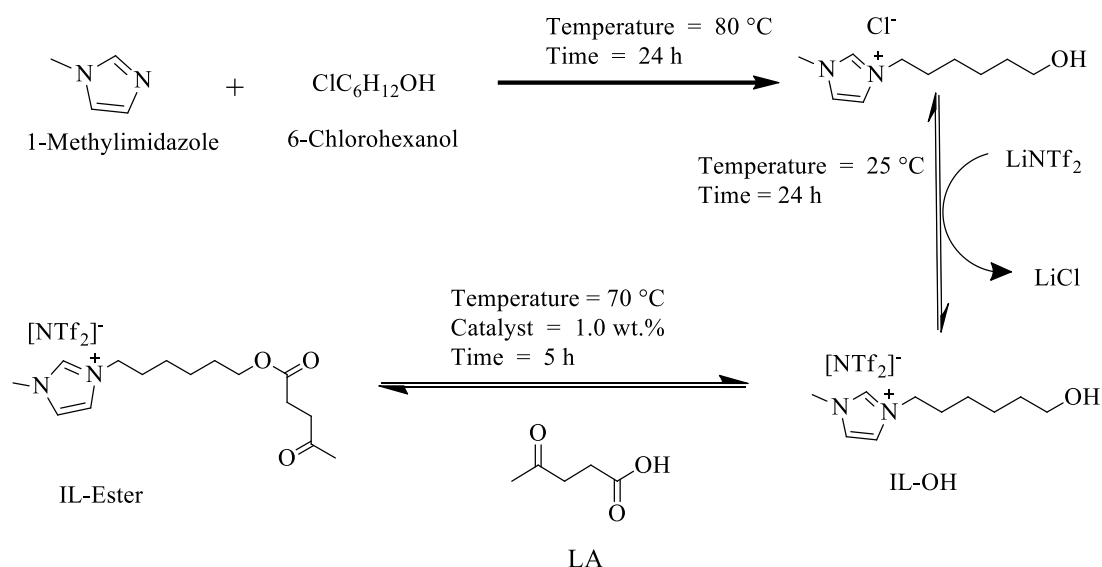
Several experiments were conducted to address the objectives of the study. The details of the experimental conditions are presented in the sub-sections herein.

### 3.2 Preparation of ILs

The ILs used in this study were derived from 1-(6-hydroxyhexyl)-3-methylimidazolium chloride ([HOC<sub>6</sub>MIM]Cl), which in turn had been synthesized via the alkylation of 1-methylimidazole using 6-chlorohexanol. The IL 1-(6-hydroxyhexyl)-3-



methylimidazolium chloride was converted to 1-(6-hydroxyhexyl)-3-methylimidazolium bis(trifluoromethanesulfonyl)imide ([HOC<sub>6</sub>MIM][NTf<sub>2</sub>]; IL-OH) via an ion exchange reaction where chloride ions were exchanged for bis(trifluoromethanesulfonyl)imide to induce hydrophobicity. The IL 1-(hexyl-4-oxopentanoate)-3-methylimidazolium bis(trifluoromethanesulfonyl) imide (IL-Ester) was prepared by esterification of IL-OH with levulinic acid, in the presence of 1.0 wt.% methanesulfonic acid as a catalyst. Scheme 3.1 summarizes the different reactions employed for the synthesis of the respective ILs.



Scheme 3.1: Synthesis of functionalized ILs.

### 3.2.1 Alkylation reaction

The alkylation reaction was carried out in a round-bottomed flask at 80 °C. A separating funnel was used to add 50.0780 g (0.6100 mol) of 1-methylimidazole into the flask containing 100.0852 g (0.7326 mol) of the alkylating agent, 6-chlorohexanol while stirring. The flask was heated in an oil bath at 80 °C for 24 h. The reaction mixture turned yellow and became highly viscous. The product was isolated from unreacted materials by extracting the unreacted materials in a continuous setup of a modified Soxhlet extraction. Diethyl ether was used as the extraction solvent at 40 °C for 12 h. The residual solvent in the product was removed with a rotary evaporator at a pressure and temperature of 72 mbar and 40 °C, respectively. The IL was transferred onto a Schlenk line for further drying under vacuum at 60 °C for another 8 h. The dried IL was

stored in a tightly closed bottle. A yield of 129.2566 g (0.5929 moles) of [HOC<sub>6</sub>MIM]Cl was achieved and this is equivalent to 97.2% yield. The product was identified by means of <sup>1</sup>H NMR spectrometry. Furthermore, the purity of the IL was also derived from <sup>1</sup>H NMR data. The chemical shifts, splitting patterns, and integration of signals identified in the <sup>1</sup>H NMR spectrum were consistent with the anticipated response.

### 3.2.2 Ion-exchange reaction

The precursor IL, [HOC<sub>6</sub>MIM]Cl, was converted into [HOC<sub>6</sub>MIM][NTf<sub>2</sub>] via an ion exchange reaction. In this case, 145.1872 g (0.5057 mol) of Li[NTf<sub>2</sub>] were dissolved in 50.0 cm<sup>3</sup> of deionized water in a 500.0 cm<sup>3</sup> round-bottomed flask. Quantitatively, 92.1728 g (0.4214 mol) of [HOC<sub>6</sub>MIM]Cl were added to the flask. The mixture was gently stirred at room temperature for 24 h after which it became biphasic. The mixture was transferred into a separating funnel and allowed to settle, and the phases separated. The IL phase formed the bottom layer, and it was extracted with 5 × 100 cm<sup>3</sup> portions of deionized water for the purpose of purification. The aqueous extracts were tested by the addition of a drop of 0.1 M AgNO<sub>3</sub>, and extraction continued until the silver chloride test was negative. The resulting IL-OH was dried using the same procedure as for the [HOC<sub>6</sub>MIM]Cl. The yield of IL-OH was found to be 183.4974 g (0.3960 moles) which is equivalent to a practical yield of 93.96%.

The purity of the IL-OH was confirmed by testing for the water content as well as the halide content. The water content was measured by a coulometric Karl-Fischer titration as described in Section 3.3.3. The halide content was determined by making use of the Nessler's test as described by Stark et al. [146]. A commercial 5.0 M NaCl standard solution was used to prepare a series of analytical standards in the concentration range of 10 – 100 mg L<sup>-1</sup>. Separately, 30 cm<sup>3</sup> of each standard was added into a Nessler tube containing 0.5 cm<sup>3</sup> of 65% HNO<sub>3</sub> and 1.0 cm<sup>3</sup> of 0.01 M AgNO<sub>3</sub>. The volume of each tube was made up to 50.0 cm<sup>3</sup> with deionized water. The tubes were left to stand in the dark for 30 minutes.

The analytical sample was prepared by dissolving 1.5321 g (0.0033 mol) of IL-OH in 20 cm<sup>3</sup> of ethanol and mixing with 0.5 cm<sup>3</sup> 65% HNO<sub>3</sub> and 1.0 cm<sup>3</sup> 0.01 M AgNO<sub>3</sub>. The tube was also placed in a dark cupboard. The opacity of the analytical sample was compared against those of the analytical standards to estimate the chloride content of the

analytical sample. It was observed that the analytical sample was more transparent and clearer than the analytical standard with a nominal concentration of 10.0 mg L<sup>-1</sup>. Therefore, it could be concluded that if there were any chloride ions present at all, the IL-OH contained chloride ions below 10.0 mg L<sup>-1</sup>.

### 3.2.3 Esterification reaction

IL-Ester was prepared from the solvent-free esterification of IL-OH with levulinic acid. In this case, 100.0 g (0.2158 mol) of IL-OH was weighed into a 250.0 cm<sup>3</sup> round-bottomed flask. A mass of 30.0704 g (0.2589 mol) of levulinic acid was added to the round-bottomed flask with the aid of a separating funnel whilst stirring gently. When the mixture was homogeneously mixed, 1.3007 g (0.0135 mol) of methanesulfonic acid was added to the flask as a catalyst. The round-bottomed flask was connected to a reflux condenser and transferred into an oil bath preheated to 70 °C. The reaction maintained a single phase throughout. After 5 h, the reaction was monitored hourly by means of HPLC as described in Section 3.3.1. until no IL-OH could be detected. At this point the reaction was quenched by placing the flask in an ice-bath for 10 minutes.

The product, IL-Ester, was isolated from the reaction mixture by extracting the acid catalyst and the excess levulinic acid with a total of 10 × 100 cm<sup>3</sup> portions of deionized water. A neutral pH in the aqueous phase indicated the absence of any acid in the reaction product. The IL-Ester was dried on a rotary evaporator at 40 °C under reduced pressure of 72 mbar for 12 h. The IL-Ester was transferred onto a Schlenk line for further drying under vacuum at 60 °C for another 8 h. The yield of IL-Ester was found to be 1.4993 g (0.0028 moles) which is equivalent to a practical yield of 95.13%.

The purity of IL-Ester was confirmed by means of HPLC as described in Section 3.3.1. Furthermore, a coulometric Karl-Fischer titration was used to determine any residual traces of moisture present in the IL-Ester. <sup>1</sup>H NMR and FTIR spectroscopic techniques described in Sections 3.3.4 and 3.3.5, respectively, were used to confirm IL-Ester. The purity of IL-Ester was determined by HPLC.

The purities of a representative batch of the respective ILs are presented in Table 3.3. The synthesis procedures were repeated when necessary, during this study. The chloride content of all batches was < 10 mg L<sup>-1</sup>. The ionic liquids are hygroscopic, but the water contents were < 5.5 mg g<sup>-1</sup> in all cases.

Table 3.3: Purity of ILs synthesized in this study.

<b>IL</b>	<b>Halide content/mg L<sup>-1</sup></b>	<b>Water content/mg g<sup>-1</sup></b>
[HOC <sub>6</sub> MIM]Cl	Not applicable	21.34
IL-OH	< 10	5.45
IL-Ester	< 10	2.76

### 3.3 Instrumental methods

Various analytical tools were employed in this study to monitor reactions for quantitative and qualitative purposes. The analytical techniques included HPLC, GC-FID, Karl-Fischer titrations, <sup>1</sup>H NMR, <sup>13</sup>C NMR, <sup>19</sup>F NMR and FTIR.

#### 3.3.1 HPLC method

A SHIMADZU high performance liquid chromatograph (HPLC) equipped with an LC-20AT pump and SPD-20A UV-Vis detector was used for the determination and quantification of IL-OH, IL-Ester, levulinic acid and fumaric acid. High purity samples of the analytes (as per <sup>1</sup>H NMR spectroscopy) were used to prepare reference standards. Similar chromatographic conditions were used for all the four analytes. A mobile phase of 70% acetonitrile and 30% 0.035 M phosphate buffer (70:30 v/v%) was used to elute the analytes from a ZORBAX HILIC Plus column (4.6 × 100 mm, 3.5 μm) at a flowrate of 1.0 cm<sup>3</sup> min<sup>-1</sup>. For each analysis, 20 μL of sample was injected onto the column and the detection wavelength was set at 211 nm.

##### 3.3.1.1 Calibration

Calibration curves were developed for both IL-OH and IL-Ester. For IL-OH and IL-Ester, stock solutions of 1000 μg cm<sup>-3</sup> were freshly prepared during inter-day calibration. Working standard solutions in the concentration range between 0 – 200 μg cm<sup>-3</sup> were prepared from accurate dilution of the stock solutions. The standard solutions were analysed based on the method described in Section 3.3.1.

In the case of fumaric acid, a standard stock solution of 500.0 mg L<sup>-1</sup> was prepared using commercially available fumaric acid with a purity of ≥ 99.0%. Analytical standards in

the concentration range 5.0 – 50.0 µg cm<sup>-3</sup> were prepared from the standard stock solution by dilution.

The calibration curve for levulinic acid was made by using analytical standards prepared from a 0.026 M standard stock solution of 98% levulinic acid. Analytical standards in the concentration range 30 – 305 mg L<sup>-1</sup> were prepared from the standard stock solution by dilution.

The calibration data was analysed by linear regression at the 95% confidence level using the Analysis ToolPak in Microsoft Excel. Calibration plots and unweighted residuals plots were established. The limits of detection (LOD) and limits of quantification (LOQ) of the HPLC method were calculated from the standard error of the slope ( $\sigma$ ) and the calibration slope ( $S'$ ) as shown in Equations 3.1 and 3.2 respectively. See Appendix A for the HPLC calibration curves for all analytes.

$$LOD = 3.3 \times \frac{\sigma}{S'} \quad (3.1)$$

and

$$LOQ = 10 \times \frac{\sigma}{S'} \quad (3.2)$$

#### 3.3.1.2 Quality assurance

The reliability of the HPLC analytical method is dependent upon reproducibility and repeatability of calibration parameters. The calibration parameters were retention time, linear range, accuracy, recovery, LOD, and LOQ. For each analyte, calibration runs were conducted on three different days to assess inter-day and intra-day precision and freshly prepared solutions with different concentrations of the analyte were used in each instance.

#### 3.3.2 GC method

A SHIMADZU GC-2014 instrument was used to monitor the transesterification reaction. Butanol, ethyl levulinate and butyl levulinate were eluted on a Zebron ZB-WAXPLUS capillary column (30 m × 0.25 mm × 0.25 µm) using helium as the carrier gas at a flow rate of 1.27 cm<sup>3</sup> min<sup>-1</sup>. Table 3.4 shows the column temperature programme. In each analysis, 0.5 µL of sample was injected at 250 °C using the split mode at a split ratio of 25. The hydrogen flame ionization detector was set at 250 °C. The retention times for

butanol, ethyl levulinate and butyl levulinate were 4.19, 8.72, and 10.32 minutes, respectively.

Table 3.4: Temperature programme for GC method.

	Heating rate/°C min <sup>-1</sup>	Temperature/°C	Hold time/min
0	–	60	1.00
1	15	180	2.00
2	10	210	5.00

### 3.3.2.1 GC calibration

The GC method focussed on the determination of multiple analytes; thus, a multi-component calibration was established. Butanol and butyl levulinate were the analytes of interest while ethyl levulinate was considered as the internal standard. Standard stock solutions of 1000 µg cm<sup>-3</sup> were prepared separately for the three analytes from commercial samples. Aliquots of the stock solutions were mixed and diluted to specific volumes to obtain analytical standards containing varied concentrations of butanol and butyl levulinate while the concentration of ethyl levulinate remained constant throughout. At each concentration level, the peak area ratio and the concentration ratio were calculated according to Equations 3.3 and 3.4, respectively.

$$\text{Peak area ratio} = \frac{\text{Peak area of analyte}}{\text{Peak area of internal standard}} \quad (3.3)$$

and

$$\text{Concentration ratio} = \frac{\text{Analyte concentration}/\mu\text{g cm}^{-3}}{\text{Internal standard concentration}/\mu\text{g cm}^{-3}} \quad (3.4)$$

The calibration curve was obtained by plotting the peak area ratio (Y) against the concentration ratio (X).

### 3.3.2.2 Quality assurance

The quality of the calibration data was derived from the linearity of the calibration curves as well as the retention times of the analytes. Intraday and inter-day analysis of replicate analytical standards assessed the repeatability and precision in the determination of the target analytes. The calibration parameters for the GC method were retention time, linear

range, accuracy, recovery, LOD, and LOQ, respectively. The GC calibration curves are presented in Appendix B.

### 3.3.3 Karl-Fischer titration

The water content of the IL phases was measured by means of a Mettler Toledo C105 Coulometric KF Titrator. The KF Titrator was calibrated with ROMIL KF WS1 Hi-Dry KF calibrant. The calibrant used was an anisole/propylene carbonate matrix with a nominal water concentration of 1.0 mg g<sup>-1</sup>. All measurements were conducted in triplicate and are reported as averages.

### 3.3.4 Nuclear magnetic resonance spectrometry

The products of transesterification were identified and confirmed by means of <sup>1</sup>H NMR spectroscopy. In each case, the sample was dissolved in 0.8 cm<sup>3</sup> of acetonitrile-d<sub>3</sub> and transferred into an NMR tube for analysis. The spectra were recorded on a Bruker Avance Ultrashield 400 MHz spectrophotometer. Topspin 4.0.9, a Bruker NMR propriety software, was used to analyse the spectra. In the case of <sup>19</sup>F NMR measurements, approximately 20.0 mg of sodium trifluoroacetate (CF<sub>3</sub>COONa) was added as an internal reference material. NMR data was used to determine molar ratio  $\left(\frac{n_x}{n_y}\right)$ , of IL-OH:IL-Ester after the transesterification reaction by using Equation 3.5,

$$\frac{n_x}{n_y} = \frac{I_x}{I_y} \times \frac{N_y}{N_x} \quad (3.5)$$

where I and N, refer to the integral of the selected signal, and the number of nuclei giving rise to the signal in compounds x and y, respectively. The same approach was also used to quantify the [NTf<sub>2</sub>]<sup>-</sup> anion with respect to CF<sub>3</sub>COONa.

### 3.3.5 FTIR spectroscopy

FTIR spectra of the regenerated IL-OH and butyl levulinate were recorded by means of a SHIMADZU IRSpirit spectrophotometer in the range 4000 – 400 cm<sup>-1</sup>. The spectrophotometer houses an attenuated total reflectance accessory (ATR). All the samples in this study were in the liquid state and for each measurement a drop of the liquid sample was dispensed onto the diamond crystal on the instrument. Table 3.5 summarizes the instrument acquisition parameters applied on the spectrophotometer.

Table 3.5: Instrument acquisition parameters for FTIR spectroscopy.

Parameter	Description
Range/cm <sup>-1</sup>	4000 – 400
Resolution/cm <sup>-1</sup>	16
Number of scans	64
Apodization	Happ-Genzel

### 3.4 Mutual solubility studies

The solubility of water in the IL, as well as the solubility of the IL in water, was considered. The effect of the pH of the aqueous phase and the effect of the amount of the initial acid catalyst (used in the subsequent esterification experiments) in the aqueous phase were investigated. Samples collected from the aqueous phase were analysed by HPLC for the detection and quantification of IL-OH. Simultaneously, the water content of the IL phase was determined by means of Karl-Fischer titration.

#### 3.4.1 Effect of pH on mutual solubility

The mutual solubilities of IL-OH or IL-Ester with water were measured as a function of initial pH of the aqueous phase. The pH was measured and monitored with a pH meter. The initial pH of the aqueous phase was adjusted to be in the range of 1.0 – 7.0 by the addition of appropriate amounts of either 0.05 M H<sub>2</sub>SO<sub>4</sub> or 0.05 M NaOH. In each case, 2.0 cm<sup>3</sup> of IL-OH and aqueous phase were measured in 5.0 cm<sup>3</sup> vials. The contents of the vials were mixed vigorously on a horizontal shaker for 12 h at 25 °C. The vials were left to stand for 5 hours to allow for phase separation prior to sampling and analysis of both the aqueous and IL phases. The aqueous phase was analysed for IL-OH using the HPLC method described in Section 3.3.1. while the IL phase was analysed for water content using the KF titration as described in Section 3.3.3. The experiment was repeated in duplicate, and average values are reported.

#### 3.4.2 Effect of methanesulfonic acid concentration on mutual solubility

The effect of acid catalyst concentration on the mutual solubility was investigated by varying the concentration of methanesulfonic acid in the aqueous phase. The initial



concentration of methanesulfonic acid was kept in the range between 0 – 12 wt.% of the aqueous phase. In each case, 2.0 cm<sup>3</sup> of IL-OH and aqueous phase were measured into 5.0 cm<sup>3</sup> vials. The contents of the vials were mixed vigorously on a horizontal shaker for 12 hours at room temperature. The vials were left to stand for 5 hours to allow for phase separation prior to sampling and analysis of both the aqueous and the IL phases. The aqueous phase was analysed for IL-OH using the HPLC method described in Section 3.3.1 while the IL phase was analysed for water content by the KF titration as described in Section 3.3.3. The experiments were conducted in duplicate.

### **3.5 Octanol-water partition coefficient measurements**

The experiments to determine the K<sub>OW</sub> values of IL-Ester and IL-OH were conducted using the shake-flask method. K<sub>OW</sub> values were measured at different concentrations for each IL. Aqueous solutions of IL-OH and IL-Ester were prepared in the concentration ranges of 11 – 210 mg L<sup>-1</sup> and 7 – 90 mg L<sup>-1</sup>, respectively. The lowest concentration for each IL in the aqueous phase was such that it was greater than the LOD and LOQ of the IL with respect to the HPLC analytical method described in Section 3.3.1.

The ILs were partitioned between the octanol and the aqueous phases using a 1:1 (v/v) ratio of the solvents at 25 °C. In each case, 10 cm<sup>3</sup> of the aqueous phase was added to a 50 cm<sup>3</sup> vial containing 10 cm<sup>3</sup> of 1-octanol. The vials were agitated vigorously for 3 h using a horizontal shaker. An OHAUS Frontier™ 5706 centrifuge was used to separate the organic phase from the aqueous phase. Samples were collected from the aqueous phase and analysed immediately without further treatment. The concentration of the IL in the aqueous phase was determined using the HPLC method described in Section 3.3.1. The amount of IL partitioned into the octanol phase was subsequently calculated by mass balance. K<sub>OW</sub> was then calculated from the equilibrium concentrations of the ILs in both phases.

### **3.6 Extraction of carboxylic acids using IL-OH**

Two carboxylic acids, namely levulinic acid or fumaric acid, were extracted from their aqueous solutions using IL-OH. In each case, both physical and chemical extraction were investigated. The difference between the physical and chemical extraction experiments was the incorporation of methanesulfonic acid as a catalyst during chemical extraction. Otherwise, all other experimental conditions were identical. The model solutions of

levulinic or fumaric acid were designed based on arbitrary concentration values within the productivities of microorganisms during fermentation.

### **3.6.1 Preparation of levulinic acid solutions**

A stock solution of 0.32 M levulinic acid was prepared from 98% levulinic acid and deionized water. The stock solution was diluted to prepare solutions within the concentration range 0.05 – 0.32 M. The physical and chemical extraction of levulinic acid from aqueous solution using IL-OH was investigated using the prepared solutions.

### **3.6.2 Preparation of fumaric acid solutions**

A stock solution of 4.002 g L<sup>-1</sup> fumaric acid was prepared from commercially available fumaric acid and deionized water. The stock solution was diluted to prepare solutions in the concentration range 0 – 4.002 g L<sup>-1</sup>. The physical and chemical extraction of fumaric acid aqueous solution using IL-OH from was investigated using the prepared solutions.

### **3.6.3 Physical extraction**

Levulinic acid or fumaric acid were physically extracted from aqueous solution in the absence of a chemical reaction. In each case, 2.0 cm<sup>3</sup> of IL-OH were added into a 5.0 cm<sup>3</sup> vial containing 2.0 cm<sup>3</sup> of the respective acid solution. The vials were left to shake vigorously on a horizontal shaker for 12 h at room temperature. The vials were left to stand to allow for phase separation prior to sampling and analysis of both the aqueous and the IL phases. The aqueous phase was analysed for either levulinic acid or fumaric acid and IL-OH using the HPLC method described in Section 3.3.1. The amount of acid extracted by the IL was calculated using mass balance. The IL phase was analysed for water content by means of the KF titration described in Section 3.3.3.

### **3.6.4 Reactive extraction**

The reactive extraction of levulinic acid or fumaric acid from aqueous solution using IL-OH was investigated by addition of methanesulfonic acid as a catalyst. In each case, 2.0 cm<sup>3</sup> of IL-OH were added into a 5.0 cm<sup>3</sup> vial containing 2.0 cm<sup>3</sup> of the respective acid solution. Methanesulfonic acid was added to the vial at a proportion of 1.0 wt.% of the IL-OH. The vials were placed on the shaker and allowed to shake vigorously for 12 h at room temperature. The vials were left to stand to allow for phase separation prior to sampling and analysis of both the aqueous and the IL phases. The aqueous phase was

analysed for the carboxylic acid and IL-OH using the HPLC method described in Section 3.3.1. The amount of the  $[\text{NTf}_2]^-$  anion in the aqueous phase were determined by means of  $^{19}\text{F}$  NMR as described in Section 3.3.4. A mass balance was used to calculate the amount of acid extracted by the IL. The water content of the IL phase after the extraction was determined by KF titration as described in Section 3.3.3.

The feasibility of aqueous phase esterification of IL-OH with levulinic acid was investigated at different levels of water loading at 70 °C in the presence of 1.0 wt.% catalyst loading. The water loading was varied between 5.7 – 520.7 mg g<sup>-1</sup>. The water loading was done with respect to the amount of water per gram of IL-OH. The conversion of IL-OH to IL-Ester was monitored by means of HPLC as described in Section 3.3.1.

### **3.7 Transesterification of IL-Ester with butanol**

Either ester hydrolysis or transesterification can be considered for the regeneration of IL-OH. Ester hydrolysis results in the carboxylic acid as product, which would have to be separated from IL-OH by extraction, since carboxylic acids are relatively difficult to distil or sublime. It can be foreseen that due to the relatively similar nature of the two components, selective extraction would be challenging. Hence, in this work, transesterification was considered to regenerate the IL-OH while producing a relatively volatile organic ester. The transesterification of IL-Ester with n-butanol was investigated. Batch experiments were conducted to determine and establish the effects of selected variables on the transesterification reaction.

#### **3.7.1 Transesterification procedure**

The variables for the transesterification reaction were the molar ratio of reactants, reaction temperature, and amount of catalyst. Commercially available methanesulfonic acid of high purity ( $\geq 99\%$ ) was used as the catalyst for the reaction. Measured quantities of reactants and catalyst were added to a round-bottomed flask and connected to the condenser. The flask was heated to the selected temperature by means of a continuously stirred oil bath.

##### *3.7.1.1 Effect of reactant molar ratio on the transesterification reaction*

The effect of molar ratio of reactants on the transesterification reaction was investigated at 85 °C in the presence of methanesulfonic acid as a catalyst. The catalyst loading was

at 1.0 wt.% relative to IL-Ester. In each case, 10 g (0.0181 mol) of IL-Ester was accurately weighed into a two-necked round-bottomed flask. This was followed by the addition of a specific amount of butanol to make a molar ratio of IL-Ester to butanol of 1:3 and 1:10 respectively. The acid catalyst was then added to the flask. The reaction mixture formed a completely homogeneous phase. The reaction mixture was placed in an oil-bath preheated to 85 °C. Samples were withdrawn from the reaction mixture after 10, 30, 60, 120, 180, 240, 300, 360 and 420 minutes and analysed by means of the HPLC method described in Section 3.3.1.

#### *3.7.1.2 Effect of reaction temperature on transesterification*

The effect of temperature on the transesterification of IL-Ester with butanol was investigated using a constant alcohol to IL-Ester ratio of 1:10 and a catalyst loading of 1.0 wt.% relative to IL-Ester. The reaction was carried out at 85 and 115 °C. In each case, 10 g IL-Ester was added to the two-necked round-bottomed flask and preheated to the selected temperature. Exactly 16.5 cm<sup>3</sup> (0.18 mol) of butanol were added to the flask, forming a completely homogeneous phase. Samples were withdrawn from the reaction mixture after 10, 30, 60, 120, 180 and 240 minutes and analysed by means of the HPLC method described in Section 3.3.1.

#### *3.7.1.3 Effect of catalyst loading on the transesterification reaction*

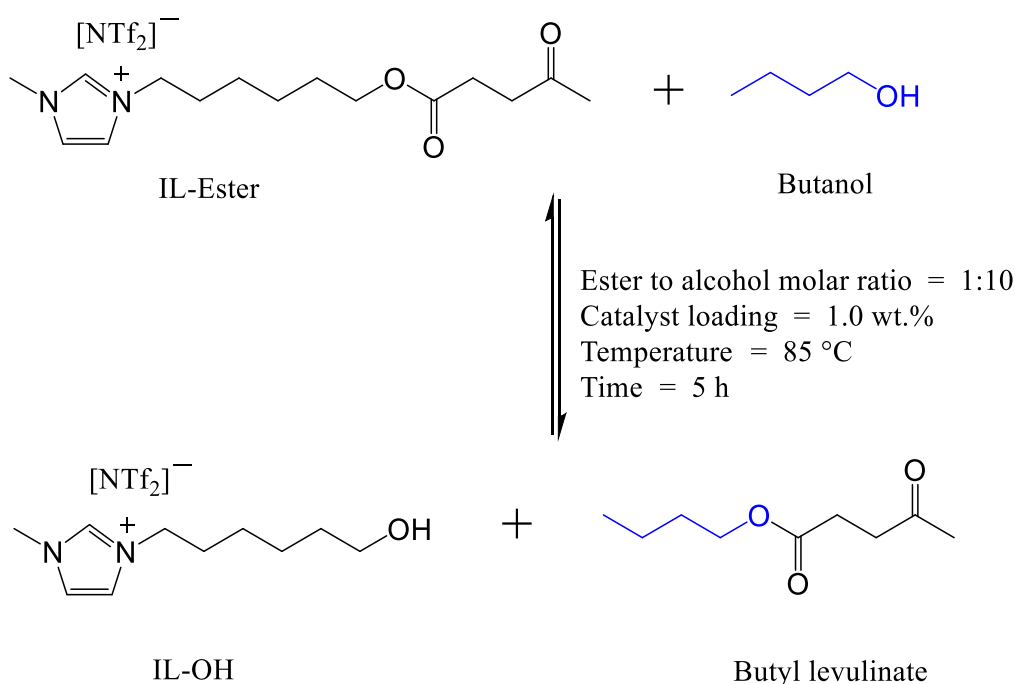
The effect of the amount of catalyst on the transesterification reaction was investigated at three different catalyst loadings. Methanesulfonic acid was chosen as the catalyst for this study and the amount of the catalyst was varied from 0 – 5.0 wt.% of the total reaction mixture. In each case, the molar ratio of IL-Ester to butanol and the reaction temperature were kept constant at 1:10 and 85 °C, respectively.

In the presence of 0 wt.% catalyst loading, addition of butanol to IL-Ester resulted in the formation of an emulsion. When the catalyst loadings were increased to 1.0 wt.% and 5.0 wt.% respectively, the reaction mixture formed a homogeneous phase.

Samples were withdrawn from the reaction mixture after 10, 30, 60, 120, 180 and 240 minutes and analysed by means of the HPLC method described in Section 3.3.1.

### 3.7.2 Sample handling and analysis

All the samples that were collected during the transesterification reaction were immediately cooled in an ice-bath and analysed immediately. In each case, 0.5 mg of the sample was weighed into a 10.0 cm<sup>3</sup> volumetric flask and diluted to the mark with the mobile phase for the HPLC method described in Section 3.3.1. The diluted samples were analysed for IL-OH and IL-Ester. The conversion and yield of the transesterification were calculated from this data. The transesterification of IL-Ester with butanol is shown in Scheme 3.2.



Scheme 3.2: Transesterification of IL-Ester with butanol in the presence of methanesulfonic acid as a catalyst.

### 3.7.3 Purification of the reaction mixture

A transesterification reaction was conducted using an IL-Ester: butanol molar ratio of 1:10 at 85 °C in the presence of 1 wt.% methanesulfonic acid catalyst. The reaction was left to proceed for 5 h prior to quenching. At this point, the flask contained a mixture of unreacted butanol, butyl levulinate, IL-OH, unreacted IL-Ester and methanesulfonic acid. The excess butanol was removed from the reaction mixture by means of vacuum distillation at 40 °C for 1 h. The recovered butanol was quantified gravimetrically and then its purity determined by GC-FID. A 20 µL sample of the recovered butanol was

dissolved in 10.0 cm<sup>3</sup> of hexane and analysed on the GC-FID using the method described in Section 3.3.2.

After the removal of excess butanol, a mixture of methanesulfonic acid, butyl levulinate, IL-Ester, and IL-OH was left behind. Butyl levulinate was extracted from the mixture by means of solvent extraction using diethyl ether as the solvent. The extraction was repeated five times with 50 cm<sup>3</sup> portions of fresh diethyl ether. The extracts were combined in a 1 000 cm<sup>3</sup> pear-shaped evaporating flask and the butyl levulinate was isolated from the extract on a rotary evaporator at 1 atm pressure and 40 °C for 1 h. The butyl levulinate was quantified gravimetrically and its purity determined using the GC-FID method described in Section 3.3.2. The remaining mixture of IL-OH, IL-Ester and methanesulfonic acid was considered as the regenerated IL thereafter. The molar ratio of IL-OH:IL-Ester was estimated from <sup>1</sup>H NMR and <sup>19</sup>F NMR spectrometry as described in Section 3.3.4. The NMR data was corroborated by analyzing the regenerated IL using the HPLC method that is described in Section 3.3.1.

### **3.8 Chapter summary**

This chapter described the experimental methodologies and analytical techniques that were employed within the current study. The experiments are presented in a cumulative order with to address the research questions. The following set of experiments were conducted,

- i. Preparation of IL-OH and IL-Ester
- ii. Mutual solubility studies for the [IL-OH + water] system
- iii. Determination of K<sub>ow</sub> values for IL-Ester and IL-OH
- iv. Extraction of levulinic acid and fumaric acid from aqueous solution
- v. Acid-catalysed transesterification of IL-Ester with butanol and recovery of IL-OH

The synthesized ILs are not available on the commercial market, however, their physicochemical properties were presented in the MSc dissertation preceding the current study [51]. All the respective chemical analyses conducted herein were carried out using validated analytical methods.

## 4 Results and Discussion

A detailed description of the results and observations made during this study are reported herein. The chapter discusses the preparation of IL-OH, the application of IL-OH as a reactive extractant for carboxylic acids and, finally, the transesterification of IL-Ester. Various analytical methods such as HPLC, FTIR and GC were developed and validated for the purpose of monitoring the respective experiments in this study. The behaviour and performance of IL-OH in the reactive extraction of carboxylic acids from aqueous solution is also discussed herein. Finally, the regeneration of IL-OH via the transesterification of IL-Ester with butanol is presented.

### 4.1 Preparation of ILs

The ILs used in this study were first synthesized. A detailed description on the preparation and characterization of  $[\text{HOC}_6\text{MIM}]\text{Cl}$ , IL-OH and IL-Ester can be found in Section 3.2 as well as in the MSc dissertation, where physico-chemical properties of IL-OH and IL-Ester were determined [51]. The organic purities of IL-OH and IL-Ester were estimated from  $^1\text{H}$  NMR to be at least 97.42 and 91.94% respectively.

The cation:anion ratio in IL-Ester was investigated to determine whether there was any leaching of either ion during the esterification reaction. For this purpose, a quantitative  $^{19}\text{F}$  NMR analysis of assumably pure IL-Ester was conducted in the presence of  $\text{CF}_3\text{CO}_2\text{Na}$  as an internal standard. The experimental quantities of IL-Ester and  $\text{CF}_3\text{CO}_2\text{Na}$  in the analytical sample are presented in Table 4.1.

Table 4.1: Sample preparation for quantitative  $^{19}\text{F}$  NMR analysis.

Analyte	Mass/g	Moles	Mole fraction	Theoretical integral
IL-Ester	0.0121	$2.155 \times 10^{-5}$	0.127	0.29
$\text{CF}_3\text{CO}_2\text{Na}$	0.0202	$1.485 \times 10^{-4}$	0.873	

The  $^{19}\text{F}$  NMR spectrum of IL-Ester in the presence of  $\text{CF}_3\text{CO}_2\text{Na}$  as an internal standard is presented in Figure 4.1. Chemical shifts observed at -76.1157 and -80.0211 ppm are due to the fluorine nuclei in  $\text{CF}_3\text{CO}_2\text{Na}$  and IL-Ester, respectively. The chemical structures of the anions are shown adjacent to the respective peaks on the spectrum.

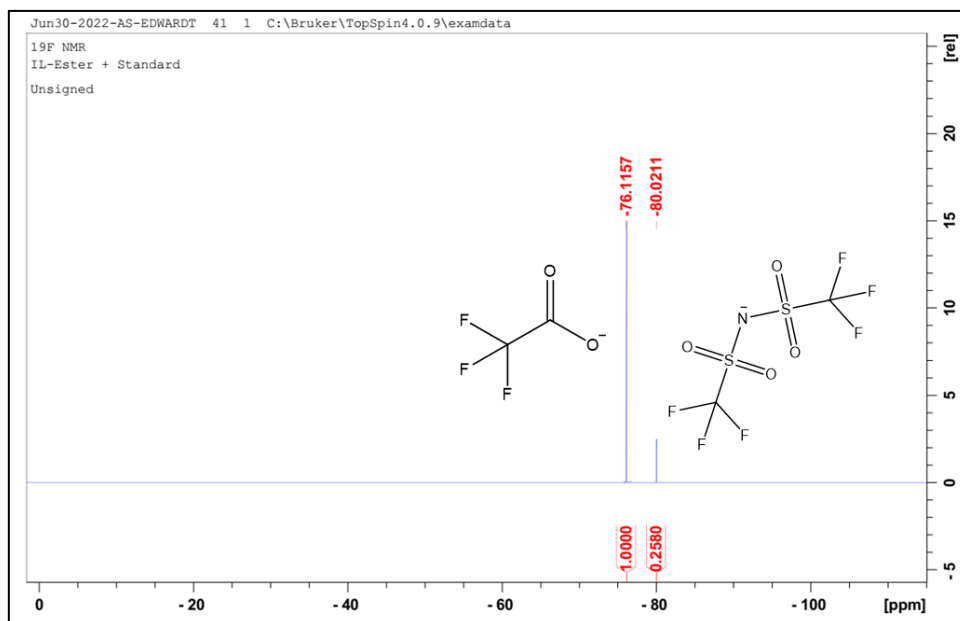


Figure 4.1:  $^{19}\text{F}$  NMR spectrum of IL-Ester and  $\text{CF}_3\text{CO}_2\text{Na}$  in acetonitrile- $\text{d}_3$ , with  $\text{CF}_3\text{CO}_2\text{Na}$  acting as the internal standard.

The mole fraction of IL-Ester in the analytical sample was determined from the  $^{19}\text{F}$  NMR spectrum using Equation 3.5. The mole fraction of IL-Ester in the sample was found to be 0.129, which is consistent with the value reported in Table 4.1. This suggests that IL-Ester contains evenly distributed ions. The differences in the mole fraction values determined gravimetrically and from the spectrum can be attributed to analytical error during sample preparation.

## 4.2 HPLC method development

The performance of IL-OH in the reactive extraction of carboxylic acids was monitored by means of an HPLC method. This analytical method was used to monitor the following experiments:

- Mutual solubility between IL-OH and water
- Reactive extraction of fumaric and levulinic acids using IL-OH
- Transesterification of IL-Ester with butanol
- Composition of regenerated IL

The nature of the experiment at hand determined which analyte(s) was/were to be monitored during that experiment.



The chromatographic method described in Section 3.3.1 provided a reproducible elution with excellent resolution between the targeted analytes. For example, sample chromatogram showing the elution of IL-Ester and IL-OH is presented in Figure 4.2

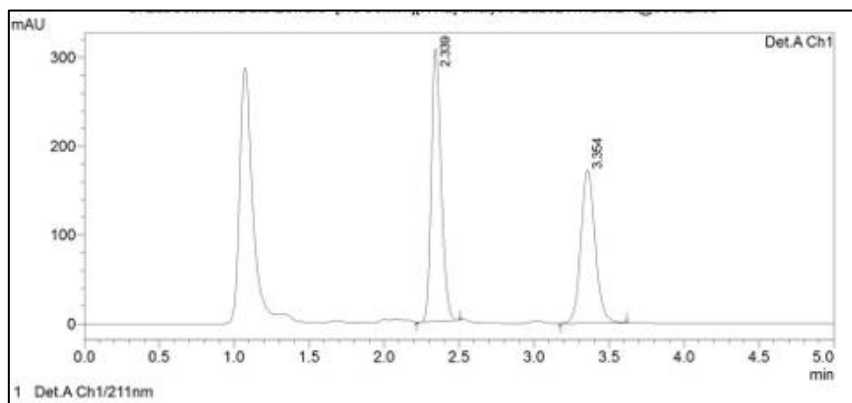


Figure 4.2: Sample chromatogram for the determination of IL-Ester and IL-OH by means of HPLC. IL-Ester and IL-OH were eluted from the column at 2.339 and 3.354 minutes, respectively. The solvent front eluted at  $t = 1.0$  minutes.

The repeatability of the HPLC analytical method was assessed by the precision in the retention times of the analytes. Five replicate samples containing a fixed concentration of IL-Ester and IL-OH were analysed and the precision on the retention times for the elution of each respective analyte is shown in Table 4.2.

Table 4.2: HPLC retention times of various analytes at 211 nm.

Analyte	Retention time/minutes					Mean	Standard deviation
	1	2	3	4	5		
Levulinic acid	1.385	1.381	1.376	1.382	1.390	1.383	0.005
Fumaric acid	1.938	1.944	1.947	1.937	1.930	1.939	0.006
IL-Ester	2.306	2.293	2.236	2.221	2.274	2.266	0.036
IL-OH	3.354	3.287	3.218	3.202	3.197	3.237	0.034

The HPLC method was primarily designed for the purpose of quantitative analysis. As a result, the calibration protocol ensured the suitability of the HPLC method in determining the concentrations of targeted analytes in the different experiments that were conducted

within the study. The calibration parameters for the HPLC determination of fumaric acid, IL-Ester, and IL-OH are presented in Table 4.3.

Table 4.3: Calibration parameters for the HPLC method.

<b>Parameter</b>	<b>Fumaric acid</b>	<b>Levulinic acid</b>	<b>IL-Ester</b>	<b>IL-OH</b>
Range/ $\mu\text{g cm}^{-3}$	5 – 50	30 – 305	20 – 180	20 – 180
Slope/mAU $\text{cm}^3 \mu\text{g}^{-1}$	170298.5	987.9	7570.0	9416.8
Standard error of slope	663.5	8.6	78.9	41.5
$R^2$	0.999	0.997	0.996	0.999
LOD/ $\mu\text{g cm}^{-3}$	0.013	0.029	0.034	0.015
LOQ/ $\mu\text{g cm}^{-3}$	0.039	0.087	0.014	0.044

The calibration curves and residuals analysis plots for the calibration data for the determination of IL-OH, IL-Ester, fumaric acid and levulinic acid are presented in Appendix A. Sample chromatograms for the elution of fumaric acid and levulinic acid are also presented in Appendix A. The precision and accuracy of the HPLC method are presented in Appendix A.

The calibration parameters indicated that the HPLC analytical method was appropriate for use in the determination of IL-OH, IL-Ester, fumaric acid and levulinic acid.

### 4.3 GC-FID method development

The GC-FID method was developed to assay the recovered butanol and as well as the butyl levulinate produced from the transesterification reaction. The repeatability of the analytical method was assessed based on the precision of the retention times for the analytes as well as the internal standards. Table 4.4 shows the retention times observed during the analysis of five replicate samples of each analyte at a fixed concentration.

A calibration was made for both butanol and butyl levulinate in the presence ethyl levulinate as an internal standard and the calibration parameters are listed in Table 4.5. Ethyl levulinate has some chemical similarity to the butyl levulinate and it was not expected to be present in the sample matrix. Furthermore, the chromatographic analysis showed a good resolution between ethyl levulinate and both analytes.

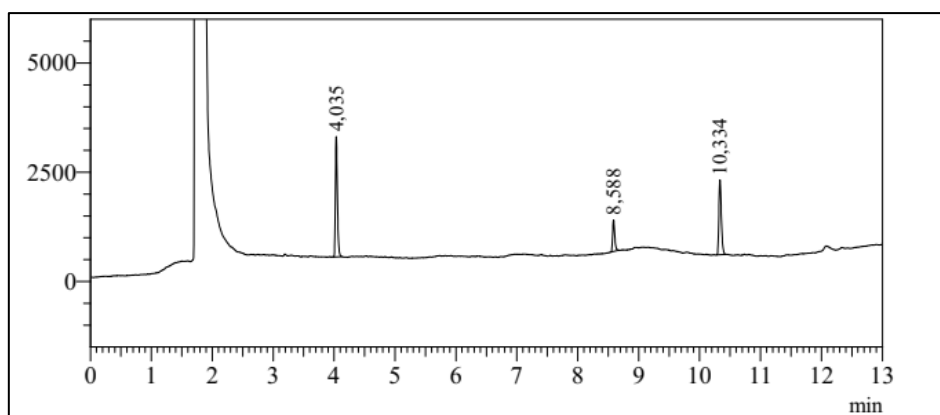


Figure 4.3: GC-FID sample chromatogram showing the elution of butanol, ethyl levulinate and butyl levulinate from a Zebron ZB-WAXPLUS column at 4.035, 8.5884, and 10.334 minutes, respectively.

Table 4.4: GC-FID retention times of butanol, ethyl levulinate and butyl levulinate.

Compound	Retention time/min					Mean	Standard deviation
	1	2	3	4	5		
Butanol	4.035	4.135	4.140	4.130	4.133	4.135	0.004
Ethyl levulinate	8.588	8.707	8.708	8.708	8.707	8.684	0.053
Butyl levulinate	10.334	10.309	10.316	10.310	10.311	10.316	0.010

Table 4.5: Calibration parameters for the GC-FID method.

Parameter	Butanol	Butyl levulinate
Calibration range/ $\mu\text{g cm}^{-3}$	15 – 200	20 – 250
Slope	1.3906	0.8897
Standard error of slope	0.0239	0.01720
$R^2$	0.9918	0.9896
LOD/ $\mu\text{g cm}^{-3}$	3.43	3.86
LOQ/ $\mu\text{g cm}^{-3}$	10.31	11.60

The precision and accuracy of the GC method are presented in Appendix B. The calibration parameters indicated that the GC analytical method was appropriate for use in the determination of butyl levulinate and butanol. Therefore, the analytical method was well-suited for the purpose of this study.

#### **4.4 Mutual solubility of IL-OH and IL-Ester with water**

The formation of a biphasic system between the aqueous phase and extractant phase is critical in the development of liquid-liquid extraction processes.

The accumulation of carboxylic acids during fermentation tends to lower the pH of the system. The optimum productivity of fermentation microbes is dependent on pH [147]. Extractive fermentation can ultimately maintain higher pH values within the system since the accumulation of the carboxylic acid is hindered. However, extractive fermentation is usually conducted at suboptimal pH ranges by using conventional molecular solvents, which may prove toxic towards the fermentation microbes [148].

During the reactive extraction of carboxylic acids, the aqueous phase initially contains the carboxylic acid to be extracted. Additionally, an acid catalyst such as methanesulfonic acid, may be needed for the reaction between IL-OH and the carboxylic acid during chemical extraction. Thus, it becomes important to understand the phase behaviour of the extraction system, i.e., the dependence of the [IL-OH + water] system on pH as well as acid catalyst concentration.

Furthermore, knowledge on the mechanism of the reactive extraction can be derived from mutual solubility measurements: the carboxylic acid molecules residing in the aqueous phase must be exposed to the extractant, IL-OH. Dependent on the solubility of IL-OH in water, the esterification reaction could either occur in the aqueous phase, the IL phase, or the interphase.

##### **4.4.1 Effect of pH on mutual solubility**

The solubility of water in an imidazolium-based IL is entirely dependent upon the formation of hydrogen bonds between the acidic hydrogen of the imidazolium cation and the oxygen of water [122, 128]. Strong hydrogen bonding tendency in the polar domains of the IL enhances the solubility of water in the IL. It can be assumed that at low pH, the cation of the IL is highly polarized, thereby increasing its tendency to form hydrogen bonds with water molecules. The solubility of water in IL-OH showed a small decrease

with increasing pH of the aqueous phase (Figure 4.4). Hence, over the pH range between 1 and 6.5 at 25 °C, the water solubility in IL-OH decreases only slightly, but clearly linearly from 61.09 mg g<sup>-1</sup> to 51.46 mg g<sup>-1</sup>.

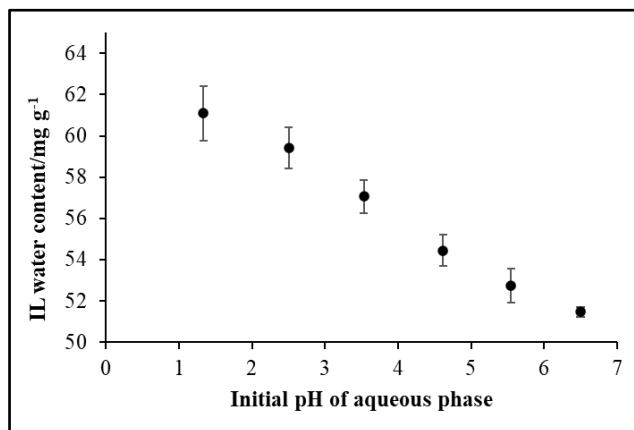


Figure 4.4: Effect of the initial pH of the aqueous phase on the amount of water dissolved in the IL-OH at 25 °C (n = 3).

Figure 4.5 shows that the mole fraction of water in IL-OH does not change significantly irrespective of the pH of the aqueous phase. Simultaneously, the solubility of IL-OH into the aqueous phase initially decreased as the pH was increased. There was a sharp decrease in the solubility of IL-OH in water between pH = 1 and pH = 2, but the change was almost negligible in the pH range between 2 – 7.

Table 4.6: Effect of initial pH of the aqueous phase on the solubility of water in IL-OH.

Parameter	pH	
	1	6.5
IL-OH water content/mg g <sup>-1</sup>	61.09	51.46
Mass of water per 100 g sample/g	6.109	5.146
Mass of IL-OH per 100 g sample/g	93.891	94.854
Mole fraction of water	0.625	0.582
Mole fraction of IL-OH	0.375	0.418

Freire et al. [122] reported similar observations on the mutual solubilities of imidazolium-based ILs with water. It has been recognized that the ability of the IL ions to interact with other solutes and solvents enhances the mutual solubility of ILs with water [128].

ILs are mesostructured materials with well-defined polar and non-polar regions. As a result, there is preferential solvation of the polar domains compared with the non-polar regions.

As shown in Figure 4.5, the solubility of IL-OH in water is relatively higher at low pH due to the increased tendency of water molecules to interact with the polar domains of the cation.

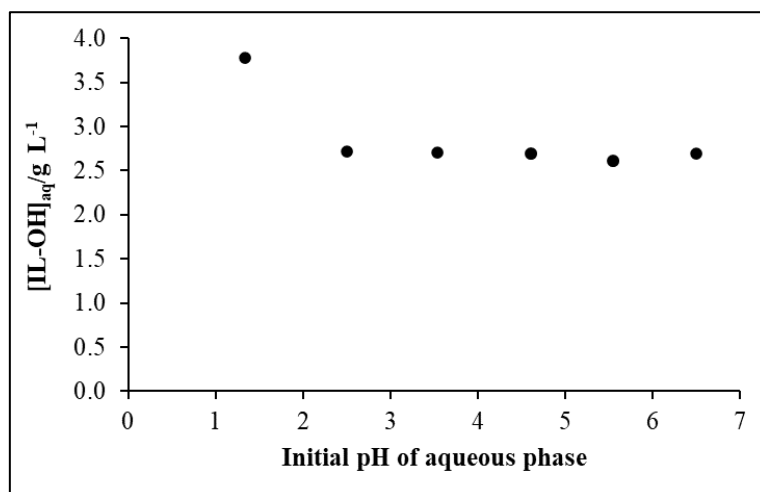


Figure 4.5: Effect of the initial pH of the aqueous phase on the solubility of IL-OH in water at 25 °C.

The polarity of IL-Ester is significantly lower than that of IL-OH. As a result, the solubility of water in IL-Ester is lower than in IL-OH. Figure 4.6 shows the influence of initial pH of the aqueous phase on the solubility of water in IL-Ester. The reduced solubility of water in IL-Ester is due to the reduced tendency of IL-Ester to form hydrogen bonds with water. This is because, as the chain length of the alkyl group increases, the charge density is reduced, thereby reducing the polarity of the IL. Secondly, the behaviour of [IL + water] systems is influenced by the formation of hydrogen bonds between water and the acidic proton of the imidazolium cation [122]. However, the long alkyl-chain in IL-Ester induces steric hindrance, reducing the interaction of the C2 -proton with water molecules.

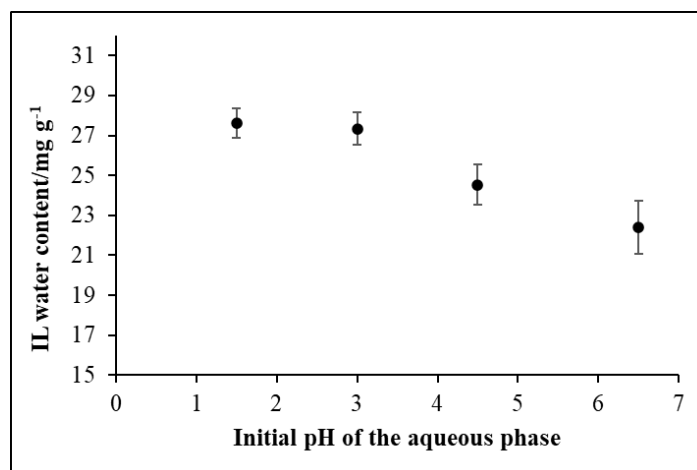


Figure 4.6: Effect of the initial pH of the aqueous phase on the amount of water dissolved in the IL-Ester at 25 °C (n = 3).

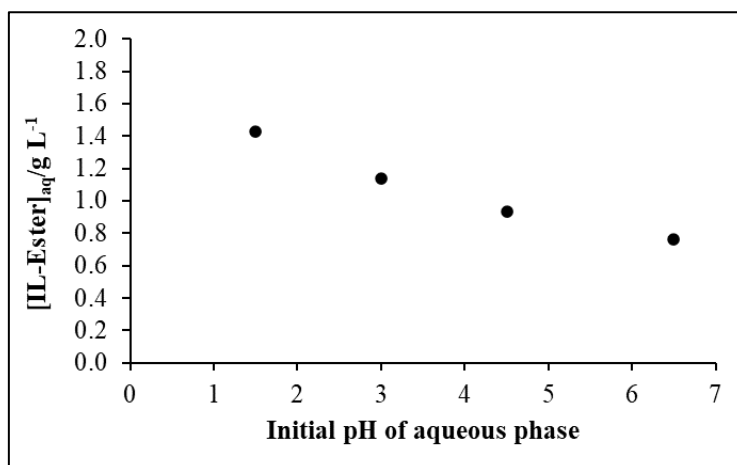


Figure 4.7: Effect of the initial pH of the aqueous phase on the solubility of IL-Ester in water at 25 °C.

Zhou and co-workers used COSMO-RS calculations to systematically predict the mutual solubilities of 1500 ILs with water [125]. It was argued that the molecular size of the IL is critical to its solubility in water. Increasing the molecular size of the IL tends to lower the charge density and polarity of the respective IL, resulting in a large non-polar surface. This in turn reduces the effective interactions with water molecules thereby increasing the misfit energy, resulting in reduced solubility. The alkyl substituent has a non-negligible effect on the mutual solubilities of IL-OH and IL-Ester with water and this is consistent with the theoretical predictions of Zhou et al. [125].

In the absence of auxiliary acid catalyst, a higher initial pH of the aqueous phase results in less water present in the IL phase. This is advantageous as it limits the water available for any potential hydrolysis of the IL-Ester. A higher initial pH also results in somewhat lower IL-OH concentration in the aqueous phase. This could be advantageous as it limits the loss of IL-OH after phase separation. However, for esterification, an auxiliary acid is often required as a catalyst. Therefore, the mutual solubility of IL-OH and water in the presence of increasing amounts of methanesulfonic acid was investigated.

#### 4.4.2 Effect of acid catalyst concentration on the aqueous solubility of IL-OH

Figure 4.8 shows the effect of the addition of the acid catalyst on the solubility of water in the IL phase: In the absence of methanesulfonic acid, the aqueous phase has a neutral pH and the solubility of water in the IL phase was approximately 50 mg g<sup>-1</sup>. Adding the smallest amount of methanesulfonic acid into the system resulted in the pH of the aqueous phase changing from a neutral pH to pH < 1, as expected when using a strong acid. Furthermore, the water uptake by IL-OH increased by approximately 10 mg g<sup>-1</sup> irrespective of the amount of methanesulfonic acid added (up to 12 wt.% tested). The mutual solubility of water and IL-OH in the presence of methanesulfonic acid is similar to when sulfuric acid is used, as previously shown in Section 4.4.1.

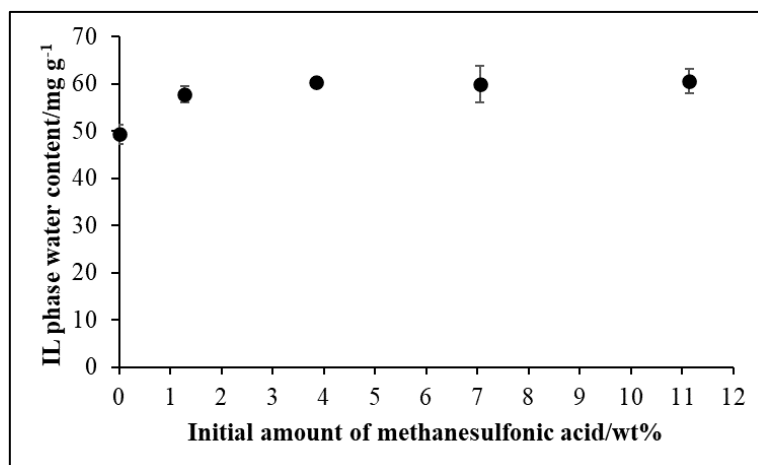


Figure 4.8: Solubility of water in IL-OH as a function of initial methanesulfonic acid concentration in the aqueous phase at 25 °C (n = 3).



In a study by Mazan et al. [127] the mutual solubility of water and a series of hydrophobic ILs, [C<sub>2</sub>MIM][NTf<sub>2</sub>], [C<sub>4</sub>MIM][NTf<sub>2</sub>], [C<sub>6</sub>MIM][NTf<sub>2</sub>], [C<sub>8</sub>MIM][NTf<sub>2</sub>] as a function of the initial concentration of hydrochloric acid, in the range 0 to 8 M was investigated.

Similarly, the solubility of IL-OH into the aqueous phase is independent of the initial amount of methanesulfonic acid in the aqueous phase. The effect of initial amount of methanesulfonic acid in the aqueous phase on the aqueous solubility of IL-OH is shown in Figure 4.9.

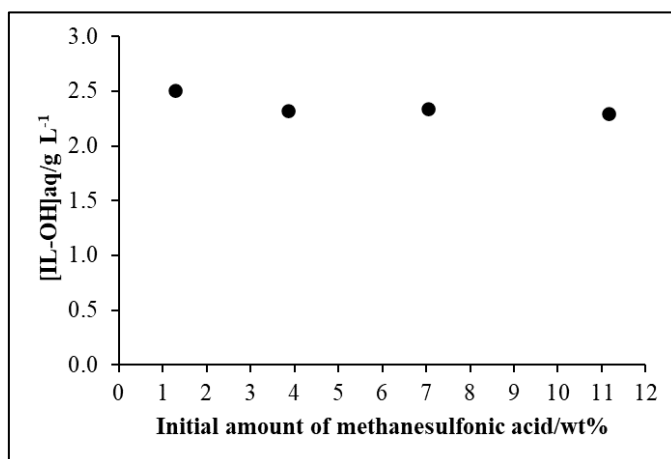


Figure 4.9: Effect of the initial amount of acid catalyst on the aqueous solubility of IL-OH at 25 °C.

The pH of the aqueous phase was monitored during the equilibration to establish whether methanesulfonic acid prefers to reside in the IL phase or the aqueous phase. In this case, a catalyst loading of 1.0 wt.% was added to separate vials containing equal volumes of IL-OH and water. In one vial, the acid catalyst was added to the aqueous phase while in the other vial it was added to the IL phase. In each case, the temperature was maintained at 25 °C. The pH of the aqueous phases was measured before and after 12 h of equilibration as shown in Appendix C, Table C5. The pH change for the [IL-OH + (water + 1.0 wt.% catalyst)] system shows that the H<sup>+</sup> concentration of the aqueous phase changed from approximately 0.049 M to 0.030 M. It can be assumed that the difference is due to the methanesulfonic acid being transferred into the IL-OH. The distribution of methanesulfonic acid into the IL phase is approximately 0.633. The optimal catalyst loading should be investigated in the future, for example via response surface optimization, where productivity and separation efficiency are the responses.

The aqueous solubility of IL-OH is an important factor when considering solvent losses and release into the environment. Furthermore, physicochemical properties of ILs are altered by the ILs' water content. It must be noted that for hydrophobic ILs of a given anion, the solubility in water is largely dependent on the structure of the cation. The hydrophobicity of the cation can be enhanced by increasing the length of the alkyl substituents. Alternatively, the salting-out principle can be employed to reduce the solubility of IL-OH in water [149-152]: The mutual solubility of water and IL-OH can be reduced by increasing the ionic strength of the aqueous phase. Therefore, salting-out-assisted reactive extraction can enhance the applicability of IL-OH in aqueous biphasic systems, improving the extractability of carboxylic acids. For example, Freire et al. [150] studied the effect of sodium salts on the solubility of [C<sub>4</sub>MIM][NTf<sub>2</sub>] and they observed that the aqueous solubility of the IL is governed by the specific ion effects and the salting-out of ILs is favoured by increasing the salt concentration. The choice and amount of the salting-out agent must be optimized with respect to IL-OH. The underlying specific ion effects behind the salting-out-assisted reactive extraction of carboxylic acid using IL-OH will need further investigation.

Alternatively, the sustainability and efficiency of IL-based reactive extraction can be enhanced by recycling the raffinate stream, using it as a makeup solvent for the fermentation. This can be achieved through process design, integration, and intensification.

#### **4.5 Octanol-water partitioning**

Octanol has a hydroxyl headgroup attached to a lipophilic alkyl chain. The polar-to-nonpolar balance within the octanol molecule allows it to be used as a surrogate for natural organic materials. Furthermore, the octanol molecule closely mimics the lipid molecules found in biological materials. As a result, the partition of chemical compounds between water and octanol ( $K_{ow}$ ) is used to estimate the environmental impact of the compounds [153]. The environmental impact is estimated based on correlations such as the soil sorption coefficient, bioconcentration factor (BCF) and bioaccumulation factors (BAF).

In this study the  $K_{ow}$  values IL-Ester and IL-OH were determined as a function of initial IL concentration in the aqueous phase. The investigated concentration ranges of IL-Ester

and IL-OH were guided by the LOD and LOQ of the HPLC analytical method, as well as the aqueous solubility of the ILs. The equilibrium concentrations of the ILs are presented in Figure 4.10. The amount of IL-OH transferred into the octanol phase is very low and increases slightly with increasing initial concentration of the IL in the aqueous phase. In the case of IL-Ester, there is a significant increase. IL-Ester has higher  $K_{ow}$  values compared to IL-OH and this can be attributed to differences in the structural composition. There is a greater tendency of hydrogen bonding interactions in IL-OH than in IL-Ester and, hence, the concentration of IL-OH will be greater in the aqueous phase than the octanol phase thus lowering the value of  $K_{ow}$  [154]. Additionally, there are also other interactions such as van der Waals forces that help to increase the affinity of IL-Ester for octanol.

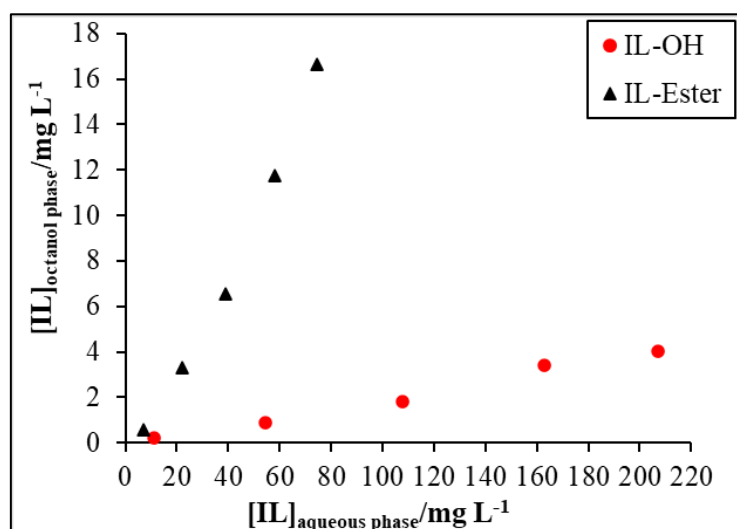


Figure 4.10: Equilibrium concentrations of IL-OH and IL-Ester in the octanol and aqueous phases at 25 °C.

Montalbán et al. [105] studied the effect of the anion and alkyl chain substituents on the  $K_{ow}$  values of various imidazolium-based ILs. It was observed that the lipophilicity of ILs can be lowered by inserting a hydroxyl moiety on the cation. For example, the  $K_{ow}$  of 1-ethyl-3-methylimidazolium chloride ([EMIM]Cl) and 1-hydroxy-ethyl-3-methylimidazolium chloride ([HEMIM]Cl) were found to be 0.0849 and 0.0111, respectively. In the same study, the  $K_{ow}$  of 1-hexyl-3-methylimidazolium bis(trifluoromethanesulfonyl)imide ([HMIM][NTf<sub>2</sub>]) was found to be 0.0352 while in this study the  $K_{ow}$  of IL-OH was found to be approximately 0.0194. This is keeping with the

trend observed by Montalbán et al. [105]. The effect of initial IL concentration on the  $K_{OW}$  value of IL-Ester and IL-OH is summarized in Tables 4.7 and 4.8, respectively.

Table 4.7: Effect of initial IL-Ester concentration on  $K_{OW}$ .

<b>[IL-Ester]<sub>initial, aq</sub></b> <b>/mg L<sup>-1</sup></b>	<b>[IL-Ester]<sub>eq, aq</sub></b> <b>/mg L<sup>-1</sup></b>	<b>[IL-Ester]<sub>eq, octanol</sub></b> <b>/mg L<sup>-1</sup></b>	<b>K<sub>OW</sub></b>	<b>Log K<sub>OW</sub></b>
7.64	7.09	0.55	0.08	-1.11
25.20	21.89	3.31	0.15	-0.82
45.49	38.92	6.57	0.17	-0.77
69.80	58.02	11.77	0.20	-0.69
91.06	74.41	16.65	0.22	-0.65

Table 4.8: Effect of initial IL-OH concentration on  $K_{OW}$ .

<b>[IL-OH]<sub>initial, aq</sub></b> <b>/mg L<sup>-1</sup></b>	<b>[IL-OH]<sub>eq, aq</sub></b> <b>/mg L<sup>-1</sup></b>	<b>[IL-OH]<sub>eq, octanol</sub></b> <b>/mg L<sup>-1</sup></b>	<b>K<sub>OW</sub></b>	<b>Log K<sub>OW</sub></b>
11.46	11.26	0.20	0.02	-1.74
55.33	54.42	0.91	0.02	-1.78
109.72	107.88	1.84	0.02	-1.77
16.21	162.82	3.39	0.02	-1.68
210.95	206.93	4.02	0.02	-1.71

Figure 4.11 shows the concentration dependence of  $K_{OW}$  for both IL-Ester and IL-OH. In the case of IL-OH, there is almost no significant change in the  $K_{OW}$  as the initial concentration of the IL in the aqueous phase was increased, resulting in a  $K_{OW}$  of 0.02.  $K_{OW}$  values for IL-Ester increased with increasing initial concentration of IL-Ester in the aqueous phase.

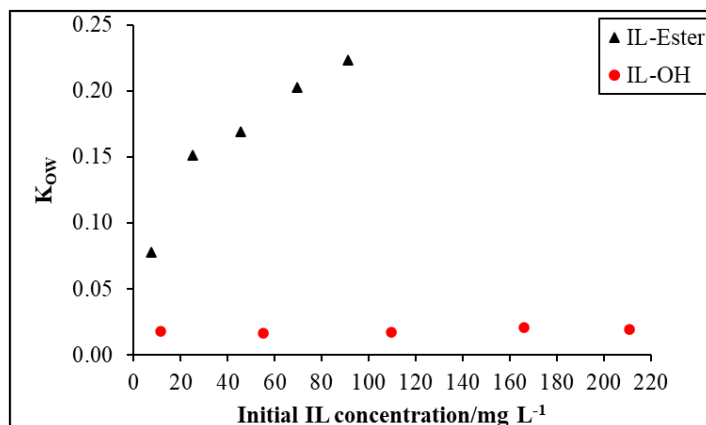


Figure 4.11: Effect of initial IL concentration on  $K_{ow}$  at 25 °C.

The  $K_{ow}$  of IL-Ester tends to be concentration-dependent. In a related study,  $K_{ow}$  of [BMIM][NTf<sub>2</sub>] was investigated as a function of IL concentration by Köddermann et al. [155]. The concentration-dependence of  $K_{ow}$  of [BMIM][NTf<sub>2</sub>] was attributed to the partial dissociation of ion pairs. The study emphasized that there is a higher extent of dissociation in the aqueous phase than the octanol phase.

Jain and Kumar [156], reported that  $\log K_{ow}$  values for [BMIM][HSO<sub>4</sub>] decreased from approximately -2.01 to -2.11 when the concentration was increased from 0.01 to 0.03 M. Similar observations were made for [HMIM][HSO<sub>4</sub>] and [OMIM][HSO<sub>4</sub>]. The shift from hydrophilic to hydrophobic character, with the size of the aggregates being heavily dependent on the IL's concentration is attributed to the tendency of ILs to self-aggregate in aqueous solutions [157, 158].

As discussed earlier in Section 2.6.3, the criteria for the identification and classification of toxic organic pollutants is well defined by the Stockholm Convention on persistent organic pollutants [106]. Based on the measured  $\log K_{ow}$  values of IL-Ester and IL-OH, the probability of IL-Ester and IL-OH to bioaccumulate is low, therefore, it can be concluded that the environmental impact of both ILs is low. IL-OH is a hydrolysis product of IL-Ester, therefore the environmental impact of all the ILs present during the application of IL-OH and IL-Ester is minimal.

Compared to the widely used molecular weight solvents, the  $K_{ow}$  values of IL-Ester and IL-OH are much closer to those of low molecular solvents such as seen in Table 2.7. In addition,  $K_{ow}$  values of ILs are significantly lower than those of high molecular weight

solvents such as tertiary amines and halogenated solvents. By interpretation of the  $K_{ow}$  values, and subsequent lipophilicity, it can be concluded that both IL-Ester and IL-OH have a low potential for bioaccumulation.

#### 4.6 Extraction of carboxylic acids from aqueous solution using IL-OH

The applicability of IL-OH as a reactive extractant for carboxylic acids was investigated using simulated aqueous solutions of levulinic acid or fumaric acid as model fermentation solutions. The selected carboxylic acids represent mono- and di-carboxylic acids as shown in Table 4.9.

Table 4.9: Solubilities of fumaric acid and levulinic acid in water at 25 °C

Carboxylic acid	Type	Molar mass/ g mol <sup>-1</sup>	Solubility/ g L <sup>-1</sup>	pK <sub>a</sub> , 25 °C	Ref
Fumaric acid	di-	116.07	7	3.03 (1), 4.54 (2)	[159]
Levulinic acid	mono-	116.11	675	4.65	[160]

The extraction of fumaric or levulinic acids was investigated separately, using initial acid concentrations in the ranges between 0 – 4.0 g L<sup>-1</sup> and 0.05 – 0.32 M, respectively. In the case of both fumaric and levulinic acid, the initial pH of the aqueous phase was in the range between 2 – 3. The addition of 1.0 wt.% methanesulfonic acid further reduced the initial pH of the aqueous phases such that pH < 2. The chemical extraction described herein refers to the extraction conducted in the presence of methanesulfonic acid as a catalyst for the esterification of the carboxylic acid with IL-OH.

The behaviour of the IL and aqueous phases during the extraction of the carboxylic acids was consistent with the observations made during the mutual solubility studies in Section 4.4. The solubility of water in the IL phase slightly increased with increasing initial concentration of the carboxylic acids. In addition, the presence of methanesulfonic acid in the system somewhat increased the solubility of water by a small fraction as shown in Figures 4.12 and 4.13.

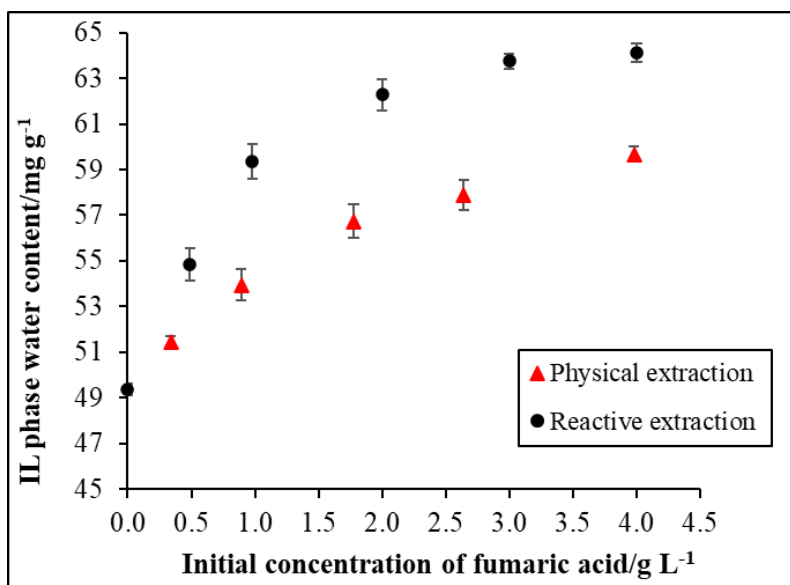


Figure 4.12: Solubility of water in the IL phase during the extraction of fumaric acid.

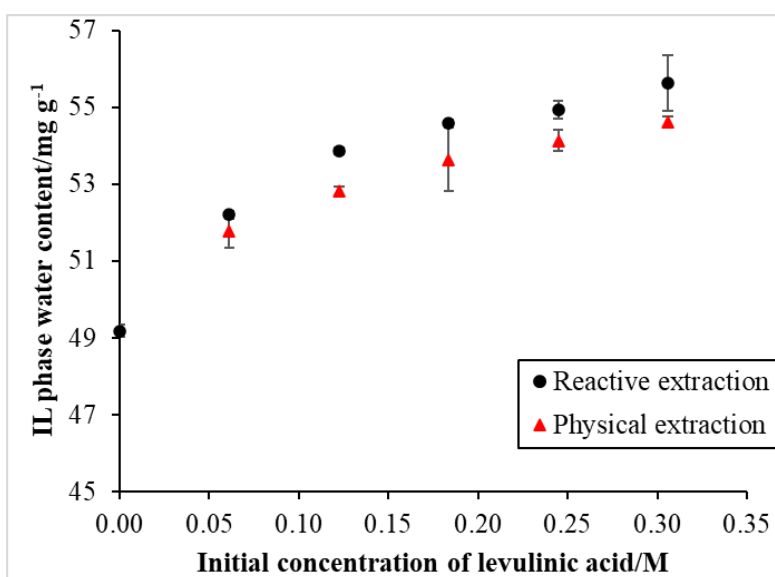


Figure 4.13: Solubility of water in the IL phase during the extraction of levulinic acid.

The amount of water transferred into the IL phase increases with increasing initial concentration of the carboxylic acid for both physical and chemical extraction. This can be explained in terms of the increase in the number of hydrogen bonding sites in the IL phase. Water molecules will be able to form hydrogen bonds with both the IL and the extracted carboxylic acid. Furthermore, since fumaric acid is a dicarboxylic acid, it has an extra hydrogen bonding site compared with levulinic acid which is a monocarboxylic

acid. The extra hydrogen bonding sites allow for further water molecules to dissolve into the IL phase and interact with the carboxylic acid molecules already in the IL phase.

During reactive extraction, the solubility of water in the organic phase is dependent on the nature of the extracted acid as well as its concentration. Tamada and King [19] observed that dicarboxylic acids carry more water with them into the organic phase during reactive extraction than monocarboxylic acids. It can be hypothesized that the extracted water interacts with the carboxylate group of the carboxylic acid via hydrogen bonding.

The solubility of IL-OH in the aqueous phase during the extraction of fumaric and levulinic acids was also consistent with the observations made in Section 4.4. There is an increased solubility of IL-OH in the aqueous phase during chemical extraction compared with physical extraction as shown Figures 4.14 and 4.15. The solubility of IL-OH in the aqueous phase during the physical extraction of either levulinic or fumaric acid is comparable. Similar observations were made for the solubility of IL-OH during chemical extraction.

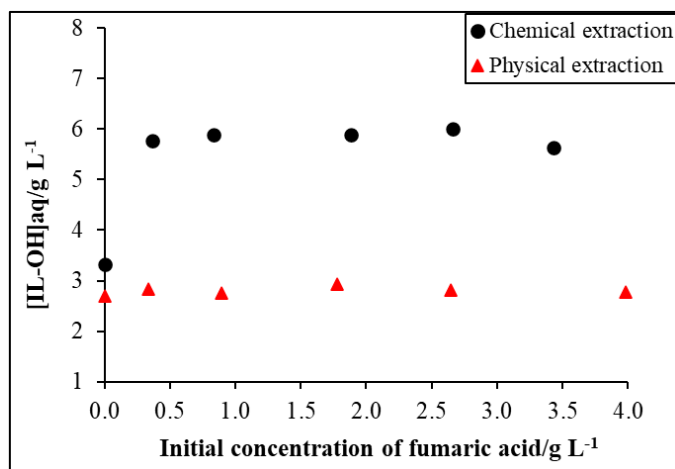


Figure 4.14: Effect of initial concentration of fumaric acid on the aqueous solubility of IL-OH during the extraction of fumaric acid with IL-OH at 25 °C.

The physical extraction was conducted in the absence of an auxiliary catalyst thereby reducing the possibility of the occurrence of the esterification reaction. On the other hand, a catalyst loading of 1.0 wt.% was added to the system during chemical extraction to facilitate the esterification reaction.



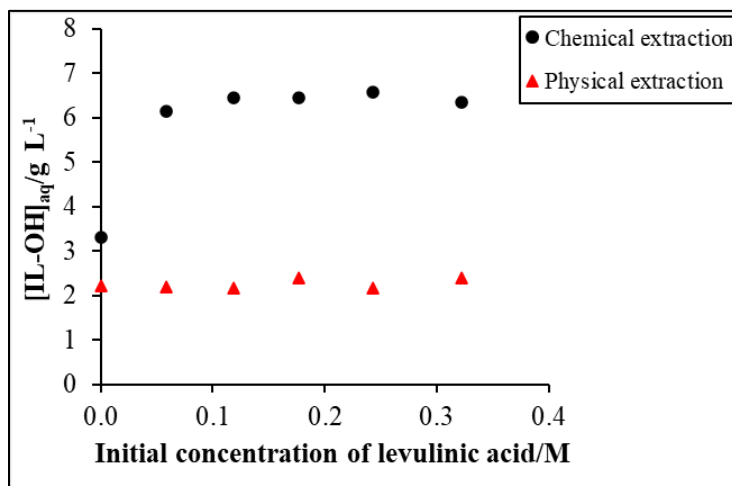


Figure 4.15: Effect of initial concentration of levulinic acid on the aqueous solubility of IL-OH during the extraction of levulinic acid with IL-OH at 25 °C.

IL-OH was able to physically extract both fumaric and levulinic acids from their respective aqueous solutions. The distribution of both carboxylic acids from the aqueous solution into the IL phase was dependent on the initial concentrations of the carboxylic acid in the aqueous phase. In the absence of a catalyst, the carboxylic acid would exhibit preferential solubility in the IL phase, exploiting various electrostatic forces and hydrogen bonding to make stable interactions with the IL. The HPLC analytical method described in Section 3.3.1 was used to analyse both the IL and aqueous phases, and the concentration of IL-Ester in the aqueous phase was below the detection limit ( $0.034 \mu\text{g cm}^{-3}$ ). This means there was no evidence of a chemical reaction between IL-OH and either of the carboxylic acids during physical extraction. Figure 4.16 shows the chromatogram from the analysis of the aqueous phase sampled after the physical extraction of levulinic acid. The method could only detect levulinic acid and IL-OH. The presence of IL-Ester would have been expected at a retention time of 2.266 minutes where the chromatogram is clear.

The expected reaction is equilibrium-driven, and it follows the Fischer esterification mechanism, producing water as one of the products. During physical extraction, the aqueous phase provides a large excess of water which hinders the esterification reaction. In addition, the extraction was conducted at 25 °C, which does not favour the forward reaction of esterification reaction, a strongly exothermic reaction.

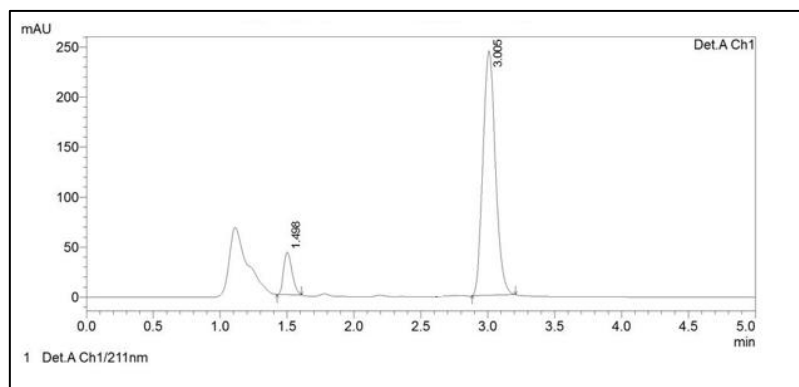


Figure 4.16: Sample HPLC chromatogram showing the elution of levulinic acid and IL-OH at 1.498 and 3.005 minutes, respectively. The analysed sample was taken from the aqueous phase after the physical extraction of levulinic acid using IL-OH at 25 °C.

The operating conditions used during the physical extraction were modified by adding methanesulfonic acid, which acted as an acid catalyst. The presence of an acid catalyst enhanced the probability of the occurrence of the esterification reaction.

The conversion of IL-OH to IL-Ester in aqueous solution is very low due to equilibrium limitations. Increasing the reaction temperature can be used to augment the conversion. Furthermore, increasing the initial concentration of carboxylic acid tends to increase the amount of acid available to chemically react with the IL-OH, thereby driving the reaction forward.

The effect of the initial water content of the reaction mixture on the esterification reaction was investigated. Increasing the amount of water during an esterification reaction does not only affect the equilibrium based on Le Chatelier's principle. The concentration of the protonated acid in the reaction matrix is reduced by the presence of excess water, hindering the reaction. Altıokka and Çıtak [161] and Goto et al. [162] reported similar retardation effects due to the presence of water in an esterification reaction.

Figure 4.17 shows the effect of initial water content during the esterification of IL-OH with levulinic acid at 70 °C. The conversion of IL-OH decreased with increasing the initial amount of water present in the reaction mixture. The reaction mixture was monophasic in the presence of low amounts of water that are within the mutual solubility

limits of IL-OH, and a biphasic mixture is formed when the initial amount of water in IL-OH exceeds 10 wt.% of the IL. On a molecular level, this translates to a molar percentage of 74% water in the mixture. It is remarkable that the conversion of IL-OH with an equimolar amount of levulinic acid still exceeds 80%.

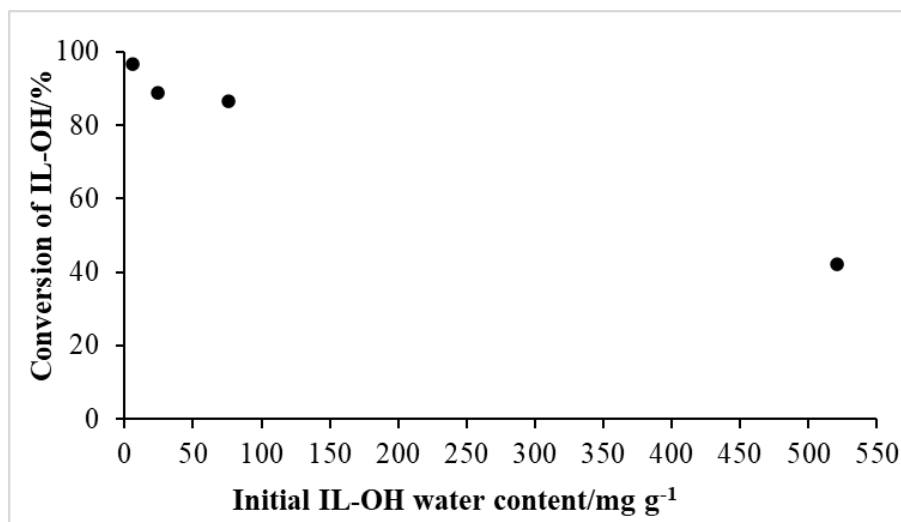


Figure 4.17: Effect of water loading on the esterification of IL-OH with levulinic acid in the presence of methanesulfonic acid as a catalyst (1 wt.% with respect to IL-OH) at 70 °C using a 1:1 molar ratio of IL-OH to levulinic acid.

Regardless of the amount of water in the system, the conversion of IL-OH to IL-Ester was achieved during the acid-catalysed esterification of reaction in the presence of water. Adopting continuous reactor set-ups will be more ideal for the reactive extraction of carboxylic acids from aqueous solutions. The conversion of IL-OH is still expected to be consistent with other process variables such as reaction temperature.

The amount of IL-OH used for the extraction of fumaric and levulinic acid was kept constant in all batch experiments. Hence, the molar ratio of fumaric and levulinic acid with respect to IL-OH increased with increasing initial concentration of fumaric and levulinic acid in the aqueous phase as shown in Figures 4.18 and 4.19.

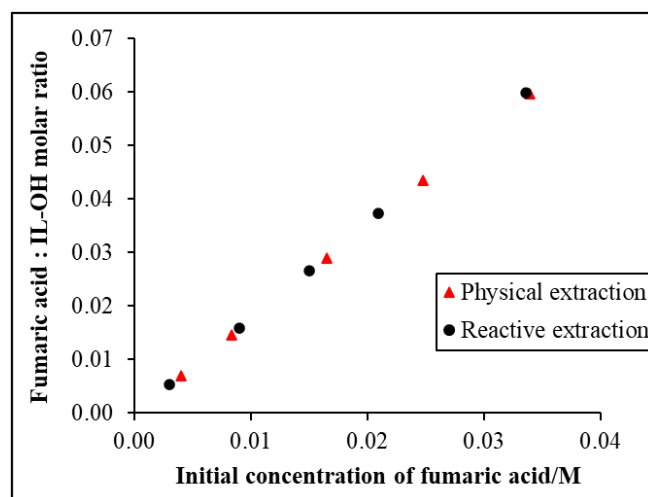


Figure 4.18: Effect of increasing the initial concentration of fumaric acid on the initial molar ratio of fumaric acid with respect to IL-OH at 25 °C. The reactive extraction experiments were conducted in the presence of 1.0 wt.% methanesulfonic acid as a catalyst.

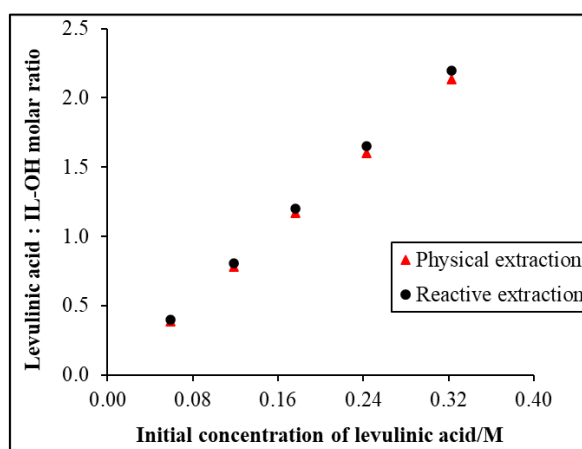


Figure 4.19: Effect of increasing the initial concentration of levulinic acid on the molar ratio of levulinic acid with respect to IL-OH at 25 °C. The reactive extraction experiments were conducted in the presence of 1.0 wt.% methanesulfonic acid as a catalyst.

Increasing the initial concentration of the carboxylic acid in the feed increases its extractability by IL-OH as seen in Figures 4.20 and 4.21. Secondly, the presence of methanesulfonic acid during reactive extraction enhanced the extractability of both

fumaric and levulinic acid by IL-OH. The increase in extractability can be attributed to the reaction enhanced extraction of fumaric acid by IL-OH where an esterification reaction occurs between fumaric acid and IL-OH.

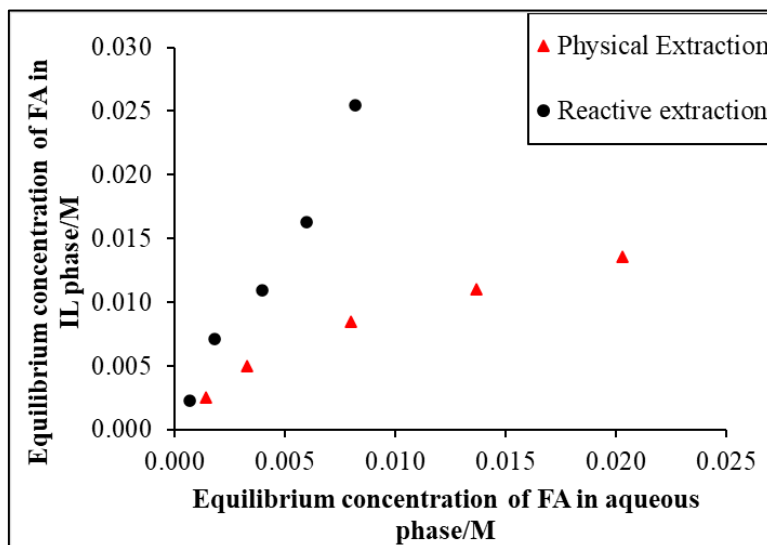


Figure 4.20: Extraction equilibria of fumaric acid with IL-OH at 25 °C. Experiments were conducted using a 1:1 v/v phase ratio. The reactive extraction was conducted in the presence of 1.0 wt.% methanesulfonic acid as a catalyst.

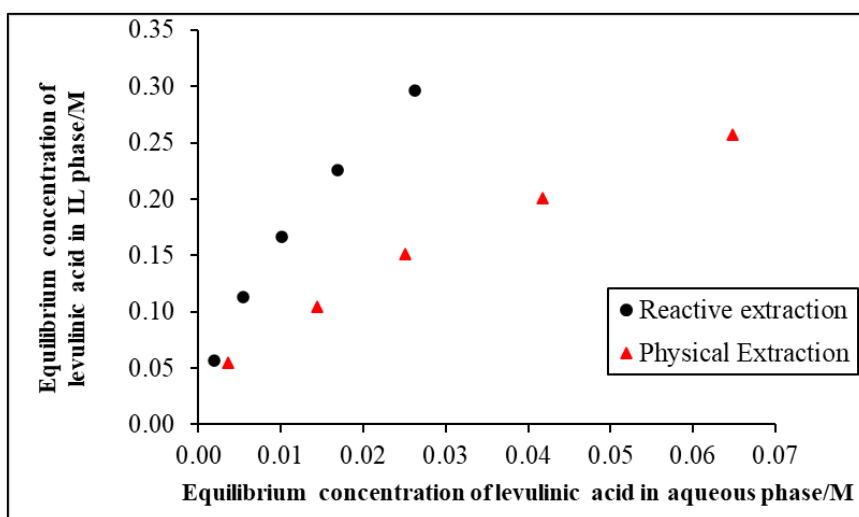


Figure 4.21: Extraction equilibria of levulinic acid with IL-OH at 25 °C. Experiments were conducted using a 1:1 v/v phase ratio. The reactive extraction was conducted in the presence of 1.0 wt.% methanesulfonic acid as a catalyst.

Detailed data on the extraction is presented in Appendix D.

## 4.7 Transesterification of IL-Ester with butanol, and regeneration of IL-OH

The transesterification of IL-Ester with butanol provides a pathway through which the extracted carboxylic acid can be stripped from the IL-OH and is recovered as a value-added chemical. Only the IL-Ester derived from levulinic acid was subjected to the acid-catalysed transesterification reaction.

### 4.7.1 Factors affecting the transesterification of IL-Ester with butanol

The effect of reaction temperature, reactant molar ratio, and catalyst loading on the transesterification of IL-Ester with butanol was investigated via batch experiments.

#### 4.7.1.1 Effect of reactant molar ratio on the transesterification reaction

The transesterification reaction of IL-Ester with butanol is a reversible reaction. Therefore, a stoichiometric excess of butanol was used to create a forward bias [163]. As shown in Figure 4.22, increasing the molar ratio from 1:3 to 1:10 significantly increased the reaction rate, as well as the maximum conversion after 420 minutes from 72.4 to 94.2%. This shows that a higher ratio of alcohol is needed to ensure a rapid rate of reaction resulting in a significantly higher yield than at lower ratios.

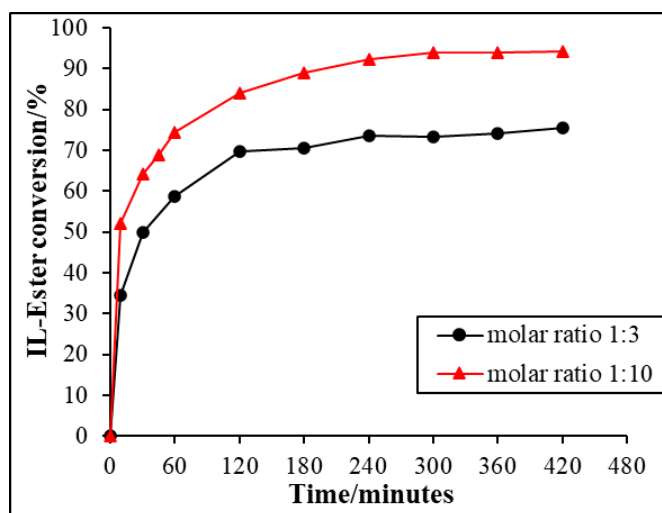


Figure 4.22: Effect of reactant IL-Ester: butanol molar ratio on the transesterification reaction at 85 °C and 1.0 wt.% methanesulfonic acid as a catalyst.

#### 4.7.1.2 Effect of temperature on transesterification reaction

The effect of temperature on the transesterification of IL-Ester with n-butanol was investigated at 85 and 115 °C. Increasing the reaction temperature increases the rate of conversion of the reaction as seen in Figure 4.23. This is because by increasing the temperature, the kinetic energy of the reactant particles is increased. The concomitant increase in reaction rate allows the system to reach equilibrium faster. In the presence of 1.0 wt.% catalyst, the conversion of IL-Ester after 300 minutes increased from 93.86 to 96.4% due to the increase in reaction temperature from 85 to 115 °C.

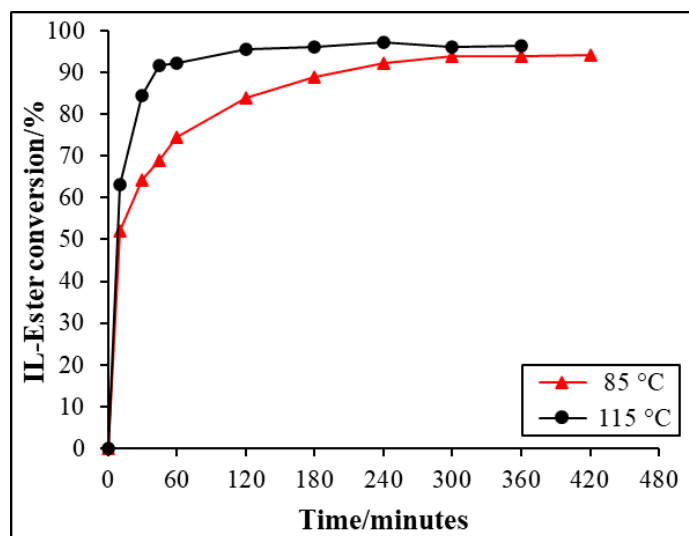


Figure 4.23: Effect of temperature on the transesterification of IL-Ester with butanol in the presence of 1.0 wt.% methanesulfonic acid catalyst and an IL: alcohol ratio of 1:10.

#### 4.7.1.3 Effect of catalyst loading on the transesterification reaction

The effect of the amount of catalyst on the transesterification of IL-Ester with butanol was investigated within the range 0 – 5.0 wt.%. In the absence of a catalyst, the reactants were immiscible, and they formed a biphasic mixture and as a result this catalyst level was not pursued. As shown in Figure 4.24 increasing catalyst loading from 1.0 to 5.0 wt.% resulted in a significant increase in the rate of conversion of IL-Ester into butyl levulinate and IL-OH. After 300 minutes, the conversion of IL-Ester increased from 92.3 to 96.61% due to the increase in catalyst loading from 1 to 5.0 wt.% at 85 °C.

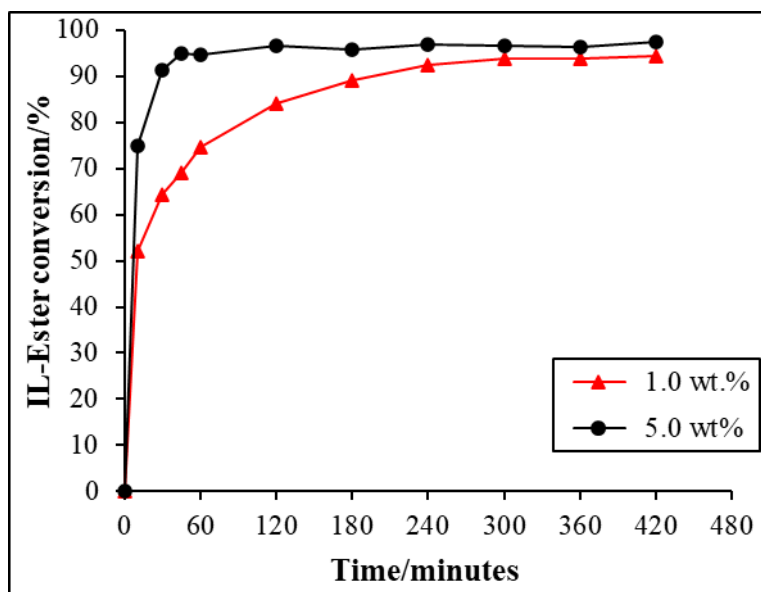


Figure 4.24: Effect of methanesulfonic acid catalyst loading on the transesterification of IL-Ester with butanol using an IL: alcohol ratio of 1:10 at 85 °C.

It is important to note that neither the reaction temperature nor the amount of catalyst influences the overall degree of conversion but simply the speed of conversion. However, increasing the ratio of butanol increases the percentage conversion during the batch esterification of IL-Ester with butanol in the presence of an acid catalyst.

In principle, the transesterification reaction will be employed to strip the extracted carboxylic acids from the IL phase and regenerate the IL. The regenerated IL will be recycled and applied again as a reactive extractant for carboxylic acids.

#### 4.7.2 Product separation and regeneration of IL-OH

After transesterification, the reaction mixture comprises of regenerated IL-OH, remaining IL-Ester, butyl levulinate, butanol, as well as methanesulfonic acid. To regenerate IL-OH, butanol was first removed by distillation, followed by the extraction of butyl levulinate. The remaining mixture of IL-OH, IL-Ester and methanesulfonic acid would then be returned to the extraction of the carboxylic acid. In the following, these steps are investigated in detail.

##### 4.7.2.1 Removal of butanol by distillation

The unreacted butanol was recovered from the reaction mixture by vacuum distillation at 40 °C. When the recovered butanol was analysed with the GC-FID method described in



Section 3.3.2, no trace of other organic compounds could be detected. The average retention time for butyl levulinate is approximately 10.312 minutes. This is evidence that the recovered butanol is not contaminated with the reaction products. Figure 4.25 shows a GC-FID chromatogram of the recovered butanol, where butanol and the internal standard eluted at 4.026 and 8.587 minutes, respectively.

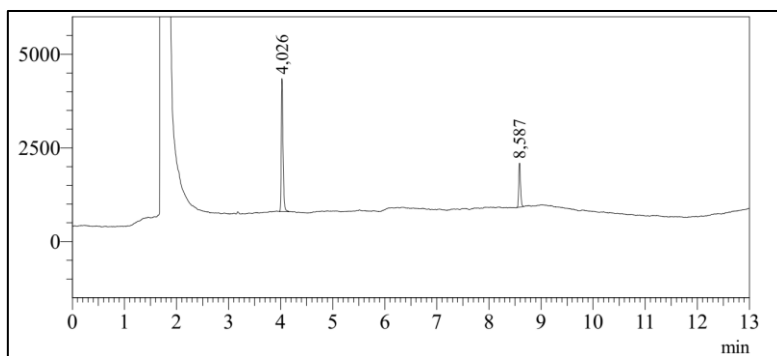


Figure 4.25: GC-FID determination of butanol on a Zebron ZB-WAXPLUS capillary column, in the presence of ethyl levulinate as an internal standard and helium as the carrier gas at a flow rate of  $1.27 \text{ cm}^3 \text{ min}^{-1}$ .

The recovered butanol was quantified gravimetrically, and it correlated to the reaction yield of the transesterification. The recovered butanol can be re-used, thereby reducing the overall input costs. This will result in the reduction of pollution due to the release of butanol into the environment. Table 4.10 shows that the recovery of unreacted butanol from the transesterification reaction is independent of the reactant molar ratio. The observed differences can be attributed to process and handling losses.

Table 4.10: Recovery of butanol as a function of reactants' molar ratio.

<b>IL-Ester: Butanol</b>	<b>Recovery yield/%</b>
1:3	94.11
1:10	93.83

An excess of butanol in the product mixture increases the alcohol to product molar ratio. Such operating conditions complicate downstream separation and purification protocols, imposing additional costs [164]. For example, a higher ratio means that more energy is required to recover the excess butanol from the reaction mixture. On the other hand, a

higher butanol to IL-Ester ratio is important for the maximum conversion during the transesterification reaction. A compromise can be made to use the lowest excess molar ratio of butanol in the reactants. The influence of process conditions on the total annual cost and yield will need to be optimized via a multi-response optimization to establish a trade-off between cost of downstream separation and excess butanol needed.

#### 4.7.2.2 Recovery of butyl levulinate

The acid-catalysed transesterification of IL-Ester with butanol produces butyl levulinate (IUPAC name butyl 4-oxopentanoic acid) as the targeted product. Butyl levulinate is isolated from the reaction mixture by means of solvent extraction with diethyl ether. A yield of 94.62% was achieved. The isolated butyl levulinate was characterized using  $^1\text{H}$  NMR,  $^{13}\text{C}$  NMR, FTIR and GC-FID methods to establish its composition and purity. The structure of the isolated product was confirmed using  $^1\text{H}$  NMR. The proton environments appearing in the spectrum shown in Figure 4.26 are consistent with the chemical structure of butyl levulinate. The absence of typical chemical shifts for the imidazolium ring in the range 7 – 9 ppm suggest that the isolated product, butyl levulinate is free from contamination by either IL-OH or IL-Ester.

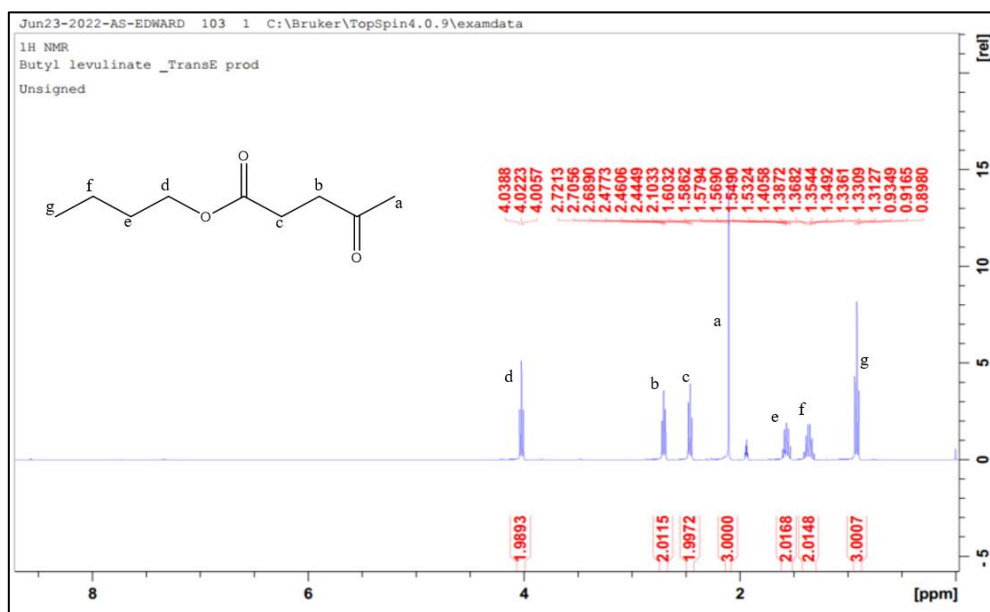


Figure 4.26:  $^1\text{H}$  NMR spectrum for butyl levulinate recovered by solvent extraction in diethyl ether. 10 mg of sample was dissolved in  $0.7\text{ cm}^3$  of acetonitrile- $\text{d}_3$ .

The chemical structure of butyl levulinate has only three functional groups, namely C=O, C-O, and C-H. The FTIR spectrum shown in Figure 4.27 is consistent with the functional groups present in butyl levulinate. The C=O and C-H bands can be observed at approximately 1720 and 2960  $\text{cm}^{-1}$ , respectively.

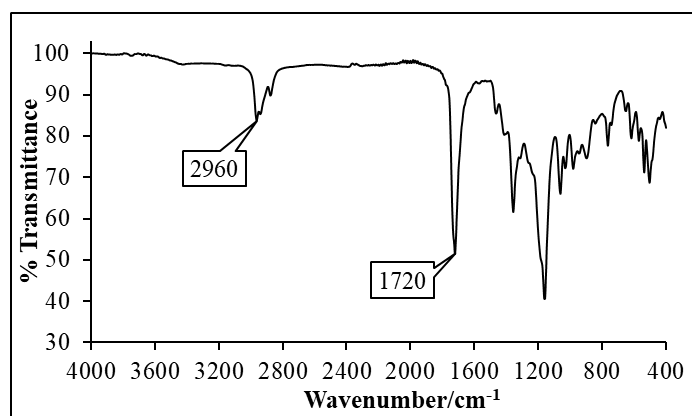


Figure 4.27: FTIR spectrum for butyl levulinate.

#### 4.7.2.3 Regenerated IL-OH

Irrespective of temperature and catalyst loading used in the transesterification, approximately 95% of the IL-OH was regenerated from IL-Ester, as determined by HPLC (see Figure 4.28).

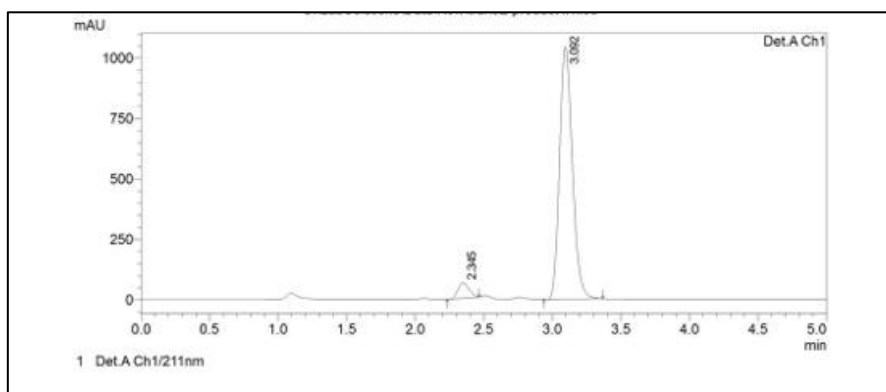


Figure 4.28: Sample HPLC chromatogram for the regenerated IL showing IL-OH with traces of IL-Ester eluting at 3.002 and 2.345 minutes, respectively.

$^1\text{H}$  NMR spectra for the regenerated IL were compared against those of pure IL-Ester and IL-OH as shown in Figure 4.29. The chemical shifts for the protons in the regenerated IL were consistent with those of a mixture of IL-Ester and IL-OH. This confirmed that

the behaviour of the cation during the transesterification reaction was consistent with the expected reaction mechanism. The composition of the regenerated IL was also determined from the  $^1\text{H}$  NMR spectrum using Equation 3.5. The full  $^1\text{H}$  NMR spectrum is presented in Appendix F. The mole fractions of IL-Ester and IL-OH in the regenerated IL were estimated to be 0.059 and 0.941, respectively, confirming the HPLC result.

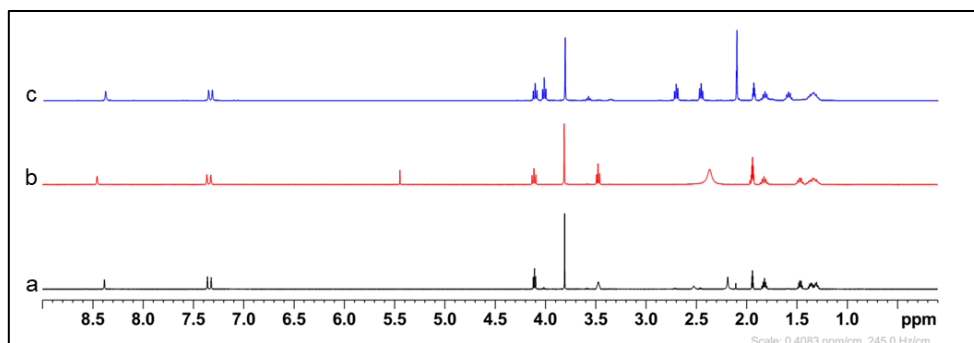


Figure 4.29: Comparison of  $^1\text{H}$  NMR spectra for (a) regenerated IL, (b) IL-OH, and (c) IL-Ester, recorded in acetonitrile- $\text{d}_3$ .

$^{19}\text{F}$  NMR spectrometry was used to monitor the  $[\text{NTf}_2]^-$  anion. The respective ILs were analysed after the ion exchange, the esterification, and the transesterification reactions. Figure 4.30 shows the  $^{19}\text{F}$  NMR spectra of the regenerated IL mixture, as well as pure IL-OH and IL-Ester, with the chemical shifts of the fluorine nuclei occurring at approximately -80 ppm for all three ILs.

The spectra for all the ILs show a single environment for the fluorine nuclei, which is consistent with the  $[\text{NTf}_2]^-$  anion. Both the esterification and transesterification reactions did not result in the formation of new environments with respect to the fluorine nuclei. Therefore, it can be concluded that the integrity of the anion is maintained during the application of IL-OH as a reactive extractant for carboxylic acids.

The  $[\text{NTf}_2]^-$  anion in the regenerated IL is common to both IL-OH and IL-Ester. Therefore, the number of moles of the  $[\text{NTf}_2]^-$  anion present in the regenerated IL would be the sum of the anion in IL-Ester and IL-OH collectively.

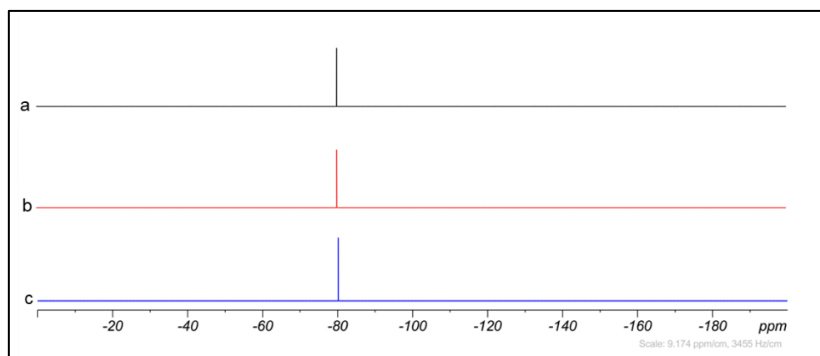


Figure 4.30:  $^{19}\text{F}$  NMR spectra of (a) regenerated IL mixture, (b) IL-OH and (c) IL-Ester recorded in acetonitrile- $\text{d}_3$ .

Figure 4.31 shows the  $^{19}\text{F}$  NMR spectrum of the regenerated IL in the presence of  $\text{CF}_3\text{CO}_2\text{Na}$  as an internal standard. The NMR sample was prepared by dissolving 12.8 mg of the regenerated IL and 13.1 mg of  $\text{CF}_3\text{COONa}$  in  $0.8\text{ cm}^3$  of acetonitrile- $\text{d}_3$ . Chemical shifts due to the fluorine nuclei were observed at  $-76.1467$  and  $-80.1995$  ppm for  $\text{CF}_3\text{CO}_2\text{Na}$  and the regenerated IL, respectively. The number of moles of the  $[\text{NTf}_2]^-$  anion in the sample was determined from the  $^{19}\text{F}$  NMR spectrum by using Equation 3.5. The total number of moles of the  $[\text{NTf}_2]^-$  anion was found to be  $1.946 \times 10^{-5}$  in this sample.

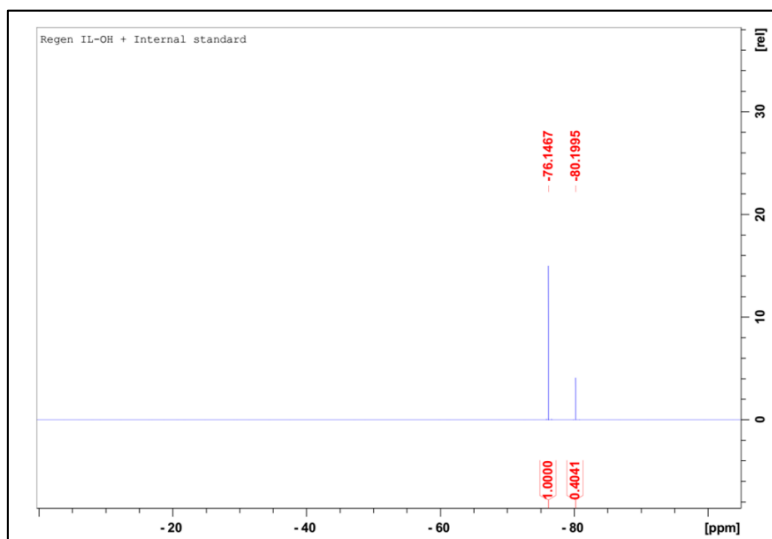


Figure 4.31:  $^{19}\text{F}$  NMR spectrum of regenerated IL in the presence of  $\text{CF}_3\text{CO}_2\text{Na}$  as an internal standard. The spectrum was recorded in acetonitrile- $\text{d}_3$ .

The cation composition of the regenerated IL mixture was also established by means of HPLC. In this case, 9.1 mg of sample was dissolved in 10 cm<sup>3</sup> of the HPLC mobile phase and analysed using the analytical protocol described in Section 3.3.1. The composition of the regenerated IL-OH as well as the resulting number of mols of [NTf<sub>2</sub>]<sup>-</sup> is presented in Table 4.11, assuming that the cations are paired with [NTf<sub>2</sub>]<sup>-</sup> anions. By comparison with the value found by <sup>19</sup>F NMR, it is concluded that all IL cations are paired with the [NTf<sub>2</sub>]<sup>-</sup> anion, and no ion exchange has occurred during the reactive extraction and transesterification.

Table 4.11: Composition of the regenerated IL determined by HPLC.

IL	% Content in the sample	Mass/g	Moles	Total number of moles of ILs in sample
IL-Ester	5.02	$4.57 \times 10^{-4}$	$8.142 \times 10^{-7}$	$1.946 \times 10^{-5}$
IL-OH	94.98	$8.64 \times 10^{-3}$	$1.865 \times 10^{-5}$	

The FTIR spectrum of the regenerated IL was compared with those of pure IL-OH and IL-Ester. The functional groups in the regenerated IL are consistent with those present in both IL-OH and IL-Ester as shown in Figure 4.32.

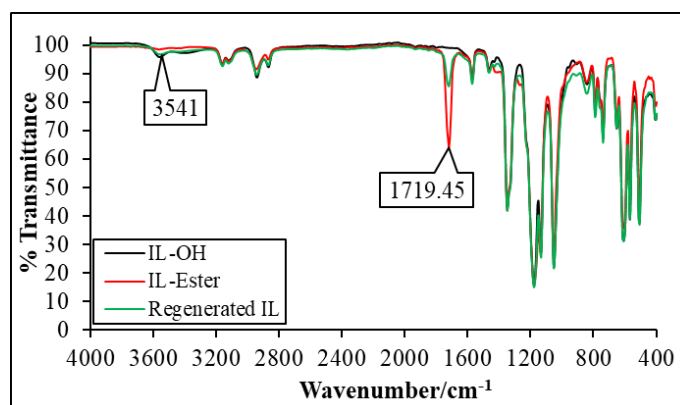


Figure 4.32: Comparison of FTIR spectra for pure IL-OH, IL-Ester, and regenerated IL.

The carbonyl band observed at 1719.45 cm<sup>-1</sup> is expected for IL-Ester. However, the same band exists in the spectrum for regenerated IL, qualitatively confirming the presence of traces of IL-Ester in the regenerated IL, which was not converted to IL-OH during transesterification.

## 4.8 Comparison with common extractants

The application of IL-OH as a reactive extractant for carboxylic acids is a promising innovation. The formation of IL-Ester during the reactive extraction is evidence of an actual chemical reaction leading to formation of covalent bonds between the solute and the extractant. The physicochemical properties of IL-Ester as well as IL-OH are well understood. This is important in evaluating the adherence of this extraction system to the solvent selection criteria as described in Table 2.4.

A comparison of the physicochemical properties of IL-OH and IL Ester with commonly used reactive extractants for carboxylic acids is presented in Table 4.12. The proposed extractant system satisfies the solvent selection criteria.

The reported viscosities of CYPHOS IL-104, [HMIM][PF<sub>6</sub>] and [HMIM][NTf<sub>2</sub>] at 25 °C are approximately 2011.7, 1572, and 269.4 mPa s, respectively [165]. These ILs have previously been investigated as reactive extractants for carboxylic acids from aqueous solution [36]. The viscosity of IL-OH at 25 °C is approximately 135 mPa s, which is significantly lower than the other ILs. An inverse relationship exist between the diffusion coefficient and the viscosity of the system [166].

The behaviour of IL-OH and IL-Ester in the presence of aqueous media has been investigated, allowing reactor designs to recover the IL from the effluent streams prior to discharge into the environment. Although the solubilities of IL-OH and IL-Ester are relatively low, some thought will have to be given to the treatment of the aqueous effluent, which is to be done in future studies. The reactive extraction is conducted at ambient temperature while the solvent regeneration step is done at an elevated temperature. The operating conditions for both the extraction and the regeneration step are within the thermal stability range for both IL-Ester and IL-OH.

A comparison of the physicochemical properties of IL-Ester, IL-OH, TOA and Cyphos IL-104 is presented in Table 4.12. TOA and Cyphos IL-104 are examples of a molecular organic solvent and an IL used for the extraction of carboxylic acids from aqueous solution, respectively.

The thermal stabilities of IL-OH and IL-Ester show that the IL can be adapted to a wide temperature range for both the esterification and transesterification reactions.

Furthermore, the log Kow value TOA translate to a high risk of bioaccumulation compared with both IL-OH and IL-Ester.

Table 4.12: Physicochemical properties of IL-OH, IL-Ester, TOA and CYPHOS IL-104.

	<b>TOA<sup>a</sup></b>	<b>CYPHOS IL-104<sup>b</sup></b>	<b>IL-OH<sup>c</sup></b>	<b>IL-Ester<sup>c</sup></b>
Molar mass/g mol <sup>-1</sup>	353.67	773.27	463.42	561.52
Density <sub>25 °C</sub> /g cm <sup>-3</sup>	0.809	0.895	1.40	1.36
Viscosity <sub>25 °C</sub> /mPa s	8.325	2011.7	135	151.56
Solubility in water <sub>25 °C</sub> /g L <sup>-1</sup>	0.000039	0.01	2.7	0.8
Water saturation <sub>25 °C</sub> /wt. %	Immiscible	15.3	5.15	2.55
Log Kow	10.5	Unknown	-1.72	-0.69
Thermal stability/°C	Not available	≤ 250	≤ 297	≤ 258
Chemical reactivity	Not available	Not available	Yes	Yes

The superscripts <sup>a</sup>, <sup>b</sup> and <sup>c</sup> refer to work adapted from [116], [167] and [168], respectively.

The biphasic systems based on either TOA and Cyphos IL-104 are such that the aqueous phase is the dense layer and hence it settles as the bottom phase. However, in aqueous extractions based on IL-OH and IL-Ester, the aqueous phase settles as the lighter phase on top of the dense IL phase.

#### **4.9 Process configuration and preliminary mass balances for reactive extraction of carboxylic acids**

The design of commercial industrial processes follows a three-step strategy. The key step focuses on proof of concept and feasibility studies, defining process objectives and variables [169]. It is important to establish an overall statement regarding the conservation of materials involved in the process during this stage. Data derived from feasibility studies is used for the optimization of design configurations during the later stages of process design. Simulation models are developed at this stage. Inputs such as reactor sizing, pumping requirements, as well as heat and energy needs, are estimated based on the previous step.



Figure 4.33 shows one of several possible process configurations for the application of IL-OH in the reactive extraction of carboxylic acids from aqueous solution. Ideally, the fermentation broth would be derived from a bioreactor. Prior to the extraction step, the broth will be filtered-off to recover the microorganisms for reuse in the fermentation process. The carboxylic acids will be extracted from the filtered broth using IL-OH. Regardless of mutual solubility, the density difference between the IL phase and water readily allows for phase separation to take place, thus the raffinate phase can be easily isolated and recycled. The IL phase is subjected to a transesterification reaction in the presence of an excess amount of alcohol, where the extracted carboxylic acid is converted to an alkyl-ester. This reaction simultaneously regenerates IL-OH. The composition of the IL phase is determined by the extent of conversion during the reaction. The unreacted alcohol is recovered through vacuum distillation and reused in the transesterification stage. A solvent extraction protocol will then be employed to isolate and purify the alkyl-ester. A solvent extraction protocol will then be employed to isolate and purify the alkyl-ester.

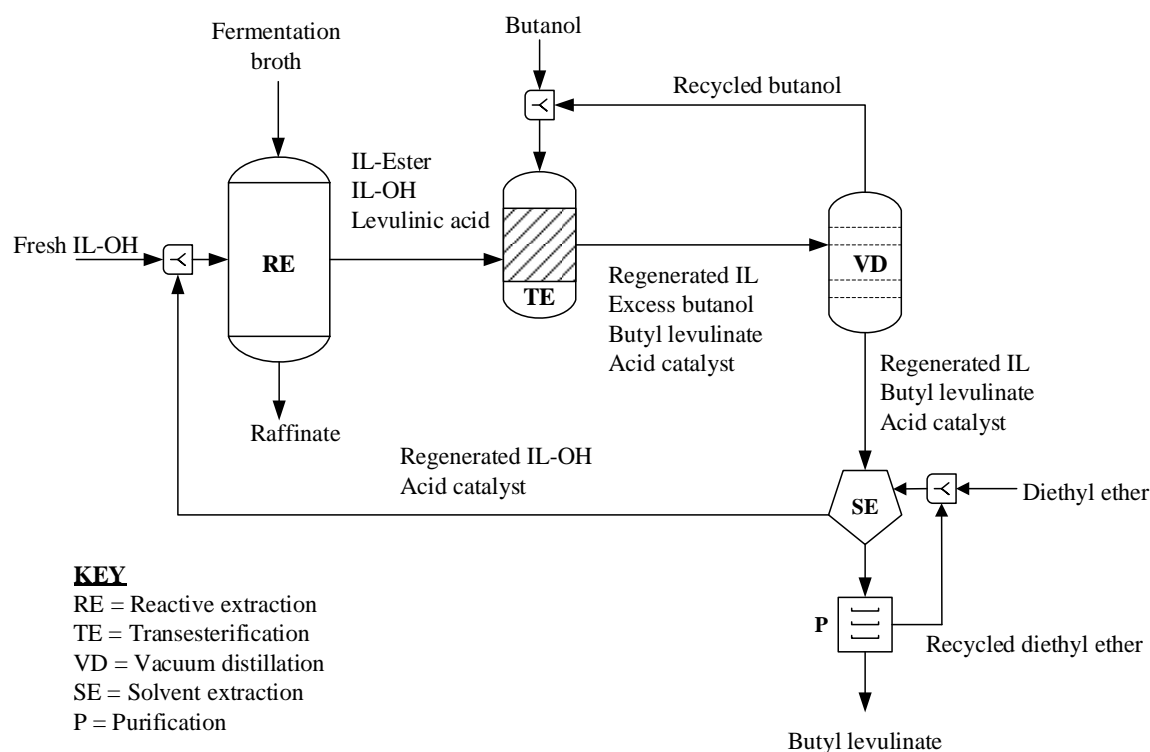


Figure 4.33: Idealized set-up for the reactive extraction of carboxylic acids using IL-OH.

A techno-economic assessment (TEA) of the whole process can then be conducted as the final step in process design. TEA is based on the optimum operating conditions and, therefore, validates the process [169, 170]. The validation process forms a basis for informed decision-making regarding the adoption and commercialization of the proposed technological designs. TEA is very useful in the identification of investment opportunities.

A case scenario was conducted with a solution of 10 wt.% levulinic acid. A mass balance calculation was established based on the experimental data from this study. The following assumptions were used as a basis in the respective calculations:

- i. The solubility of IL-OH in water at 25 °C is 2.5 g L<sup>-1</sup>.
- ii. The solubility of water in IL-OH at 25 °C is 56 mg g<sup>-1</sup> IL.
- iii. The distribution coefficient of levulinic acid for the reactive extraction of levulinic acid with IL-OH is 9.96 when the initial concentration of levulinic acid is 10 wt.%.
- iv. 6.05% of the extracted levulinic acid is bound in IL-Ester.
- v. At 85 °C, the transesterification of IL-Ester with butanol, and esterification of levulinic acid, in the presence of a 1:10 molar excess of butanol achieves approximately 95% conversion.
- vi. Methanesulfonic acid dissolved in the IL phase during the reactive extraction step remains quantitatively within the system and is also utilised as a catalyst during the transesterification reaction. However, the methanesulfonic acid transferred into the raffinate can be compensated by careful top-ups.
- vii. The vacuum distillation step achieves complete removal of unreacted butanol from the transesterification mixture.

The mass balance calculation is based on changes in the composition of interconnecting streams between the extraction and the back-extraction stages. Tables 4.13 – 4.16 present the tabulated mass balance data with respect to individual stages during the process. The extraction of levulinic acid is achieved by mixing IL-OH with the aqueous solution, at which point phase miscibility, physical extraction, and chemical extraction are inevitable. Chemical extraction is driven by an acid-catalysed esterification reaction between IL-OH

and levulinic acid. The esterification reaction introduces new chemical species, i.e., IL-Ester into the system.

Table 4.13: Mass balance data for the reactive extraction of levulinic acid stage.

Component	Mass/g		
	IN	OUT aq phase	OUT IL phase
<b>IL-OH</b>	701	2.511	656.04
<b>IL-Ester</b>	0	1.43	49.77
<b>Methanesulfonic acid</b>	7.01	4.21	2.80
<b>Levulinic acid</b>	100.24	9.33	80.80
<b>Water</b>	1000	963.26	36.74
<b>Total</b>	<b>1808.25</b>	<b>980.75</b>	<b>826.15</b>

$$\Sigma \text{IN} - \Sigma \text{OUT} = 1.35 \text{ g}$$

The water produced during the esterification reaction cannot be neglected. The IL and aqueous phases readily phase separate, and two streams exit the extractor. The IL phase identifies as the product stream since it carries the extracted carboxylic acid. The aqueous phase contains the unextracted carboxylic acid, hence it identifies as the raffinate phase. The principal component of solvent extraction-based processes is the solvent. Phase-trapping due to mutual solubility is undesirable as it leads to cumulative solvent losses. The re-use of solvent and minimization of solvent losses is of great importance. The entrainment of IL-OH in the aqueous stream can be reduced by improving phase separation and recycling the raffinate stream to the fermentation broth. The overall changes in the composition of both phases are dependent on the extraction and separation efficiencies.

The transesterification stage is driven by a chemical reaction and a new component, butyl levulinate is produced in the system, reducing both IL-Ester and levulinic acid while increasing IL-OH.

Table 4.14: Mass balance data for the transesterification stage.

Component	Mass/g	
	IN	OUT
<b>IL-OH</b>	656.04	695.06
<b>Methanesulfonic acid</b>	2.80	2.8
<b>Levulinic acid</b>	80.80	4.04
<b>Water</b>	36.74	48.65
<b>IL-Ester</b>	49.77	2.49
<b>Butanol</b>	581.51	526.27
<b>Butyl levulinate</b>		128.36
<b>Total</b>	<b>1407.67</b>	<b>1407.67</b>

$$\Sigma \text{IN} - \Sigma \text{OUT} = -0.0059 \text{ g}$$

The vacuum distillation stage is a separation process aimed towards the recovery of unreacted butanol from the transesterification mixture. The absence of a chemical reaction at this stage must be emphasised, therefore no accumulation or consumption of chemical species is expected. The recovered butanol is recycled back into the transesterification stage to improve mass utilisation efficiency.

Table 4.15: Mass balance data for the vacuum distillation stage.

Component	Mass/g		
	IN	OUT Condensate	OUT IL phase
<b>IL-OH</b>	695.06		695.06
<b>Methanesulfonic acid</b>	2.80		2.80
<b>Levulinic acid</b>	4.04		4.04
<b>Water</b>	48.65		48.65
<b>IL-Ester</b>	2.49		2.49
<b>Butanol</b>	526.27	526.27	
<b>Butyl levulinate</b>	128.36		128.36
<b>Total</b>	<b>1407.67</b>	<b>526.27</b>	<b>881.40</b>

$$\Sigma \text{IN} - \Sigma \text{OUT} = 0.00 \text{ g}$$

Table 4.16: Mass balance data for the solvent recovery of butyl levulinate.

Component	Mass/g		
	IN	OUT Extract phase	OUT IL phase
<b>IL-OH</b>	695.06		695.06
<b>Methanesulfonic acid</b>	2.80		2.80
<b>Levulinic acid</b>	4.04		4.04
<b>Water</b>	48.65		48.65
<b>IL-Ester</b>	2.49		2.49
<b>Butyl levulinate</b>	128.36	128.36	
<b>Total</b>	<b>881.40</b>	<b>128.36</b>	<b>753.04</b>

$$\Sigma \text{IN} - \Sigma \text{OUT} = 0.00 \text{ g}$$

Butyl levulinate is isolated from the regenerated IL-OH by means of solvent extraction. The regenerated IL-OH is recycled and reused without further purification. The overall mass balance is presented in Figure 4.34.

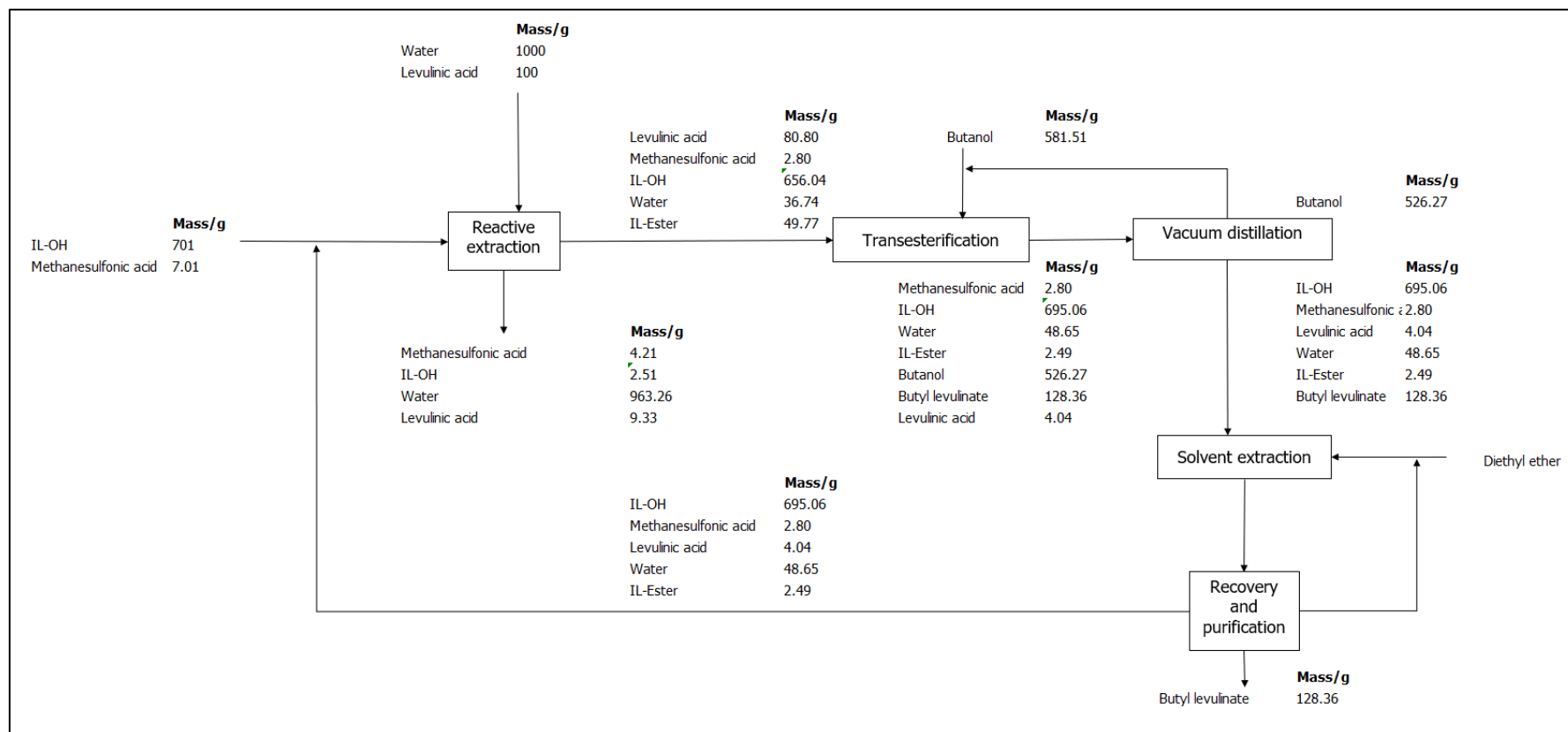
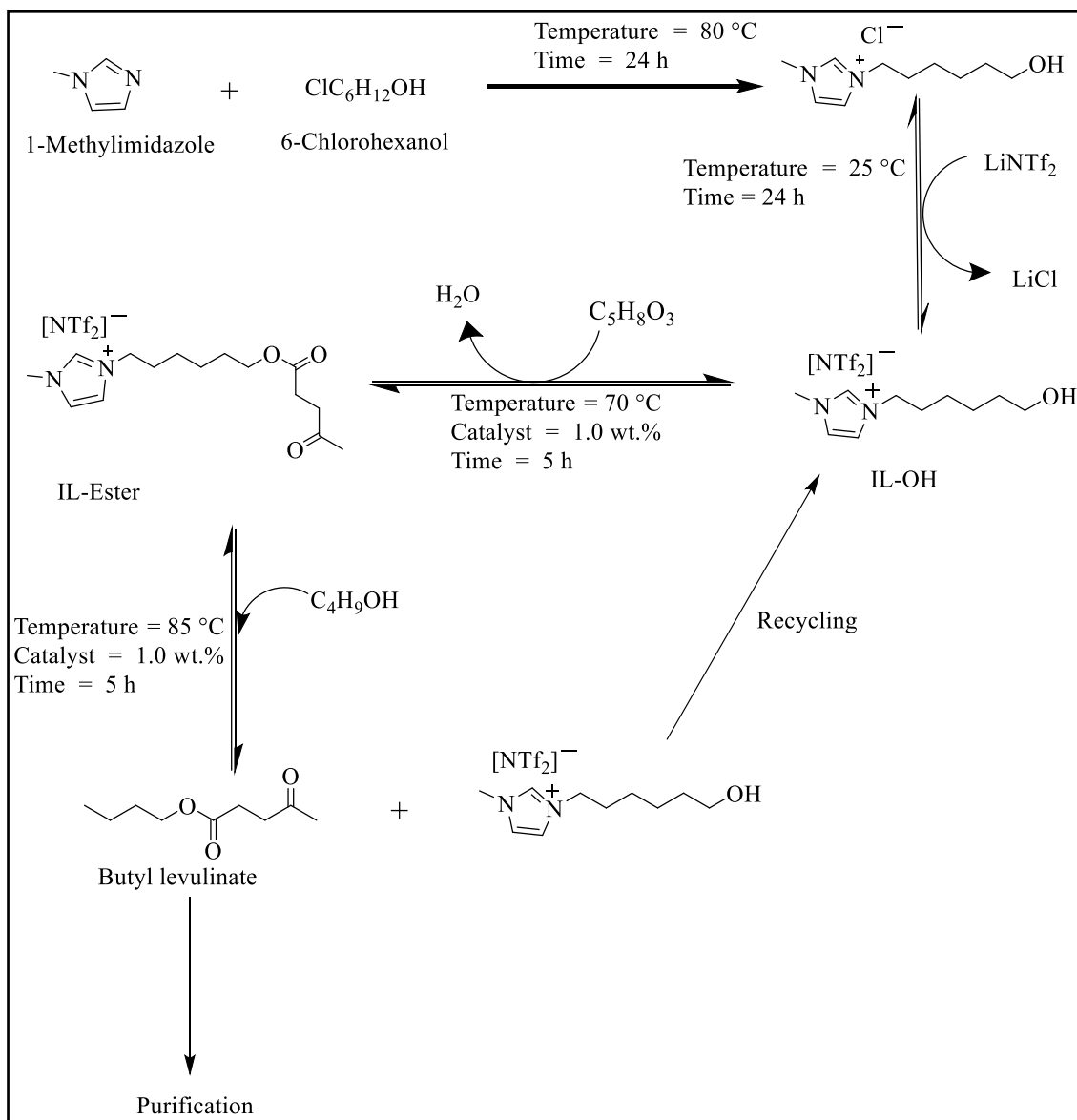


Figure 4.34: Overall mass balance for the reactive extraction of levulinic acid from aqueous solution using IL-OH.

## 4.10 Chapter summary

This chapter discussed the synthesis and application of a hydroxyl-functionalized IL in the reactive extraction of levulinic acid from aqueous solution. A summary of all the chemical reactions occurring during the synthesis and application of IL-OH are shown in Scheme 4.1. Levulinic acid was chosen as a model carboxylic acid and similar reactions are expected in the case of other carboxylic acids, as was the case for fumaric acid.



Scheme 4.1: Synthesis and application of IL-OH in the reactive extraction of levulinic acid.

Data derived from this study will serve as a basis for the design of IL-based reactive extraction systems, incorporating minor modifications, adjustments, and optimizations where necessary.

Minimal environmental risk is presented by the discharge of IL-OH and IL-Ester into the environment as predicted from their low  $K_{ow}$  values, that indicate little likelihood of bioaccumulation.



## **5 Conclusions, outlook, and future work**

The conclusions derived from the application of a hydroxyl-functionalized IL in the recovery of carboxylic acids such as levulinic and fumaric acids from aqueous solutions are described herein. The conclusions are based on the objectives and results of the study. The chapter also outlines the implications of the conclusions made herein, establishing the basis and recommendations for future work.

### **5.1 Conclusions**

The current study investigated the application of IL-OH as a reactive extractant for levulinic and fumaric acids from aqueous solution. The operating conditions for reactive extraction result in a noticeable mutual solubility between water and both IL-OH and IL-Ester. The mutual solubility of water and the ILs must be reduced to minimize solvent losses, thereby enhancing the economic viability of IL-based reactive extraction processes. Details of the conclusions are presented in the following subsections.

#### **5.1.1 Application of IL-OH as a reactive extractant**

IL-OH is an alcohol-functionalized IL that can extract carboxylic acids from aqueous solution by means of an esterification reaction. The chemical reaction enhances the extractability of the carboxylic acids from aqueous solutions. It can be concluded that the application of IL-OH as a reactive extractant for carboxylic acids from aqueous solution is an attractive alternative.

The extractability of both levulinic acid and fumaric acid were found to be dependent on the following factors:

- Initial concentration of the carboxylic acid, and
- Extraction mechanism (physical and chemical extraction).

In physical extraction, the extractability of both fumaric and levulinic acid was driven by the physical partitioning of the acids from the aqueous phase into the IL phase. When investigations were conducted using 0.034 M and 0.059 M solutions of fumaric and levulinic acid, respectively, the extraction efficiencies for fumaric and levulinic acid were found to be 40.13% and 82.80%, respectively. In the presence of 1.0 wt.% methanesulfonic acid as a catalyst, the extraction efficiency improved for both fumaric

and levulinic acid. The extraction efficiency of fumaric and levulinic acid improved to 75.68% and 93.03%, respectively.

Physical and chemical extraction efficiencies decreased with increasing initial acid concentration for both acids. Increasing the initial concentration of fumaric acid from 0.004 M to 0.034 M resulted in the extraction efficiency decreasing from 64.45 to 40.13% and 77.53 to 75.68% during chemical extraction, respectively. Similarly, the extraction efficiency of levulinic acid decreased from 93.88 to 82.80% and 96.76 to 91.87% during physical and chemical extraction from 0.059 M – 0.243 M solutions, respectively. However, the extractability of both fumaric and levulinic acid was significantly higher during chemical extraction.

Multiple carboxylic acids and alcohols are produced as co-products in a real fermentation broth. For example, formic acid and acetic acid are typical co-products of fermentation. The rate at which products accumulate is determined by the process conditions. The selectivity of the reactive extraction system is therefore influenced by the available composition of the products and feed. Extractive fermentation will convert the different carboxylic acids in the broth into different esters of the IL-OH which can easily be separated by reactive distillation. The reactive distillation can be designed based on the transesterification reaction. This reduces the need for auxiliary reagents and occurrence of undesirable side-reactions.

It is important to optimize product selectivity in the esterification of polycarboxylic acids with IL-OH. The nature of the ester present in the transesterification stage is influential to the reaction conditions needed for the efficient back-extraction of the extracted carboxylic acid.

### **5.1.2 Isolation of extracted carboxylic acid and regeneration of IL-OH**

The recovery of the chemically extracted carboxylic acid from the IL phase can be achieved by means of an acid-catalysed transesterification reaction. A stoichiometric excess of butanol is used to drive the reaction to completion. In principle, the choice of the resulting alky-ester can be customized through a careful selection of the alcohol used for the alcoholysis of IL-Ester. Furthermore, the physically extracted carboxylic acid will also react with the alcohol in an esterification reaction, producing the same alky-ester as

the transesterification reaction. The recovery of excess butanol from the transesterification mixture exceeds 95%, allowing for recirculation and recycling of the alcohol. Liquid-liquid extraction with diethyl ether can be employed to isolate and purify the alkyl-ester.

### **5.1.3 Environmental impact of IL-OH and IL-Ester**

As for other solvents, being used in the extraction of aqueous fermentation broths, there is a risk that ILs are emitted from the proposed process. While there is no biodegradability and toxicity data on these novel ILs developed, the environmental impact of IL-OH and IL-Ester was estimated from their respective  $K_{ow}$  values. The magnitudes of the  $K_{ow}$  values of both IL-Ester and IL-OH allow for the assumption that the environmental impact (bioaccumulation) of both ILs is minimal. IL-OH and IL-Ester have lower environmental impact than conventional solvents that are currently employed as extractants.

The environmental impact of the precursors, intermediates and degradation products involved in the preparation and utilization of ILs also needs to be clearly defined. The discharge and accumulation of ILs into the environment is inevitable. The degradation of IL-OH and IL-Ester in an aqueous environment must be investigated. A detailed study on the degradation mechanisms of hydroxyl- and ester-functionalized ILs must be conducted to establish the actual degradation products.

### **5.1.4 Process design and configuration**

The proposed model has clearly defined stages for the reactive extraction of carboxylic acids from aqueous solution with IL-OH. In the case of mixed-acid solutions, the reaction conditions can be designed to favour selectivity to multiple esters of IL-OH, which can later be transformed to alkyl esters during the transesterification stage. The resulting alkyl esters have different boiling points and can easily be separated by vacuum distillation. It is expected that the proposed model provides a conceptual framework for understanding the stages of IL-based reactive extraction, and thus serves as a basis for the design of reactive extraction processes.

## 5.2 Outlook

The proof of concept for the application of IL-OH for the reactive extraction of carboxylic acids has been established herein. The proposed IL displays properties of a promising reactive extractant and can be applied for the recovery any carboxylic acid. However, optimization of the individual process conditions will be required for different carboxylic acids. Secondly, novel IL-Esters can be synthesized and characterized to determine the physicochemical data. Such data is useful in predicting the phase behaviour during the application of IL-OH.

The use of IL-OH as a reactive extractant during extractive fermentation will promote the continuous removal of accumulating product(s), thereby reducing chances of end-product inhibition, thereby increasing the productivity of the microbes. The preparation and application of IL-OH must be evaluated based on a technoeconomic assessment criteria. The economic feasibility is used as a basis for financial investment as discussed by Dzigbor and Chimphango [171].

Finally, the presence of carboxylic acids in aqueous media can be attributed to both biochemical and chemical processes. Carboxylic acids are value-added chemicals, and their recovery, isolation, and purification are of great importance at industrial level. This study has developed a platform that can be applied for the recovery and isolation of a large range of carboxylic acids from aqueous media. Additionally, this platform may be applicable to the extraction of carboxylic acids from industrial waste waters to lower COD.

## 5.3 Future work

This study sets the foundation for further research on the selective recovery of carboxylic acids from aqueous solutions using IL-OH. The current study encountered several limitations which were beyond the scope of the study. To allow for the adoption of IL-based reactive extraction systems in biorefineries, there is need for further improvements of the reactive extraction process proposed herein. Furthermore, the extractability of carboxylic acids must also be investigated as a function of the productivity of the fermentation broth.

The application of IL-OH in the reactive extraction of carboxylic acids from aqueous solution demonstrates an integrated, extractive fermentation process. However, it must be noted, that the esterification reactive extraction is an acid-catalysed reaction, thus operates at suboptimal pH levels with respect to the fermentation process. Similarly, the transesterification reaction used for the back-extraction of the carboxylic acids and solvent regeneration is also an acid-catalysed reaction. Unfortunately, such operating conditions lead to reduced productivity during the fermentation process. Therefore, it is important to investigate the potential impact of the acid catalysts on extractive fermentation processes.

It is important to investigate the selectivity of the esterification of mixed carboxylic acids with IL-OH. Furthermore, product selectivity in the case of polycarboxylic acids must also be investigated. The optimal acid catalyst concentrations needed for the extractive esterification must be determined via response surface optimization.

Future work must be directed on establishing the kinetics of the reactive extraction of carboxylic acids with IL-OH. The effect of the chemical reaction on mass transfer during the reactive extraction of carboxylic acids with IL-OH must be explained using kinetic parameters. It is also important to determine whether the reaction is occurring in the aqueous phase, at the liquid interface, or in the IL phase.

The model proposed herein must be validated. The validations will establish the extent to which the model deviates from experimental data. Furthermore, the validated model can be used to estimate the energy requirements of the overall process, providing a detailed framework and methodology for calculating parameters such as feed targets, concentrations, and reactor sizing, among others. Future studies must attempt to integrate the proposed model in TEA models to establish optimal designs for IL-based extractive fermentation processes.

Finally, the proposed model can be modified and adapted for the recovery of other bio-based chemicals such as alcohols and phenols from aqueous solutions. This can be achieved by modifying the IL, through substituting the hydroxyl functionality with other reactive functionalities such as carboxyl and nitrile groups.

## References

- [1]. Wang, J., Lin, M., Xu, M., and Yang, S.-T. (2016). Anaerobic fermentation for production of carboxylic acids as bulk chemicals from renewable biomass. in: Hatti-Kaul, R., Mamo, G., and Mattiasson, B. (ed). *Anaerobes in Biotechnology*. Cham, Springer International Publishing. pp. 323-361.
- [2]. Karp, E. M., Cywar, R. M., Manker, L. P., Saboe, P. O., Nimlos, C. T., Salvachúa, D., Wang, X., Black, B. A., Reed, M. L., Michener, W. E., Rorrer, N. A., and Beckham, G. T. (2018). Post-fermentation recovery of biobased carboxylic acids. *ACS Sustainable Chemistry & Engineering*. 6(11), 15273-15283. doi:10.1021/acssuschemeng.8b03703.
- [3]. Kertes, A. S. and King, C. J. (1986). Extraction chemistry of fermentation product carboxylic acids. *Biotechnology and Bioengineering*. 28(2), 269-282. doi:10.1002/bit.260280217.
- [4]. Murali, N., Srinivas, K., and Ahring, B. K. (2017). Biochemical production and separation of carboxylic acids for biorefinery applications. *Fermentation*. 3(2), 22-47. doi:10.3390/fermentation3020022.
- [5]. Straathof, A. J. J. (2011). Proportion of downstream costs in fermentative production processes. in: Moo-Young, M. (ed). *Comprehensive Biotechnology*. Burlington, Academic Press. pp. 811-814.
- [6]. Hong, Y. K., Hong, W. H., and Han, D. H. (2001). Application of reactive extraction to recovery of carboxylic acids. *Biotechnology Bioprocess Engineering*. 6(1), 386 - 394.
- [7]. Din, N. S., Lim, S. J., Maskat, M. Y., Mutalib, S. A., and Zaini, N. M. (2021). Lactic acid separation and recovery from fermentation broth by ion-exchange resin: A review. *Bioresources and Bioprocessing*. 8, 31. doi:10.1186/s40643-021-00384-4.
- [8]. Notarfrancesco, S., Morselli, E., Martinez, G. A., Harasimiuk, W., Domingos, J. M. B., Negroni, A., Fava, F., and Bertin, L. (2021). Improved recovery of carboxylic acids using sequential cationic-anionic adsorption steps: A highly competitive ion-equilibrium model. *Separation and Purification Technology*. 261, 118253. doi:10.1016/j.seppur.2020.118253.
- [9]. Handojo, L., Wardani, A. K., Regina, D., Bella, C., Kresnowati, M. T. P., and Wenten, I. G. (2019). Electro-membrane processes for organic acid recovery. *RSC Advances*. 9(14), 7854-7869. doi:10.1039/C8RA09227C.
- [10]. Gössi, A., Burgener, F., Kohler, D., Urso, A., Kolvenbach, B. A., Riedl, W., and Schuur, B. (2020). In-situ recovery of carboxylic acids from fermentation broths through membrane supported reactive extraction using membrane modules with improved stability. *Separation and Purification Technology*. 241, 116694. doi:10.1016/j.seppur.2020.116694.
- [11]. Kakku, S., Gaikwad, S. M., Gaikwad, S., Taralkar, S. V., Billa, S. B., Chakinala, A. G., and Chakinala, N. (2022). Reactive extraction of gluconic acid Using trioctylamine in different diluents. *Chemical Engineering & Technology*. 45(3), 417-424. doi:10.1002/ceat.202100373.
- [12]. Shakya, A. K., Jaiswal, Y., Pal, S. L., and Srivastava, S. (2022). Reactive extraction of acetic acid by using tri-butyl-phosphate with different diluents. *Chemical Data Collections*. 39. doi:10.1016/j.cdc.2022.100855.

- [13]. Firdous, K. and Ahmad, S. A. (2020). Reactive extraction of valeric acid with a tributyl phosphate + diluent system. *Journal of Chemical & Engineering Data*. 65(11), 5096-5100. doi:10.1021/acs.jced.0c00086.
- [14]. Canari, R. and Eyal, A. M. (2003). Extraction of carboxylic acids by amine-based extractants: Apparent extractant basicity according to the pH of half-neutralization. *Industrial & Engineering Chemistry Research*. 42(7), 1285-1292. doi:10.1021/ie010578x.
- [15]. Chemarin, F., Moussa, M., Chadni, M., Pollet, B., Lieben, P., Allais, F., Trelea, I. C., and Athes, V. (2017). New insights in reactive extraction mechanisms of organic acids: An experimental approach for 3-hydroxypropionic acid extraction with tri-n-octylamine. *Separation and Purification Technology*. 179, 523-532. doi:10.1016/j.seppur.2017.02.018.
- [16]. Kumar, T. P., Vishwanadham, B., Rani, K. N. P., Mallikarjun, M., and Rao, V. V. B. (2010). Reactive extraction of levulinic acid from aqueous solutions with tri-n-octylamine (TOA) in 1-octanol: Equilibria, kinetics, and model development. *Chemical Engineering Communications*. 198(4), 572-589. doi:10.1080/00986445.2010.512549.
- [17]. Uslu, H. and Kirbaslar, S. I. (2008). Equilibrium studies of extraction of levulinic acid by (trioctylamine (TOA) plus ester) solvents. *Journal of Chemical and Engineering Data*. 53(7), 1557-1563. doi:10.1021/jc800099a.
- [18]. Matsumoto, M., Kawamura, Y., and Kondo, K. (2004). Synergistic extraction of organic acids with tri-n-octylamine and di-2-ethylhexylphosphoric acid. *Journal of Chemical Engineering of Japan*. 37, 1150-1154. doi:10.1252/jcej.37.1150.
- [19]. Tamada, J. A. and King, C. J. (1990). Extraction of carboxylic acids with amine extractants. 3. Effect of temperature, water coextraction, and process considerations. *Industrial & Engineering Chemistry Research*. 29(7), 1333-1338. doi:10.1021/ie00103a037.
- [20]. Tamada, J. A. and King, C. J. (1990). Extraction of carboxylic acids with amine extractants. 2. Chemical interactions and interpretation of data. *Industrial & Engineering Chemistry Research*. 29(7), 1327-1333. doi:10.1021/ie00103a036.
- [21]. Djas, M. and Henczka, M. (2018). Reactive extraction of carboxylic acids using organic solvents and supercritical fluids: A review. *Separation and Purification Technology*. 201(1), 106-119. doi:10.1016/j.seppur.2018.02.010.
- [22]. Keshav, A., Wasewar, K. L., and Chand, S. (2009). Recovery of propionic acid from an aqueous stream by reactive extraction: effect of diluents. *Desalination*. 244(1-3), 12-23. doi:10.1016/j.desal.2008.04.032.
- [23]. Datta, D., Kumar, S., Wasewar, K. L., and Babu, B. V. (2012). Comparative study on reactive extraction of picolinic acid with six different extractants (phosphoric and aminic) in two different diluents (benzene and decan-1-ol). *Separation Science and Technology*. 47(7), 997-1005. doi:10.1080/01496395.2011.638961.
- [24]. Datta, D., Kumar, S., and Uslu, H. (2015). Status of the reactive extraction as a method of separation. *Journal of Chemistry*. 2015(1), 1-16. doi:10.1155/2015/853789.
- [25]. Welton, T. (2018). Ionic liquids: A brief history. *Biophysical Reviews*. 10(3), 691-706. doi:10.1007/s12551-018-0419-2.

- [26]. Stark, A. and Seddon, K. R. (2007). Ionic liquids. in: *Kirk-Othmer encyclopaedia of chemical technology*. 5th ed. Seidel, A. (ed). (eds). Hoboken, New Jersey: John Wiley & Sons, Inc. pp. 836-920.
- [27]. Wilkes, J. S. (2002). A short history of ionic liquids — from molten salts to neoteric solvents. *Green Chemistry*. 4(2), 73-80. doi:10.1039/B110838G.
- [28]. Welton, T. (1999). Room-temperature ionic liquids. Solvents for synthesis and catalysis. *Chemical Reviews*. 99(8), 2071-2084. doi:10.1021/cr980032t.
- [29]. Marták, J. and Schlosser, S. (2019). Influence of anion and cation structure of ionic liquids on carboxylic acids extraction. *Frontiers in Chemistry*. 7, 10. doi:10.3389/fchem.2019.00117.
- [30]. Sprakel, L. M. J. and Schuur, B. (2019). Solvent developments for liquid-liquid extraction of carboxylic acids in perspective. *Separation and Purification Technology*. 211, 935-957. doi:10.1016/j.seppur.2018.10.023.
- [31]. Marták, J. and Schlosser, S. (2016). New mechanism and model of butyric acid extraction by phosphonium ionic liquid. *Journal of Chemical and Engineering Data*. 61(9), 2979-2996. doi:10.1021/acs.jced.5b01082.
- [32]. Marták, J. and Schlosser, Š. (2007). Extraction of lactic acid by phosphonium ionic liquids. *Separation and Purification Technology*. 57(3), 483-494. doi:10.1016/j.seppur.2006.09.013.
- [33]. Bai, Y., Yan, R., Huo, F., Qian, J., Zhang, X., and Zhang, S. (2017). Recovery of methacrylic acid from dilute aqueous solutions by ionic liquids through hydrogen bonding interaction. *Separation and Purification Technology*. 184, 354-364. doi:10.1016/j.seppur.2017.05.013.
- [34]. Blaga, A. C., Tucaliuc, A., and Kloetzer, L. (2022). Applications of ionic liquids in carboxylic acids separation. *Membranes*. 12(8), 771.
- [35]. Pandey, S., Chomal, N., Kamsonlian, S., and Kumar, S. (2018). Theoretical and experimental studies on extraction of carboxylic acids from aqueous solution using ionic liquids. *International Journal of Chemical Engineering and Applications*. 9(1). doi:10.18178/ijcea.2018.9.1.693.
- [36]. Ayan, E., Baylan, N., and Çehreli, S. (2020). Optimization of reactive extraction of propionic acid with ionic liquids using central composite design. *Chemical Engineering Research and Design*. 153, 666-676. doi:10.1016/j.cherd.2019.11.015.
- [37]. Baylan, N. (2020). Ionic liquids as green solvents for reactive separation of glutaric acid from water. *Water Air and Soil Pollution*. 231. doi:10.1007/s11270-020-04549-3.
- [38]. Riveiro, E., Villar, L., Sas, O. G., González, B., Canosa, J., and Domínguez, Á. (2019). Extraction of carboxylic acids from aqueous solutions by using [BMim][NTf2] and salting-out agents. *Journal of Chemical & Engineering Data*. 64(11), 4717-4723. doi:10.1021/acs.jced.9b00041.
- [39]. Aşçi, Y. S. (2017). Examination of the efficiency of ionic liquids in glycolic acid separation from aqueous solution by using reactive extraction method. *Journal of the Turkish Chemical Society, Section A: Chemistry*. 4(3), 981-992. doi:10.18596/jotcsa.322687.
- [40]. Tonova, K., Svinyarov, I., and Bogdanov, M. G. (2014). Hydrophobic 3-alkyl-1-methylimidazolium saccharinates as extractants for L-lactic acid recovery. *Separation and Purification Technology*. 125, 239-246. doi:10.1016/j.seppur.2014.02.001.



- [41]. Li, X., Zhao, D., Fei, Z., and Wang, L. (2006). Applications of functionalized ionic liquids. *Science in China Series B: Chemistry*. 49(5), 385-401. doi:10.1007/s11426-006-2020-y.
- [42]. Lee, S. (2006). Functionalized imidazolium salts for task-specific ionic liquids and their applications. *Chemical Communications*. (10), 1049-1063. doi:10.1039/B514140K.
- [43]. Tang, S., Baker, G. A., and Zhao, H. (2012). Ether- and alcohol-functionalized task-specific ionic liquids: attractive properties and applications. *Chemical Society reviews*. 41(10), 4030-4066. doi:10.1039/c2cs15362a.
- [44]. Zeng, Q., Qin, H., Cheng, H., Chen, L., and Qi, Z. (2019). Development of a reactive extraction process for isobutyl isobutyrate formation intensified by bifunctional ionic liquid. *Chemical Engineering Science: X*. 1, 100001. doi:10.1016/j.cesx.2018.100001.
- [45]. Mishra, B. B. and Devi, N. (2022). Application of bifunctional ionic liquids for extraction and separation of Eu<sup>3+</sup> from chloride medium. *Transactions of Nonferrous Metals Society of China*. 32(6), 2061-2070. doi:10.1016/S1003-6326(22)65930-2.
- [46]. Li, X., Zhao, D., Fei, Z., and Wang, L. (2006). Applications of functionalized ionic liquids. *Science in China Series B-Chemistry*. 49(5), 385-401. doi:10.1007/s11426-006-2020-y.
- [47]. Inyang, V. M. and Lokhat, D. (2020). Reactive extraction of propionic acid using trioctylamine in 1-decanol by response surface methodology optimization technique. *International Journal of Low-Carbon Technologies*. 15(2), 171-179. doi:10.1093/ijlct/ctz069.
- [48]. Inyang, V. and Lokhat, D. (2020). Kinetic studies on propionic and malic acid reactive extraction using trioctylamine in 1-decanol. *Chemical Papers*. 74(10), 3597-3604. doi:10.1007/s11696-020-01194-2.
- [49]. Kumar, A. and Thakur, A. (2019). Parametric optimization of green synergistic reactive extraction of lactic acid using trioctylamine, Aliquat336, and butan-2-ol in sunflower oil by response surface methodology. *Chemical Engineering Communications*. 206(8), 1072-1086. doi:10.1080/00986445.2018.1544898.
- [50]. Marcus, Y. and Hefter, G. (2006). Ion Pairing. *Chemical Reviews*. 106(11), 4585-4621. doi:10.1021/cr040087x.
- [51]. Marondedze, E. T., Azim, M. M., Martincigh, B. S., and Stark, A. (2023). Synthesis and physico-chemical characterization of the binary mixtures of hydroxyl- and ester-functionalized ionic liquids. *Journal of Molecular Liquids*. 379. doi:10.1016/j.molliq.2023.121596.
- [52]. Roa Engel, C. A., Straathof, A. J. J., Zijlmans, T. W., van Gulik, W. M., and van der Wielen, L. A. M. (2008). Fumaric acid production by fermentation. *Applied microbiology and biotechnology*. 78(3), 379-389. doi:10.1007/s00253-007-1341-x.
- [53]. Kamm, B., Grubber, P. R., and Kamm, M. (2006). *Biorefineries-Industrial processes and products. Status quo and future directions*. Weinheim, Wiley - VCH. pp.
- [54]. de Jong, E. and Jungmeier, G. (2015). Biorefinery Concepts in Comparison to Petrochemical Refineries. in: Pandey, A., et al. (ed). *Industrial Biorefineries & White Biotechnology*. Amsterdam, Elsevier. pp. 3-33.

- [55]. Cherubini, F. (2010). The biorefinery concept: Using biomass instead of oil for producing energy and chemicals. *Energy Conversion and Management*. 51(7), 1412-1421. doi:10.1016/j.enconman.2010.01.015.
- [56]. Cherubini, F., Jungmeier, G., Wellisch, M., Willke, T., Skiadas, I., Van Ree, R., and de Jong, E. (2009). Toward a common classification approach for biorefinery systems. *Biofuels, Bioproducts and Biorefining*. 3(5), 534-546. doi:10.1002/bbb.172.
- [57]. Isikgor, F. H. and Becer, C. R. (2015). Lignocellulosic biomass: a sustainable platform for the production of bio-based chemicals and polymers. *Polymer Chemistry*. 6(25), 4497-4559. doi:10.1039/C5PY00263J.
- [58]. Takkellapati, S., Li, T., and Gonzalez, M. A. (2018). An overview of biorefinery derived platform chemicals from a cellulose and hemicellulose biorefinery. *Clean technologies and environmental policy*. 20(7), 1615-1630. doi:10.1007/s10098-018-1568-5.
- [59]. Martin-Dominguez, V., Estevez, J., de Borja, O. F., Santos, V. E., and Ladero, M. (2018). Fumaric acid production: A biorefinery perspective. *Fermentation*. 4(2), 33-50.
- [60]. Das, R. K., Brar, S. K., and Verma, M. (2016). Fumaric acid: Production and application aspects. in: Kaur Brar, S., Jyoti Sarma, S., and Pakshirajan, K. (ed). *Platform Chemical Biorefinery*. Amsterdam, Elsevier. pp. 133-157.
- [61]. Xu, Q., Li, S., Huang, H., and Wen, J. (2012). Key technologies for the industrial production of fumaric acid by fermentation. *Biotechnology Advances*. 30(6), 1685-1696. doi:10.1016/j.biotechadv.2012.08.007.
- [62]. Herrero, A. A. (1983). End-product inhibition in anaerobic fermentations. *Trends in Biotechnology*. 1(2), 49-53. doi:10.1016/0167-7799(83)90069-0.
- [63]. Abunde, N. F., Asiedu, N., and Addo, A. (2017). Dynamics of inhibition patterns during fermentation processes-Zea Mays and Sorghum Bicolor case study. *International Journal of Industrial Chemistry*. 8(1), 91-99. doi:10.1007/s40090-016-0105-9.
- [64]. Garrett, B. G., Srinivas, K., and Ahring, B. K. (2015). Performance and stability of Amberlite™ IRA-67 ion exchange resin for product extraction and pH control during homolactic fermentation of corn stover sugars. *Biochemical Engineering Journal*. 94, 1-8. doi:10.1016/j.bej.2014.11.004.
- [65]. Xiao, Z., Cheng, C., Bao, T., Liu, L., Wang, B., Tao, W., Pei, X., Yang, S.-T., and Wang, M. (2018). Production of butyric acid from acid hydrolysate of corn husk in fermentation by *Clostridium tyrobutyricum*: kinetics and process economic analysis. *Biotechnology for Biofuels*. 11. doi:10.1186/s13068-018-1165-1.
- [66]. Nunraksa, N., Rattanasansri, S., Praiboon, J., and Chirapart, A. (2019). Proximate composition and the production of fermentable sugars, levulinic acid, and HMF from *Gracilaria fisheri* and *Gracilaria tenuistipitata* cultivated in earthen ponds. *Journal of Applied Phycology*. 31(1), 683-690. doi:10.1007/s10811-018-1552-9.
- [67]. Kumar, A., Shende, D. Z., and Wasewar, K. L. (2020). Production of levulinic acid: A promising building block material for pharmaceutical and food industry. *Materials Today: Proceedings*. 29(3), 790-793. doi:10.1016/j.matpr.2020.04.749.

- [68]. Pateraki, C., Papadaki, A., Koutinas, A., and Kachrimanidou, V. (2019). 2.59 - Biorefinery Engineering. in: Moo-Young, M. (ed). *Comprehensive Biotechnology (Third Edition)*. Oxford, Pergamon. pp. 879-892.
- [69]. Liu, H., Jin, Y., Zhang, R., Ning, Y., Yu, Y., Xu, P., Deng, L., and Wang, F. (2023). Recent advances and perspectives on production of value-added organic acids through metabolic engineering. *Biotechnology Advances*. 62, 108076. doi:10.1016/j.biotechadv.2022.108076.
- [70]. Expert Market Research: Global fumaric acid market outlook. (2023). [cited 17/08/2023]; Available from: <https://www.expertmarketresearch.com/reports/fumaric-acid-market>.
- [71]. Vantage Market Research: Levulinic acid market - Global industry assessment and forecast. (2023). [cited 14/07/2023]; Available from: <https://www.vantagemarketresearch.com/industry-report/levulinic-acid-market-1993>.
- [72]. Liang, C., Wang, Y., Hu, Y., Wu, L., and Zhang, W. (2019). Study of a new process for the preparation of butyl levulinate from cellulose. *ACS Omega*. 4(6), 9828-9834. doi:10.1021/acsomega.9b00735.
- [73]. Démolis, A., Essayem, N., and Rataboul, F. (2014). Synthesis and applications of alkyl levulinates. *ACS Sustainable Chemistry & Engineering*. 2(6), 1338-1352. doi:10.1021/sc500082n.
- [74]. Merck Chemicals: Levulinic acid. (2023). [cited 14/07/2022]; Available from: <https://www.sigmaaldrich.com/ZA/en/substance/levulinicacid11612123762>.
- [75]. Merck Chemicals: Ethyl levulinate. (2023). [cited 14/07/2023]; Available from: <https://www.sigmaaldrich.com/ZA/en/product/aldrich/122629>.
- [76]. TCI Chemicals: Butyl levulinate. (2023). [cited 14/07/2023]; Available from: <https://www.tcichemicals.com/US/en/p/L0138>.
- [77]. Datta, D., Babu, B. V., and Kumar, S. (2017). Equilibrium and thermodynamic studies on reactive extraction of nicotinic acid using a biocompatible extraction system. *Journal of Chemical & Engineering Data*. 62(10), 3431-3436. doi:10.1021/acs.jced.7b00457.
- [78]. Inyang, V. and Lokhat, D. (2021). Butyric acid reactive extraction using trioctylamine in 1-decanol: Response surface methodology parametric optimization technique. *Arabian Journal for Science and Engineering*. 46(7), 6567-6577. doi:10.1007/s13369-020-05255-2.
- [79]. Bart, H.-J. (2001). *Reactive extraction*. 1<sup>st</sup> ed. Berlin, Springer. pp 4-15.
- [80]. Glasstone, S. and Lewis, D. (1960). *Elements of physical chemistry*. 2<sup>nd</sup> ed. London, Macmillan and Co. Ltd. pp 379 -381.
- [81]. Hansen, J.-P. and McDonald, I. R. (2006). Chapter 10 - Ionic Liquids. in: Hansen, J.-P. and McDonald, I. R. (ed). *Theory of Simple Liquids (Third Edition)*. Burlington, Academic Press. pp. 291-340.
- [82]. Mu, T. C. and Han, B. X. (2014). Structures and thermodynamic properties of ionic liquids. in: Zhang, S., et al. (ed). *Structures and Interactions of Ionic Liquids*. Berlin, Springer-Verlag Berlin. pp. 107-139.
- [83]. Clough, M. T., Crick, C., Grasvik, J., Hunt, P., Niedermeyer, H., Welton, T., and Whitaker, O. (2015). A physicochemical investigation of ionic liquid mixtures. *Chemical Science*. 6(2), 1101-1114. doi:10.1039/c4sc02931c.

- [84]. Fillion, J. J. and Brennecke, J. F. (2017). Viscosity of ionic liquid–ionic liquid mixtures. *Journal of Chemical & Engineering Data*. 62(6), 1884-1901. doi:10.1021/acs.jced.7b00221.
- [85]. Niedermeyer, H., Hallett, J., Villar-Garcia, I., Hunt, P., and Welton, T. (2012). Mixtures of ionic liquids. *Chemical Society Reviews*. 41(23), 7780-7802. doi:10.1039/c2cs35177c.
- [86]. Holbrey, J. D., Reichert, W. M., Swatloski, R. P., Broker, G. A., Pitner, W. R., Seddon, K. R., and Rogers, R. D. (2002). Efficient, halide free synthesis of new, low cost ionic liquids: 1,3-dialkylimidazolium salts containing methyl- and ethyl-sulfate anions. *Green Chemistry*. 4(5), 407-413. doi:10.1039/b204469b.
- [87]. Ratti, R. (2014). Ionic liquids: Synthesis and applications in catalysis. *Advances in Chemistry*. 2014(2014), 58-74. doi:10.1155/2014/729842.
- [88]. Naert, P., Rabaey, K., and Stevens, C. V. (2018). Ionic liquid ion exchange: exclusion from strong interactions condemns cations to the most weakly interacting anions and dictates reaction equilibrium. *Green Chemistry*. 20(18), 4277-4286. doi:10.1039/C8GC01869C.
- [89]. Sowmiah, S., Srinivasadesikan, V., Tseng, M. C., and Chu, Y. H. (2009). On the chemical stabilities of ionic liquids. *Molecules*. 14(9), 3780-813. doi:10.3390/molecules14093780.
- [90]. Wang, X., Heinemann, F. W., Yang, M., Melcher, B. U., Fekete, M., Mudring, A.-V., Wasserscheid, P., and Meyer, K. (2009). A new class of double alkyl-substituted, liquid crystalline imidazolium ionic liquids—a unique combination of structural features, viscosity effects, and thermal properties. *Chemical Communications*. 2009(47), 7405-7407. doi:10.1039/B914939B.
- [91]. Mehdi, H., Binnemans, K., Van Hecke, K., Van Meervelt, L., and Nockemann, P. (2010). Hydrophobic ionic liquids with strongly coordinating anions. *Chemical Communications*. 46(2), 234-236. doi:10.1039/B914977E.
- [92]. Butler, B. J. and Harper, J. B. (2019). The effect of the structure of the anion of an ionic liquid on the rate of reaction at a phosphorus centre. *Journal of Physical Organic Chemistry*. 32. doi:10.1002/poc.3819.
- [93]. Hagiwara, R. and Ito, Y. (2000). Room temperature ionic liquids of alkylimidazolium cations and fluoroanions. *Journal of Fluorine Chemistry*. 105(2), 221-227. doi:10.1016/S0022-1139(99)00267-5.
- [94]. Jacquemin, J., Husson, P., Padua, A. A. H., and Majer, V. (2006). Density and viscosity of several pure and water-saturated ionic liquids. *Green Chemistry*. 8(2), 172-180. doi:10.1039/B513231B.
- [95]. Sideris, P. J., Chen, Y., Gobet, M., and Greenbaum, S. G. (2019). Alkyl chain length effects of hydroxyl-functionalized imidazolium ionic liquids in the ionothermal synthesis of LiFePO<sub>4</sub>. *Phosphorus, Sulfur, and Silicon and the Related Elements*. 194(4-6), 292-296. doi:10.1080/10426507.2018.1528260.
- [96]. Sardar, J., Maeorg, U., Krasnou, I., Baddam, V., ViktoriaGudkova, Krumme, A., Savest, N., Tarasova, E., and Viirsalu, M. (2016). Synthesis of polymerizable ionic liquid monomer and its characterization. *IOP Conference Series: Materials Science and Engineering*. 111, 012 -021. doi:10.1088/1757-899x/111/1/012021.
- [97]. Põhako-Esko, K., Taaber, T., Saal, K., Lõhmus, R., Kink, I., and Mäeorg, U. (2013). New method for synthesis of methacrylate-type polymerizable ionic liquids. *Synthetic Communications*. 43(21), 2846-2852. doi:10.1080/00397911.2012.745159.

- [98]. Voutsas, E. (2007). Estimation of the volatilization of organic chemicals from soil. in: Letcher, T. M. (ed). *Thermodynamics, Solubility and Environmental Issues*. Amsterdam, Elsevier. pp. 205-227.
- [99]. Ropel, L., Aki, S., Stadtherr, M., and Brennecke, J. (2005). Octanol-water partition coefficients of imidazolium-based ionic liquids. *Green Chemistry*. 7(2), 83-90. doi:10.1039/b410891d.
- [100]. Hermens, J. L. M., de Bruijn, J. H. M., and Brooke, D. N. (2013). The octanol–water partition coefficient: Strengths and limitations. *Environmental Toxicology and Chemistry*. 32(4), 732-733. doi:10.1002/etc.2141.
- [101]. Amézqueta, S., Subirats, X., Fuguet, E., Rosés, M., and Ràfols, C. (2020). Chapter 6 - Octanol-water partition constant. in: Poole, C. F. (ed). *Liquid-Phase Extraction*. Elsevier. pp. 183-208.
- [102]. Sangster, J. (1989). Octanol-water partition coefficients of simple organic compounds. *Journal of Physical and Chemical Reference Data*. 18(3), 1111-1229. doi:10.1063/1.555833.
- [103]. PubChem compound summary for CID 7909, methyl isobutyl ketone. (2022). [cited October 29]; Available from: <https://pubchem.ncbi.nlm.nih.gov/compound/Methyl-isobutyl-ketone>.
- [104]. PubChem compound summary for CID 14227, trioctylamine. (2022). [cited October 29]; Available from: <https://pubchem.ncbi.nlm.nih.gov/compound/Trioctylamine>.
- [105]. Montalbán, G. M., Collado-González, M. M., Trigo, R., Díaz Baños, F. G., and Vllora, G. (2015). Experimental measurements of octanol-water partition coefficients of ionic liquids. *Journal of Advanced Chemical Engineering*. 5(3). doi:10.4172/2090-4568.1000133.
- [106]. UNEP. (2001). Stockholm convention on persistent organic pollutants. in *The conference of plenipotentiaries*. Sweden.
- [107]. Rybinska, A., Sosnowska, A., Grzonkowska, M., Barycki, M., and Puzyn, T. (2016). Filling environmental data gaps with QSPR for ionic liquids: Modeling n-octanol/water coefficient. *Journal of Hazardous Materials*. 303, 137-144. doi:10.1016/j.jhazmat.2015.10.023.
- [108]. Cumming, H. and Rücker, C. (2017). Octanol–Water Partition Coefficient Measurement by a Simple <sup>1</sup>H NMR Method. *ACS Omega*. 2(9), 6244-6249. doi:10.1021/acsomega.7b01102.
- [109]. Ulrich, N., Goss, K.-U., and Ebert, A. (2021). Exploring the octanol–water partition coefficient dataset using deep learning techniques and data augmentation. *Communications Chemistry*. 4, 90. doi:10.1038/s42004-021-00528-9.
- [110]. Yu, S., Gao, S., Gan, Y., Zhang, Y., Ruan, X., Wang, Y., Yang, L., and Shi, J. (2016). QSAR models for predicting octanol/water and organic carbon/water partition coefficients of polychlorinated biphenyls. *SAR QSAR Environ Res*. 27(4), 249-63. doi:10.1080/1062936x.2016.1158734.
- [111]. Jordan, A. and Gathergood, N. (2015). Biodegradation of ionic liquids – a critical review. *Chemical Society Reviews*. 44(22), 8200-8237. doi:10.1039/C5CS00444F.
- [112]. de Jesus, S. S. and Maciel Filho, R. (2022). Are ionic liquids eco-friendly? *Renewable and Sustainable Energy Reviews*. 157. doi:10.1016/j.rser.2021.112039.

- [113]. Inyang, V. and Lokhat, D. (2020). Reactive extraction of malic acid using trioctylamine in 1-decanol: Equilibrium studies by response surface methodology using Box Behnken optimization technique. *Scientific Reports*. 10. doi:10.1038/s41598-020-59273-z.
- [114]. Kaur, G. and Elst, K. (2014). Development of reactive extraction systems for itaconic acid: a step towards in situ product recovery for itaconic acid fermentation. *RSC Advances*. 4(85), 45029-45039. doi:10.1039/C4RA06612J.
- [115]. Yalkowsky, S. H., He, Y., and Jain, P. (2010). *Handbook of aqueous solubility data*. 2<sup>nd</sup> ed. Boca Raton, CRC Press. pp 333.
- [116]. Stenström, S., Dalene, M., and Skarping, G. (1985). The solubility of tri-n-octylamine in water and phosphoric acid solutions. *Analytica Chimica Acta*. 177, 279-284. doi:10.1016/S0003-2670(00)82962-1.
- [117]. Santana, M. D. P., Lavrador, R. B., and Pessoa, P. D. (2022). Solvent screening for liquid-liquid extraction of levulinic acid from aqueous medium. *Separation Science and Technology*. 57(10), 1575-1584. doi:10.1080/01496395.2021.1993920.
- [118]. Curzons, A. D., Constable, D. C., and Cunningham, V. L. (1999). Solvent selection guide: a guide to the integration of environmental, health and safety criteria into the selection of solvents. *Clean Products and Processes*. 1(2), 82-90. doi:10.1007/s100980050014.
- [119]. Smallwood, I. (1996). *Handbook of organic solvent properties*. London, Arnold. pp 11, 137, 141, 101, 179, 273.
- [120]. Kakiuchi, T. (2008). Mutual solubility of hydrophobic ionic liquids and water in liquid-liquid two-phase systems for analytical chemistry. *Analytical Sciences*. 24(10), 1221-1230. doi:10.2116/analsci.24.1221.
- [121]. Dietz, M. L. (2006). Ionic Liquids as Extraction Solvents: Where do we Stand? *Separation Science and Technology*. 41(10), 2047-2063. doi:10.1080/01496390600743144.
- [122]. Freire, M. G., Santos, L. M. N. B. F., Fernandes, A. M., Coutinho, J. P. A., and Marrucho, I. M. (2007). An overview of the mutual solubilities of water–imidazolium-based ionic liquids systems. *Fluid Phase Equilibria*. 261(1-2), 449-454. doi:10.1016/j.fluid.2007.07.033.
- [123]. Anthony, J. L., Brennecke, J. F., Holbrey, J. D., Maginn, E. J., Mantz, R. A., Rogers, R. D., Trulove, P. C., Visser, A. E., and Welton, T. (2003). Physicochemical Properties of Ionic Liquids. in: Wasserscheid, P. and Welton, T. (ed). *Ionic Liquids in Synthesis*. Germany, Wiley-Blackwell. pp. 41-54.
- [124]. Vidiš, A., Ohlin, C. A., Laurenczy, G., Küsters, E., Sedelmeier, G., and Dyson, P. J. (2005). Rationalisation of solvent effects in the Diels-Alder reaction between cyclopentadiene and methyl acrylate in room temperature ionic liquids. *Advanced Synthesis & Catalysis*. 347(2-3), 266-274. doi:10.1002/adsc.200404301.
- [125]. Zhou, T., Chen, L., Ye, Y., Chen, L., Qi, Z., Freund, H., and Sundmacher, K. (2012). An overview of mutual solubility of ionic liquids and water predicted by COSMO-RS. *Industrial & Engineering Chemistry Research*. 51(17), 6256-6264. doi:10.1021/ie202719z.
- [126]. Seddon, K. R., Stark, A., and Torres, M. J. (2000). Influence of chloride, water, and organic solvents on the physical properties of ionic liquids. *Pure and Applied Chemistry*. 72(12), 2275-2287. doi:10.1351/pac200072122275.

- [127]. Mazan, V., Boltoeva, M. Y., Tereshatov, E. E., and Folden, C. M. (2016). Mutual solubility of water and hydrophobic ionic liquids in the presence of hydrochloric acid. *RSC Advances*. 6(61), 56260-56270. doi:10.1039/C6RA06791C.
- [128]. Freire, M. G., Neves, C. M. S. S., Carvalho, P. J., Gardas, R. L., Fernandes, A. M., Marrucho, I. M., Santos, L. M. N. B. F., and Coutinho, J. A. P. (2007). Mutual solubilities of water and hydrophobic ionic liquids. *The Journal of Physical Chemistry B*. 111(45), 13082-13089. doi:10.1021/jp076271e.
- [129]. Hanke, C. G. and Lynden-Bell, R. M. (2003). A simulation study of water–dialkylimidazolium ionic liquid mixtures. *The Journal of Physical Chemistry B*. 107(39), 10873-10878. doi:10.1021/jp034221d.
- [130]. Seddon, K. R. and Stark, A. (2002). Selective catalytic oxidation of benzyl alcohol and alkylbenzenes in ionic liquids. *Green Chemistry*. 4(2), 119-123. doi:10.1039/B111160B.
- [131]. Dupont, D., Depuydt, D., and Binnemans, K. (2015). Overview of the Effect of salts on biphasic ionic liquid/water solvent extraction systems: Anion exchange, mutual solubility, and thermomorphic properties. *The Journal of Physical Chemistry B*. 119(22), 6747-6757. doi:10.1021/acs.jpcb.5b02980.
- [132]. Alipour, S. and Omidvarborna, H. (2016). High concentration levulinic acid production from corn stover. *RSC Advances*. 6(112), 111616-111621. doi:10.1039/C6RA23768A.
- [133]. Toth, A., Siebenhofer, M., and Lux, S. (2021). Reactive distillation for solvent regeneration after biphasic carboxylic acid esterification. *Chemie Ingenieur Technik*. 93(10), 1493-1501. doi:10.1002/cite.202100074.
- [134]. Loudon, M. G. (1995). *Organic Chemistry*. 3<sup>rd</sup> ed. California, The Benjamins/Cummings Publishing Company. pp 946-947, 971-1012.
- [135]. Karthikeyan, P., Muskawar, P. N., Aswar, S. A., Bhagat, P. R., Kumar, S. S., Satvat, P. S., and Nair, A. N. B. (2016). L-Valine functionalized ionic liquid catalyzed esterification reaction under approach. *Arabian Journal of Chemistry*. 9(2), S1679-S1682. doi:10.1016/j.arabjc.2012.04.032.
- [136]. Keyes, D. B. (1932). Esterification processes and equipment. *Industrial & Engineering Chemistry*. 24(10), 1096-1103. doi:10.1021/ie50274a003.
- [137]. Rajabi, F., Abdollahi, M., and Luque, R. (2016). Solvent-free esterification of carboxylic acids using supported iron oxide nanoparticles as an efficient and recoverable catalyst. *Materials*. 9(7), 557. doi:10.3390/ma9070557.
- [138]. Zhang, Z.-T., Taylor, S., and Wang, Y. (2017). In situ esterification and extractive fermentation for butyl butyrate production with *Clostridium tyrobutyricum*. *Biotechnology and Bioengineering*. 114(7), 1428-1437. doi:10.1002/bit.26289.
- [139]. Oliveira, A. C., Rosa, M. F., Cabral, J. M. S., and Aires-Barros, M. R. (2001). Effect of extraction and enzymatic esterification of ethanol on glucose consumption by two *Saccharomyces cerevisiae* strains: A comparative study. *Journal of Chemical Technology and Biotechnology*. 76(3), 285-290. doi:10.1002/jctb.382.
- [140]. Vu, T. H. T., Au, H. T., Nguyen, T. H. T., Nguyen, T. T. T., Do, M. H., Bui, N. Q., and Essayem, N. (2013). Esterification of lactic acid by catalytic extractive reaction: An efficient way to produce a biosolvent composition. *Catalysis Letters*. 143(9), 950-956. doi:10.1007/s10562-013-1077-4.

- [141]. Keshav, A. and Wasewar, K. L. (2010). Back extraction of propionic acid from loaded organic phase. *Chemical Engineering Science*. 65(9), 2751-2757. doi:10.1016/j.ces.2010.01.010.
- [142]. Wasewar, K. L. (2012). Reactive extraction: An intensifying approach for carboxylic acid separation. *International Journal of Chemical Engineering and Applications*. 3(4), 249-255. doi:10.7763/ijcea.2012.v3.195.
- [143]. Teke, G. M. and Pott, R. W. M. (2021). Design and evaluation of a continuous semipartition bioreactor for in situ liquid-liquid extractive fermentation. *Biotechnology and Bioengineering*. 118(1), 58-71. doi:10.1002/bit.27550.
- [144]. Day, J. N. E. and Ingold, C. K. (1941). Mechanism and kinetics of carboxylic ester hydrolysis and carboxyl esterification. *Transactions of the Faraday Society*. 37(1941), 686-705. doi:10.1039/TF9413700686.
- [145]. Patai, S. (1969). *The chemistry of carboxylic acids and esters*. Chichester, Interscience-Publishers. pp 106 - 120.
- [146]. Stark, A., Behrend, P., Braun, O., Müller, A., Ranke, J., Ondruschka, B., and Jastorff, B. (2008). Purity specification methods for ionic liquids. *Green Chemistry*. 10(11), 1152-1161. doi:10.1039/b808532c.
- [147]. Ai, B., Li, J., Chi, X., Meng, J., Jha, A. K., Liu, C., and Shi, E. (2014). Effect of pH and buffer on butyric acid production and microbial community characteristics in bioconversion of rice straw with undefined mixed culture. *Biotechnology and Bioprocess Engineering*. 19(4), 676-686. doi:10.1007/s12257-013-0655-z.
- [148]. Yang, S.-T., Huang, H., Tay, A., Qin, W., De Guzman, L., and Nicolas, E. C. S. (2007). Extractive fermentation for the production of carboxylic acids. in: Yang, S.-T. (ed). *Bioprocessing for value-added products from renewable resources*. Heidelberg, Elsevier. pp. 421-446.
- [149]. Neves, C. M. S. S., Dinis, T. B. V., Carvalho, P. J., Schröder, B., Santos, L. M. N. B. F., Freire, M. G., and Coutinho, J. A. P. (2019). Binary mixtures of ionic liquids in aqueous solution: towards an understanding of their salting-in/salting-out phenomena. *Journal of Solution Chemistry*. 48(7), 983-991. doi:10.1007/s10953-018-0836-7.
- [150]. Freire, M. G., Carvalho, P. J., Silva, A. M. S., Santos, L. M. N. B. F., Rebelo, L. P. N., Marrucho, I. M., and Coutinho, J. A. P. (2009). Ion specific effects on the mutual solubilities of water and hydrophobic ionic liquids. *The Journal of Physical Chemistry B*. 113(1), 202-211. doi:10.1021/jp8080035.
- [151]. Wianowska, D., Gil, M., and Olszowy, M. (2020). Chapter 5 - Miniaturized methods of sample preparation. in: Hussain, C. M. (ed). *Handbook on Miniaturization in Analytical Chemistry*. Elsevier. pp. 99-125.
- [152]. Hyde, A. M., Zultanski, S. L., Waldman, J. H., Zhong, Y.-L., Shevlin, M., and Peng, F. (2017). General principles and strategies for salting-out informed by the Hofmeister series. *Organic Process Research & Development*. 21(9), 1355-1370. doi:10.1021/acs.oprd.7b00197.
- [153]. Chou, C. T. (2002). *Partition and adsorption of organic contaminants in environmental systems*. New Jersey, John Wiley & Sons Inc. pp 53 - 59.
- [154]. Lee, B.-S. and Lin, S.-T. (2014). A priori prediction of the octanol–water partition coefficient ( $K_{ow}$ ) of ionic liquids. *Fluid Phase Equilibria*. 363, 233-238. doi:10.1016/j.fluid.2013.11.042.



- [155]. Köddermann, T., Reith, D., and Arnold, A. (2013). Why the partition coefficient of ionic liquids is concentration-dependent. *The Journal of Physical Chemistry B*. 117(37), 10711-10718. doi:10.1021/jp405383f.
- [156]. Jain, P. and Kumar, A. (2016). Concentration-dependent apparent partition coefficients of ionic liquids possessing ethyl- and bi-sulphate anions. *Physical Chemistry Chemical Physics*. 18(2), 1105-1113. doi:10.1039/C5CP06611E.
- [157]. Dorbritz, S., Ruth, W., and Kragl, U. (2005). Investigation on aggregate formation of ionic liquids. *Advanced Synthesis & Catalysis*. 347(9), 1273-1279. doi:10.1002/adsc.200404352.
- [158]. Bandrés, I., Meler, S., Giner, B., Cea, P., and Lafuente, C. (2009). Aggregation behavior of pyridinium-based ionic liquids in aqueous solution. *Journal of Solution Chemistry*. 38(12). doi:10.1007/s10953-009-9474-4.
- [159]. PubChem Compound Summary for CID 444972, Fumaric acid. (2023). [cited January 25]; Available from: <https://pubchem.ncbi.nlm.nih.gov/compound/Fumaric-acid>.
- [160]. PubChem Compound Summary for CID 11579, Levulinic acid. (2023). [cited January 25]; Available from: <https://pubchem.ncbi.nlm.nih.gov/compound/Levulinic-acid>.
- [161]. Altiokka, M. R. and Çıtak, A. (2003). Kinetics study of esterification of acetic acid with isobutanol in the presence of amberlite catalyst. *Applied Catalysis A: General*. 239(1-2), 141-148. doi:10.1016/s0926-860x(02)00381-2.
- [162]. Goto, S., Tagawa, T., and Yusoff, A. (1991). Kinetics of the esterification of palmitic acid with isobutyl alcohol. *International Journal of Chemical Kinetics*. 23, 17-26. doi:10.1002/kin.550230103.
- [163]. Bart, J. C. J., Palmeri, N., and Cavallaro, S. (2010). Transesterification processes for biodiesel production from oils and fats. in: Bart, J. C. J., Palmeri, N., and Cavallaro, S. (ed). *Biodiesel Science and Technology*. Woodhead Publishing. pp. 285-321.
- [164]. Musa, I. A. (2016). The effects of alcohol to oil molar ratios and the type of alcohol on biodiesel production using transesterification process. *Egyptian Journal of Petroleum*. 25(1), 21-31. doi:10.1016/j.ejpe.2015.06.007.
- [165]. Harris, K. R., Kanakubo, M., and Woolf, L. A. (2007). Temperature and pressure dependence of the viscosity of the ionic liquids 1-hexyl-3-methylimidazolium hexafluorophosphate and 1-butyl-3-methylimidazolium bis(trifluoromethylsulfonyl)imide. *Journal of Chemical & Engineering Data*. 52(3), 1080-1085. doi:10.1021/je700032n.
- [166]. Avramov, I. (2009). Relationship between diffusion, self-diffusion and viscosity. *Journal of Non-Crystalline Solids*. 355(10), 745-747. doi:10.1016/j.jnoncrysol.2009.02.009.
- [167]. Blahušiak, M., Schlosser, Š., Cvengroš, J., and Marták, J. (2011). New approach to regeneration of an ionic liquid containing solvent by molecular distillation. *Chemical Papers*. 65(5), 603-607. doi:10.2478/s11696-011-0053-y.
- [168]. Marondedze, E. T. (2018). Hydroxyl-functionalized ionic liquids: Synthesis, characterization and application in the reactive extraction of levulinic acid from aqueous solution. MSc Thesis. University of KwaZulu-Natal. Durban.
- [169]. Mizzi, B., Meyer, M., Prat, L., Augier, F., and Leinekugel-Le-Cocq, D. (2017). General design methodology for reactive liquid-liquid extraction: Application to dicarboxylic acid recovery in fermentation broth. *Chemical Engineering and*

- Processing-Process Intensification.* 113(2017), 20-34. doi:10.1016/j.cep.2016.10.003.
- [170]. Banerjee, S., Munagala, M., Shastri, Y., Vijayaraghavan, R., Patti, A. F., and Arora, A. (2022). Process design and techno-economic feasibility analysis of an integrated pineapple processing waste biorefinery. *ACS Engineering Au.* 2(3), 208-218. doi:10.1021/acsengineeringau.1c00028.
- [171]. Dzigbor, A. and Chimphango, A. (2019). An integrated strategy targeting drying and cooling unit operations to improve economic viability and reduce environmental impacts in a mango processing plant. *Clean Technologies and Environmental Policy.* 21, 139-153. doi:10.1007/s10098-018-1623-2.
- [172]. Wildman, S. A. and Crippen, G. M. (1999). Prediction of physicochemical parameters by atomic contributions. *Journal of Chemical Information and Computer Sciences.* 39(5), 868-873. doi:10.1021/ci990307l.
- [173]. Diejomaoh Abase, O. T., Azim, M. M., Martincigh, B. S., and Stark, A. (2022). Cation-fluorinated ionic liquids: Synthesis, physicochemical properties and comparison with non-fluorinated analogues. *Journal of Molecular Liquids.* 349. doi:10.1016/j.molliq.2021.118104.
- [174]. Seki, S., Tsuzuki, S., Hayamizu, K., Umebayashi, Y., Serizawa, N., Takei, K., and Miyashiro, H. (2012). Comprehensive refractive index property for room-temperature ionic liquids. *Journal of Chemical & Engineering Data.* 57(8), 2211-2216. doi:10.1021/je201289w.
- [175]. Freire, M. G., Teles, A. R. R., Rocha, M. A. A., Schröder, B., Neves, C. M. S. S., Carvalho, P. J., Evtuguin, D. V., Santos, L. M. N. B. F., and Coutinho, J. A. P. (2011). Thermophysical characterization of ionic liquids able to dissolve biomass. *Journal of Chemical & Engineering Data.* 56(12), 4813-4822. doi:10.1021/je200790q.
- [176]. Deetlefs, M., Seddon, K. R., and Shara, M. (2006). Predicting physical properties of ionic liquids. *Physical Chemistry Chemical Physics.* 8(5), 642-649. doi:10.1039/B513453F.
- [177]. Bonhôte, P., Dias, A., Papageorgiou, N., Kalyanasundaram, K., and Grätzel, M. (1996). Hydrophobic, highly conductive ambient-temperature molten salts. *Inorganic Chemistry.* 35(5), 1168-1178. doi:10.1021/ic951325x.
- [178]. Huddleston, J. G., Visser, A. E., Reichert, W. M., Willauer, H. D., Broker, G. A., and Rogers, R. D. (2001). Characterization and comparison of hydrophilic and hydrophobic room temperature ionic liquids incorporating the imidazolium cation. *Green Chemistry.* 3(4), 156-164. doi:10.1039/B103275P.

## Appendix A: HPLC data

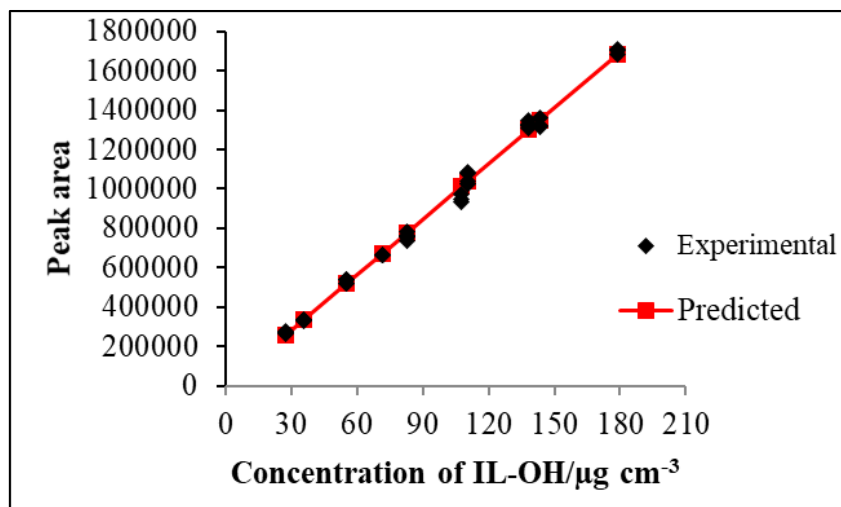


Figure A 1: HPLC calibration curve for determination of IL-OH at 211 nm.

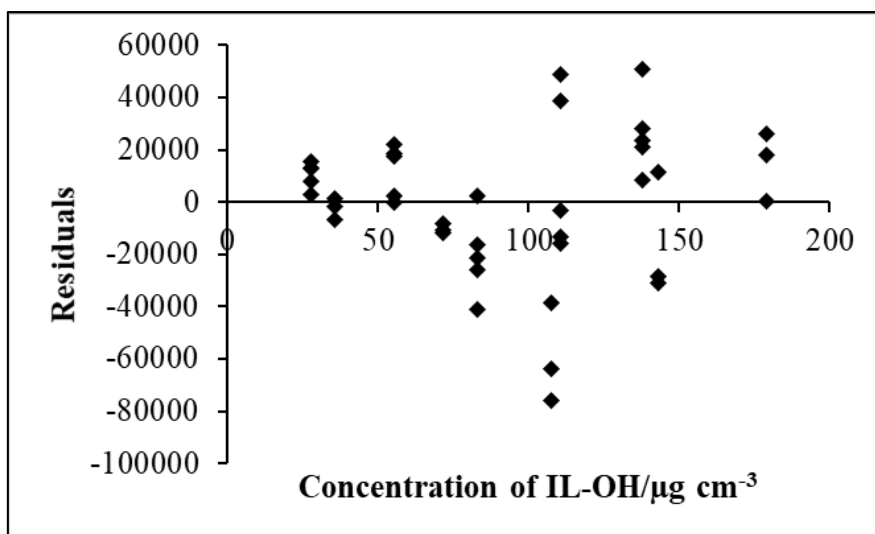


Figure A 2: Residuals plot for the calibration curve for the determination of IL-OH.

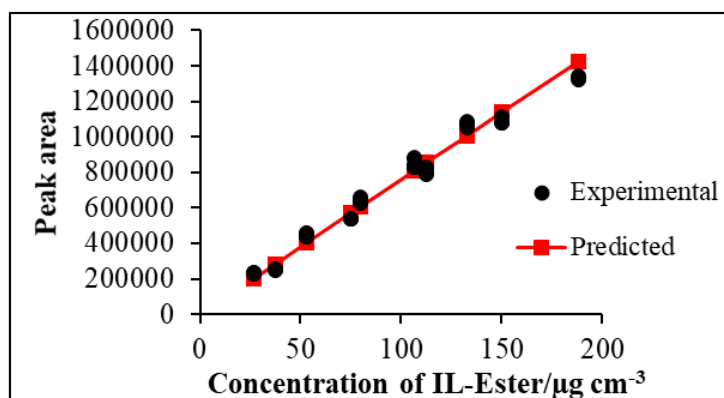


Figure A 3: HPLC calibration curve for determination of IL-Ester at 211 nm.

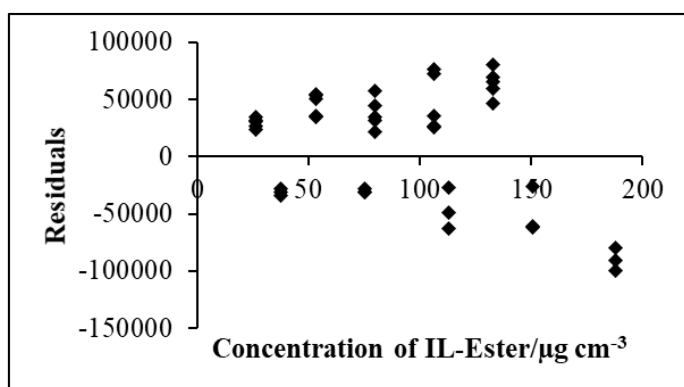


Figure A 4: Residuals plot for the calibration curve for the determination of IL-Ester.

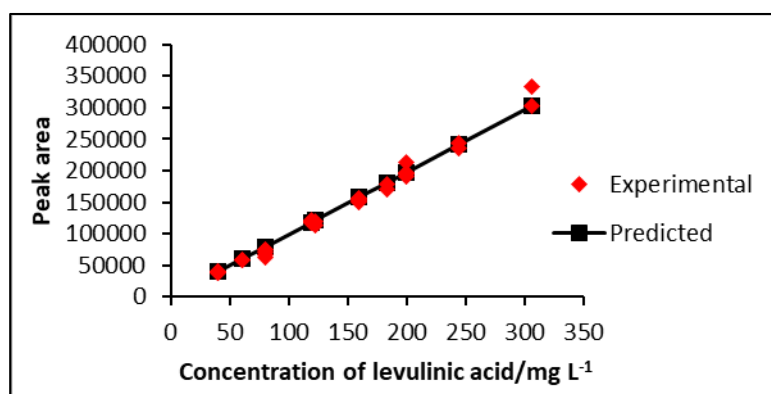


Figure A 5: HPLC calibration curve for determination of levulinic acid at 210 nm.

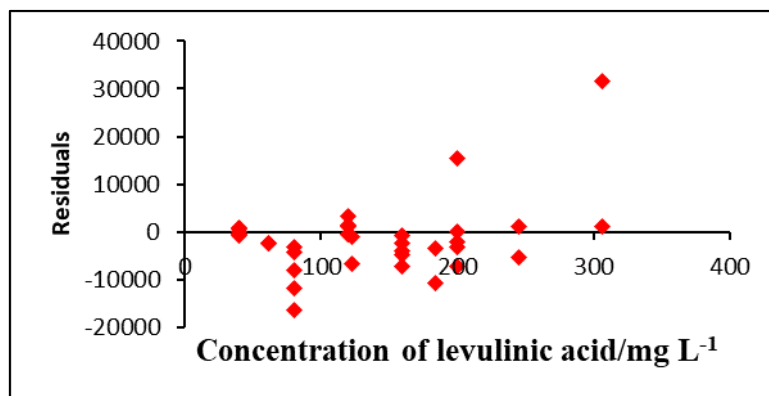


Figure A 6: Residuals plot for the calibration curve for the determination of levulinic acid.

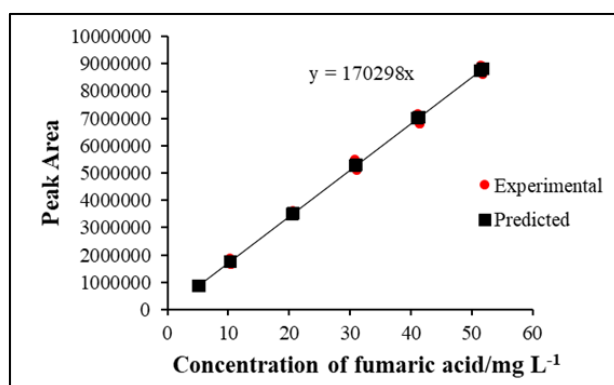


Figure A 7: HPLC calibration curve for determination of fumaric acid at 211 nm.

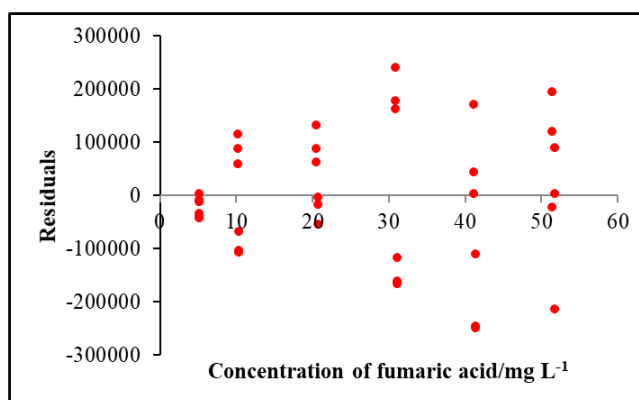


Figure A 8: Residuals plot for the calibration curve for the determination of fumaric acid.

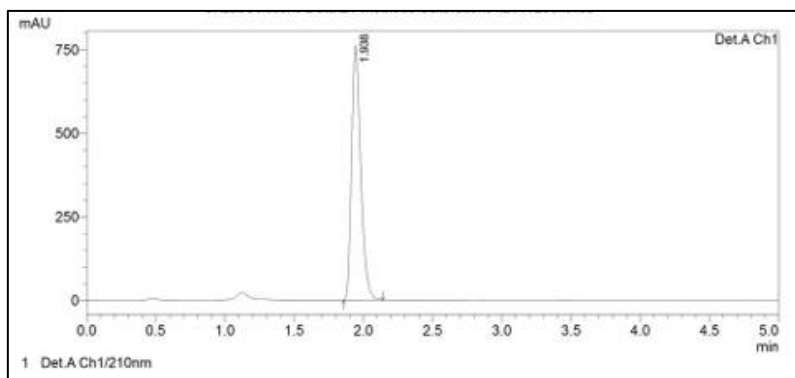


Figure A 9: Sample chromatogram for the elution of fumaric acid at 1.908 minutes using the HPLC method.

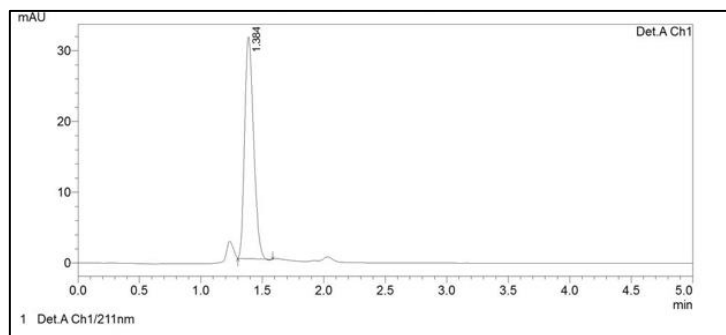


Figure A 10: Sample chromatogram for the elution of levulinic acid at 1.384 minutes using the HPLC method.

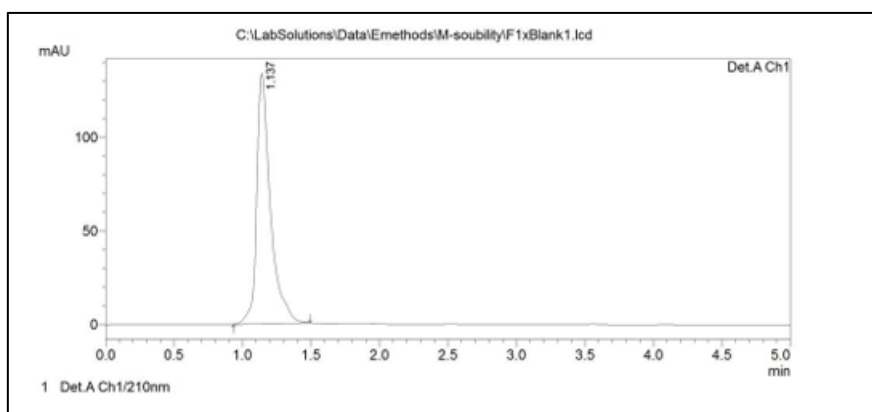


Figure A 11: Sample chromatogram for the blank injection during the HPLC analysis. The solvent front was eluted at  $t = 1.137$  minutes.

Table A 1: Analytical precision in the determination of IL-OH.

Concentration /mg L <sup>-1</sup>	Intra-day (n = 6)		Accuracy/ %	Inter-day (n = 3)		Accuracy/ %
	Mean	stdev		Mean	stdev	
10	10.08	0.21	100.8	9.92	0.47	99.2
50	50.85	0.71	101.7	51.06	1.31	102.12
200	200.06	0.74	100.03	198.37	1.70	99.19

Table A 2: Analytical precision in the determination of levulinic acid.

Concentration /mg L <sup>-1</sup>	Intra-day (n = 6)		Accuracy/ %	Inter-day (n = 3)		Accuracy/ %
	Mean	stdev		Mean	%RSD	
60	58.71	0.01	95.97	60.32	2.39	100.53
120	118.45	2.89	98.71	121.08	1.58	100.90
180	176.44	3.59	98.02	178.63	3.74	99.24

Table A 3: Analytical precision in the determination of fumaric acid.

Concentration /mg L <sup>-1</sup>	Intra-day (n = 6)		Accuracy/ %	Inter-day (n = 3)		Accuracy/ %
	Mean	%RSD		Mean	%RSD	
10	5.03	0.09	100.61	9.72	0.88	97.19
20	20.84	0.34	104.21	20.03	0.46	100.16
50	51.77	0.75	103.53	51.42	1.07	102.84

Table A 4: Analytical precision in the determination of IL-Ester.

Concentration /mg L <sup>-1</sup>	Intra-day (n = 6)		Accuracy/ %	Inter-day (n = 3)		Accuracy/ %
	Mean	%RSD		Mean	%RSD	
30	29.84	2.16	99.47	31.43	2.37	104.77
75	76.07	1.85	101.43	74.86	2.10	99.81
120	119.63	2.17	99.69	120.51	1.99	100.43

## Appendix B: GC data

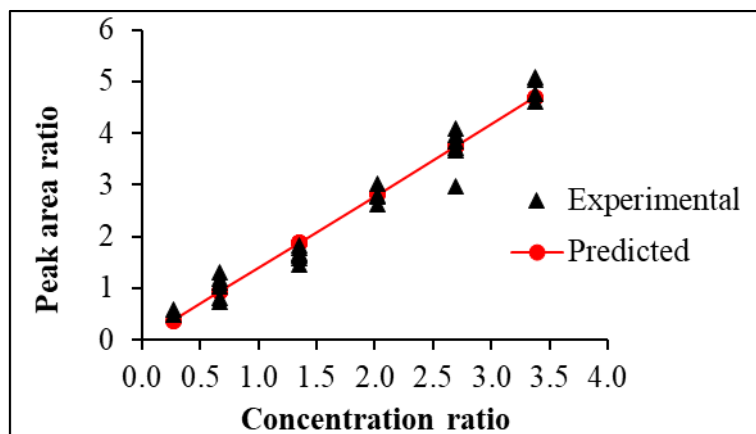


Figure B 1: GC-FID calibration curve for the determination of butanol in the presence of ethyl levulinate as an internal standard.

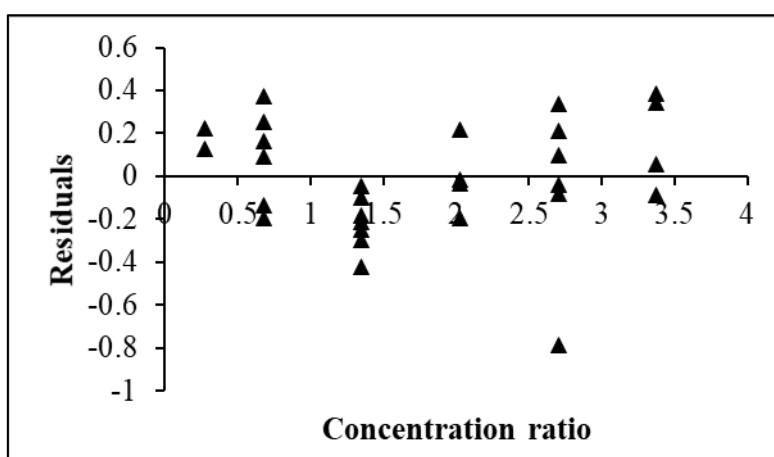


Figure B 2: Residuals plot for the calibration curve for the determination of butanol using GC-FID.



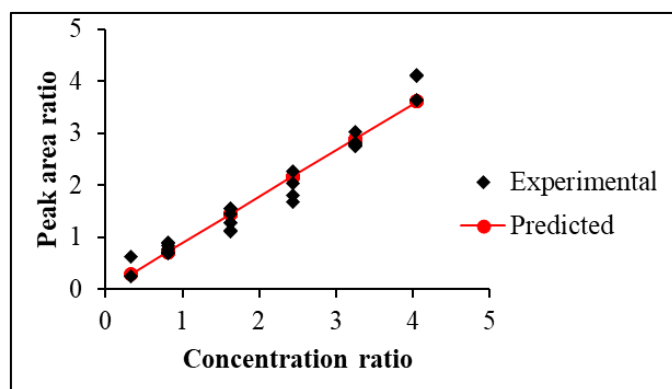


Figure B 3: GC-FID calibration curve for the determination of butyl levulinate in the presence of ethyl levulinate as an internal standard.

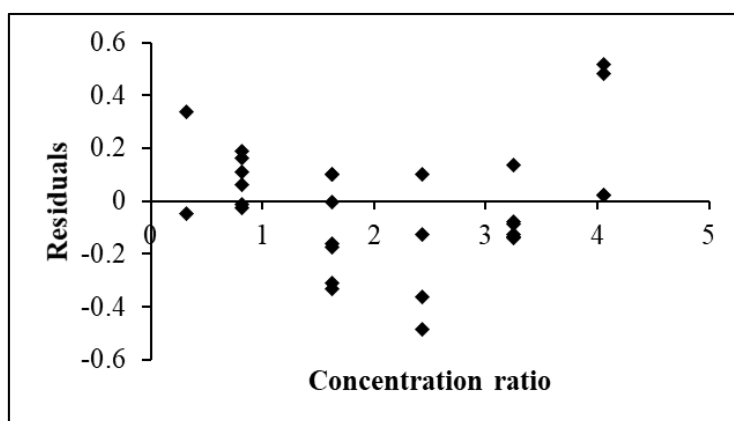


Figure B 4: Residuals plot for the calibration curve for the determination of butyl levulinate using GC-FID.

Table B 1: Analytical precision in the determination of butanol (n = 6).

Concentration /mg L <sup>-1</sup>	Intra-day		Accuracy %	Inter-day		Accuracy%
	Mean	%RSD		Mean	%RSD	
50	49.37	1.29	98.74			
85				88.05	0.63	103.58

Table B 2: Analytical precision in the determination of butyl levulinate (n = 6).

Concentration /mg L <sup>-1</sup>	Intra-day		Accuracy %	Inter-day		Accuracy%
	Mean	%RSD		Mean	%RSD	
100	101.18	2.35	106.76	99.04	2.38	99.04

## Appendix C: Extraction data

Table C 1: Physical extraction of fumaric acid with IL-OH at 25 °C.

Initial concentration/g L <sup>-1</sup>	Equilibrium concentration/ g L <sup>-1</sup>		K <sub>D</sub>	E%
	Aqueous phase	IL phase		
0.4644	0.1651	0.2993	1.8128	64.4488
0.9603	0.3820	0.5783	1.5138	60.2208
1.9127	0.9249	0.9878	1.0680	51.6443
2.8730	1.5901	1.2829	0.8068	44.6537
3.9356	2.3563	1.5793	0.6702	40.1286

Table C 2: Reactive extraction of fumaric acid with IL-OH at 25 °C.

Initial concentration/ g L <sup>-1</sup>	Equilibrium concentration/ g L <sup>-1</sup>		K <sub>D</sub>	E%
	Aqueous phase	IL phase		
0.3467	0.0779	0.2688	3.4506	77.5310
1.0402	0.2088	0.8314	3.9818	79.9269
1.7336	0.4632	1.2704	2.7427	73.2810
2.4271	0.6941	1.8958	2.7313	73.1997
3.9000	0.9486	2.9514	3.1113	75.6769

Table C 3:Physical extraction of levulinic acid with IL-OH at 25 °C.

Initial concentration/M	Equilibrium concentration/M		K <sub>D</sub>	E%
	Aqueous phase	IL phase		
0.059	0.0036	0.0551	15.3056	93.8671
0.118	0.0144	0.1040	7.2222	87.8378
0.176	0.0251	0.1513	6.0279	85.7710
0.243	0.0417	0.2008	4.8153	82.8041

Table C 4: Reactive extraction of levulinic acid with IL-OH at 25 °C.

Initial concentration/M	Equilibrium concentration/M		$K_D$	E%
	Aqueous phase	IL phase		
0.0587	0.0019	0.0568	29.8947	96.7632
0.1184	0.0054	0.1131	20.9444	95.4430
0.1764	0.0101	0.1663	16.4654	94.2744
0.2425	0.0169	0.2256	13.3491	93.0309
0.3225	0.0262	0.2962	11.3053	91.8735

*Conversion of pH to concentration*

The concentration of an aqueous phase can be derived from pH values by using Equation C 1.

$$pH = -\log [H^+] \quad (C\ 1)$$

where the molar concentration of the hydrogen ion is denoted by  $[H^+]$

Table C 5: Converting Between pH and hydrogen ion concentration.

Experiment	Aqueous phase pH			
	Initial	Final	Initial $[H^+]/M$	Final $[H^+]/M$
(IL-OH + 1.0 wt.% catalyst) + water	6.87	1.55	$1.35 \times 10^{-7}$	0.028
IL-OH + (water + 1.0 wt.% catalyst)	1.31	1.52	0.049	0.030

## Appendix D: Group contribution data

The densities of the IL-OH precursor ([HOC<sub>6</sub>MIM]Cl), IL-OH, and IL-Ester were estimated from their refractive indices, respectively. The group contribution method developed by Wildman and Crippen [172] was applied. This method estimates the molar refractivity of a compound by totalling the atomic contributions using tabulated data presented in Table D 1.

Table D 1: Atom type description and contributions to obtain  $P_{\text{calc}}$  [172].

Wildman and Crippen descriptor type (i)	Description	Atom contribution (a)
H1	H (any in the ILs assessed, except -OH)	1.057
H2	-OH	1.395
C1	CH <sub>3</sub> in acetate, methane sulfonate (anion); aliphatic CH <sub>3</sub> and CH <sub>2</sub> (levulinate and cation, except for those specified under C3),	2.503
C3	CH <sub>3</sub> or CH <sub>2</sub> -CH <sub>2</sub> on N of imidazolium and pyridinium	2.753
C4	CF <sub>3</sub> in [CF <sub>3</sub> SO <sub>3</sub> ] <sup>-</sup> and [NTf <sub>2</sub> ] <sup>-</sup>	2.731
C5	Carboxyl and carbonyl C in [CH <sub>3</sub> CO <sub>2</sub> ] <sup>-</sup> , and ester of levulinic acid	5.007
C11	Aromatic in imidazolium and pyridinium	2.582
CS	CF <sub>3</sub> and CF <sub>2</sub> of perfluoroalkyl chains	3.243
N14	N in imidazolium and pyridinium	3.359
NS	N in [NTf <sub>2</sub> ] <sup>-</sup>	2.134
S2	S in [NTf <sub>2</sub> ] <sup>-</sup> , [CF <sub>3</sub> SO <sub>3</sub> ] <sup>-</sup> and [CH <sub>3</sub> SO <sub>3</sub> ] <sup>-</sup>	7.365
OS	O in [CF <sub>3</sub> SO <sub>3</sub> ] <sup>-</sup> and [CH <sub>3</sub> SO <sub>3</sub> ] <sup>-</sup>	0.6865
O1	O in [NTf <sub>2</sub> ] <sup>-</sup>	1.080
O2	OH	0.8238
O3	-O- in levulinic acid ester	1.085
O9	=O in [CH <sub>3</sub> CO <sub>2</sub> ] <sup>-</sup> and ester of levulinic acid	0.000
Cl	chloride	5.853
F	F in [NTf <sub>2</sub> ] <sup>-</sup> , [CF <sub>3</sub> SO <sub>3</sub> ] <sup>-</sup> and perfluoroalkyl chains	1.108
I	iodide	14.02

The experimentally determined refractive index (at a given temperature) is then applied to calculate the density according to Equation D 1.

$$V_M = \frac{M}{\rho} \quad (\text{D } 1)$$

where  $V_M$ ,  $M$  and  $\rho$  are the molar volume, molar mass, and density, respectively.

From an experimental point of view, this is of interest because refractive index measurements require a much smaller sample size, and feature a smaller error compared to density measurements. Figure D 1 shows the correlation of the experimental and refractive index-derived densities of twenty ILs, including those of this and as well as a previous study on cation-fluorinated ILs conducted in our research group by Diejomaoh Abafe et al. [173]. The coefficient of determination over the whole data set is  $> 0.98$ , with an average standard deviation of 1.66%. It must be noted that Figure D 1 is based on experimental data for refractive index and density measurements at the same temperature for a given IL (see Table D 2).

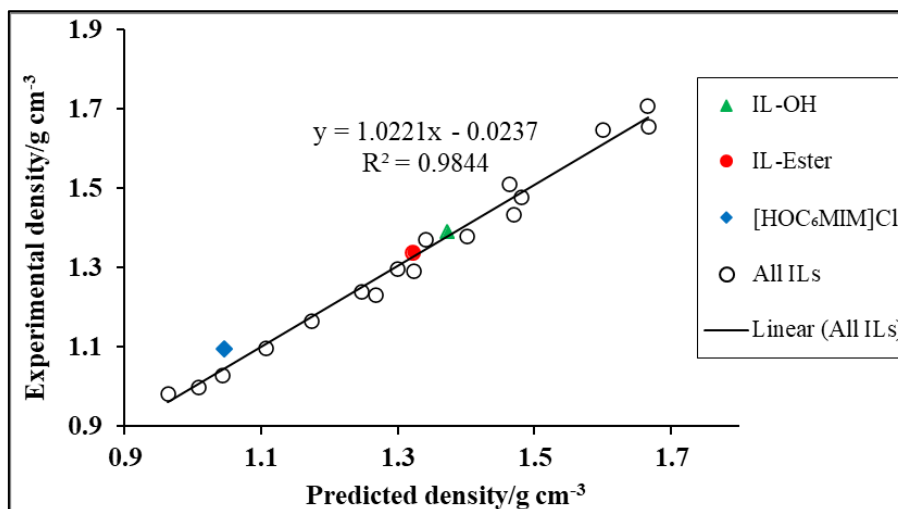


Figure D 1: Correlation of the experimental and refractive index-derived densities of a large set of ILs, Table D 2.

Also, from the temperature-dependent refractive index data of this study [168], the corresponding density at a required temperature could be obtained with coefficients of determinations  $> 0.99$  for both the pure IL-OH and IL-Ester, as well as a 1:1 mixture of [IL-OH + IL-Ester] over the temperature range from 30 – 70 °C as shown in Figure D2.

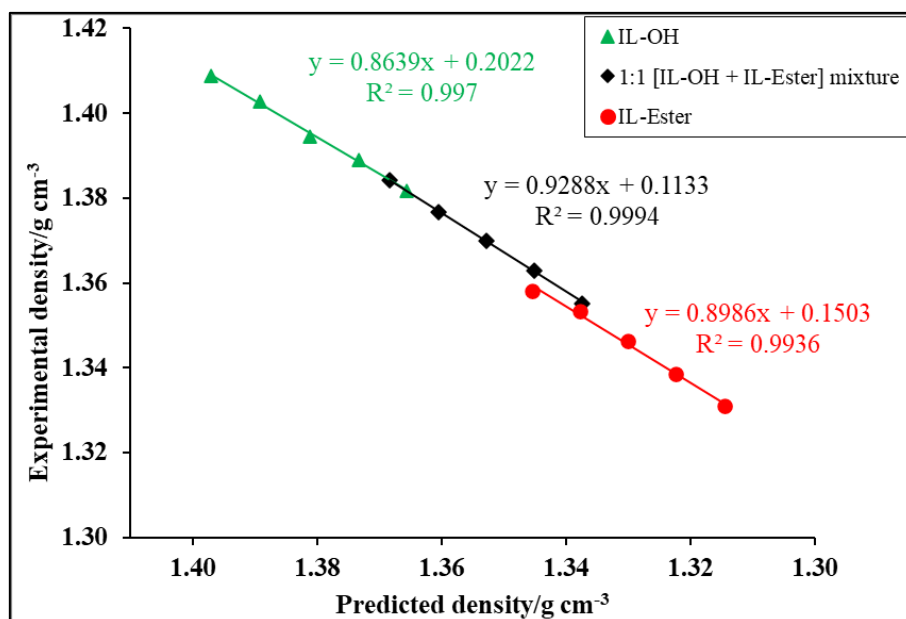


Figure D 2: Correlation of the experimental and refractive index-derived densities of IL-OH, IL-Ester and their 1:1 mixture at temperatures between 30 and 70 °C.

Table D 2: Prediction of density from molar refraction.

Ionic liquid	R (group contrib.)	M /g mol <sup>-1</sup>	n (exp)	Density (calc) /g cm <sup>-3</sup>	Density (exp) /g cm <sup>-3</sup>	Error /%	Ref. density data
[F <sub>13</sub> C <sub>8</sub> MIM][OAc]	77.8360	488.25	1.3881	1.4805	1.4786	0.13	60 °C; [173]
[F <sub>13</sub> C <sub>8</sub> MIM][OTf]	81.2615	578.27	1.3844	1.6653	1.7094	-2.58	60 °C; [173]
[F <sub>13</sub> C <sub>8</sub> MIM][NTf <sub>2</sub> ]	100.4490	709.34	1.3704	1.5988	1.6487	-3.03	60 °C; [173]
[F <sub>13</sub> C <sub>8</sub> Py][NTf <sub>2</sub> ]	98.4440	706.34	1.3811	1.6663	1.6566	0.58	60 °C; [173]
[C <sub>8</sub> MIM][OAc]	72.7330	254.37	1.4625	0.9624	0.9826	-2.06	60 °C; [173]
[C <sub>8</sub> MIM][OTf]	76.1585	344.39	1.4318	1.1724	1.1668	0.48	60 °C; [173]
[C <sub>8</sub> MIM][NTf <sub>2</sub> ]	95.3460	475.47	1.4222	1.2677	1.2316	2.93	60 °C; [173]
[C <sub>8</sub> Py][NTf <sub>2</sub> ]	93.3410	472.46	1.4354	1.3219	1.2935	2.19	60 °C; [173]
[C <sub>2</sub> MIM][NTf <sub>2</sub> ]	67.6440	391.30	1.4198	1.4633	1.5130	-3.29	30 °C; [174]
[C <sub>2</sub> MIM][OAc]	45.0310	170.21	1.4973	1.1066	1.0990	0.70	25 °C; [175]
[C <sub>2</sub> MIM][MeSO <sub>3</sub> ]	48.0755	206.26	1.4928	1.2464	1.2400	0.52	20 °C; [174]
[C <sub>4</sub> MIM][NTf <sub>2</sub> ]	76.8780	419.36	1.4510	1.4687	1.4363	2.26	25 °C; [176]
[C <sub>4</sub> MIM][OTf]	57.6905	288.29	1.4330	1.2987	1.2968	0.15	20 °C; [177]

[C <sub>6</sub> MIM]I	66.8380	294.18	1.5490	1.4000	1.3814	1.35	25 °C; [176]
[C <sub>6</sub> MIM][NTf <sub>2</sub> ]	86.1120	447.41	1.4295	1.3408	1.3710	-2.20	25 °C; [177]
[C <sub>6</sub> MIM]Cl	58.6710	202.73	1.5150	1.0420	1.0300	1.16	25 °C; [178]
[C <sub>8</sub> MIM]Cl	67.9050	230.78	1.5050	1.0080	1.0000	0.80	25 °C; [178]
IL-OH	87.2738	463.41	1.4306	1.3733	1.3890	-1.13	60 °C; This work
IL-Ester	111.0620	561.51	1.4361	1.3223	1.3385	-1.21	60 °C; This work
[HOC <sub>6</sub> MIM]Cl	59.8328	218.73	1.4840	1.0459	1.0944	-4.43	60 °C; This work

## Appendix E: NMR spectra

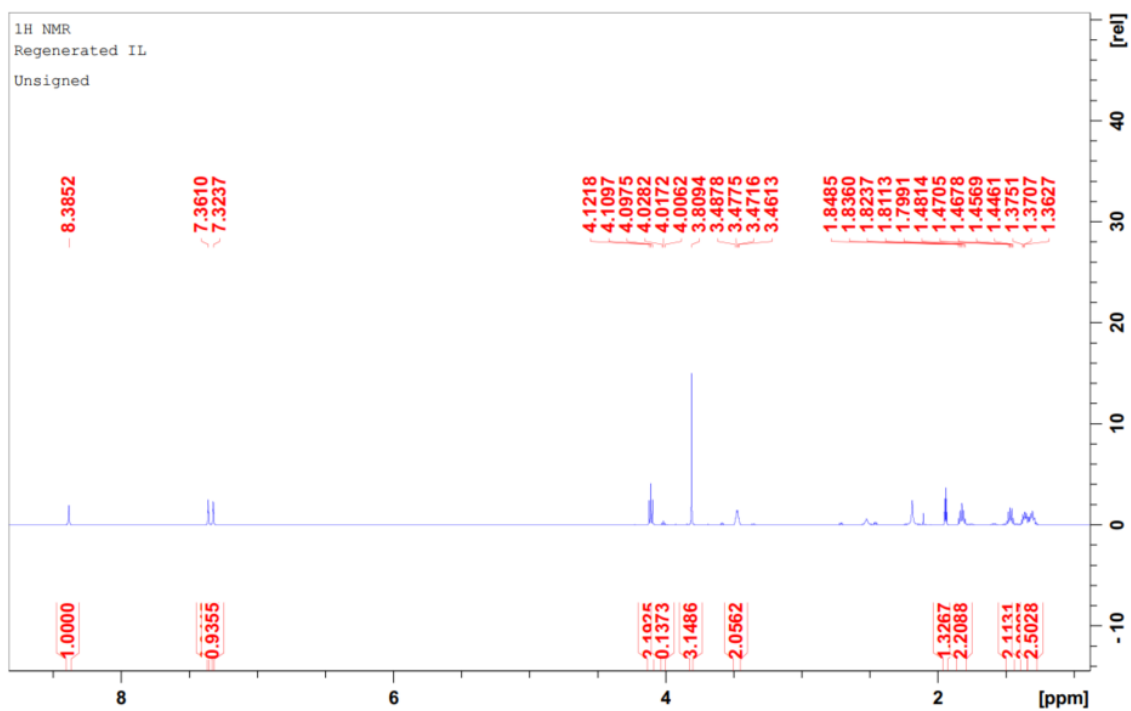


Figure E 1: <sup>1</sup>H NMR spectrum of the regenerated IL in acetonitrile-d<sub>3</sub>.