QUANTUM STRUCTURE ACTIVITY RELATIONSHIP (QSAR) METHOD TO PREDICT THE TOXICITY OF IONIC LIQUIDS.

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CHEMICAL ENGINEERING
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Quantum structure activity relationship (QSAR) method to predict the toxicity of ionic liquids

by

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17172

Dissertation submitted in partial fulfilment of

the requirements for the

Bachelor of Engineering (Hons)

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SEPTEMBER 2015

Universiti Teknologi PETRONAS,
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CERFICATION OF APPROVAL

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A project dissertation submitted to the

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BACHELOR OF ENGINEERING (Hons)
(CHEMICAL ENGINEERING)

Approved by,

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SEPTEMBER 2015

CERTIFICATION OF ORIGINALITY

This is to certify that I am responsible for the work submitted in this project, that the original work is my own except as specified in the references and acknowledgements and that the original work contained herein have not been undertaken or done by unspecified sources or persons.

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RUKESH PRUSHOTHMAN

ABSTRACT

This study focuses on the prediction of the toxicity of ionic liquid. Despite the existence of other methods with their limitation to predict toxicity of ionic liquid such as the quantitative structure activity relationship, partial least square discriminant, acute toxicity and etc., quantum structure activity relationship (QSAR) method have been used in this study to predict the toxicity of IL's at molecular level using density functional theory (DFT) based computational programme code of $Dmol^3$. The objectives are basically broken down into three level. The first level is to identify the molecular structure of the toxic ionic liquids which may give harmful effects to the environment. Next, once the samples have been identified, the geometries of molecular structure of IL's are optimized and calculate the values of the toxicity descriptors such as electrophilicity index (ω), hardness ($\boldsymbol{\eta}$), chemical potential (μ) and Total energy (E_t). Lastly, a four parameter regression correlation is developed between the values of the toxic ionic liquids and toxicity descriptors to observe the activity. Predicted toxicity of IL's have been validated with reported toxicity Equimolar Concentration 50 (EC_{50}) via experiment. So, quantum structure activity relationship (QSAR) method successfully predicted the IL's toxicity precisely as compared to other prediction method. From these correlation, the identified descriptors which contribute to the toxicity are known and an analyses is performed on the results obtained to show if the density functional theory is as effective as the other methods present. The scope of study used is basically on the density functional theory and also on the other methods available to predict toxicity so that a good set of comparison can be obtained while using the density functional theory. The research methodology present in this project is firstly to identify the molecular structure of the toxic ionic liquid. Then, the structures are optimized using the MS software to obtain values of the descriptors using the Density functional theory to make a predictive toxicity model of ionic liquids and compare these values with the existing toxicity value EC_{50}.

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CHAPTER 1

INTRODUCTION

1.1 Background study

Toxicity usually represents the measure of its harmful effects on living organisms. Toxicity can be classified into three categories which are chemical toxicity, biological toxicity and physical toxicity. Chemical toxicity includes both organic and inorganic where both have been proven to cause harmful effects on living organisms, for example, inorganic substances such as table salt, lead, and water primarily attacks the nervous system whereas organic substances such as the methyl alcohol effects the kidneys or liver . In our concern for this research paper, the focus is on the chemical toxicity from ionic liquids. Ionic liquids (ILs) have remarkable properties such as thermal stability, negligible vapour pressure, non-flammability and wide electrochemical potential window (Zhao, Zhao et al. 2014). Ionic liquids are also used extensively in the chemical processes including catalysis, extraction, electrochemistry and separation. It was also found that the ionic liquids do not contribute much to the air pollution but a certain amount of ionic liquids do get released into the soil or water after usage and hence may cause persistent pollutants and pose environment risks.

Lately, the toxicity of ionic liquids have been given more attention. Studies have shown that ionic liquids do give harmful effect to bacteria, plants, invertebrates, fish and human. Hence, ionic liquids are not so much regarded as an intrinsically "green". Since the interest on ionic liquids has raised, it's only feasible to come up with a solution to measure its toxicity without the sacrifice of animals as lab tests. This could be done by a computational approach towards the properties of the ionic liquids and developing a model to analyse the toxicity levels of the liquid. The most important theory that needs to be implemented in this research paper will be the Density Functional Theory. The density functional theory are used to compute the descriptors such as chemical potential, ionization potential, electrophilicity index,

as well as electron affinity of those ionic liquids. All these properties are later compared with the different experimental data done on laboratory animals. With these comparison, the toxicity is determined with just the information needed from the density functional theory.

1.2 Problem statement

In this project, the main concern will be to develop the density functional theory based theoretical model based on toxicity descriptors such as electrophilicity index (ω), Total energy (E_t), hardness (***η***) and chemical potential (μ) so that we can observe the co-relation of these descriptors and compare the results obtained with toxicity through experimentation. It will be much more functional to predict toxicity of a compound based on justified theoretical approach instead of conducting the lab tests on laboratory animals and in the same time getting exposed to the chemicals. (Zhao, Zhao et al.2014), have investigated roughly 100 ionic liquids and its molecular structure. They have used the Quantitative Structure-Activity relationship (QSAR) model to predict the toxicities (EC50 values) of these various ionic liquids on leukaemia rat cell line IPC-81. Four parameters were selected by the heuristics method (HM) are used to perform the studies of multiple linear regression (MLR) and support vector machine (SVM). Later, the methods are compared to see which of the results are more accurate.

As mentioned earlier, QSAR is another method used to predict toxicity using modelling but there is also other available methods such as the acute toxicity. Basically, these acute toxicity are obtained via laboratory experiment whereby they would expose the laboratory animals mainly mice and hamsters and observe the effect on the animal. It will usually take a very long time to obtain results because at times the substances take a very long time to react and hence it is not a very feasible method since the world is now fast moving. Data are needed immediately in order to prevent any further risk to the environment and humans. Besides that, the expenses are also very high since you need to hire labour work and lab technician to be in the lab constantly and observe.

Furthermore, there is also a method known as the mathematical modelling using the partial least square discriminant analysis developed by Manuel Alvarez-Guerra and Angel Irabien. It is most definitely a good method to approach toxicity.

However, the reliability of the data are not finalized before the model can fulfil its potential, it needs to be tested against other data sets such as toxicity data for cells of other species and higher organism. It is definitely time consuming. Hence, with the introduction of the density functional theory on predicting toxicity on ionic liquids will definitely reduce the use of laboratory animals as well as minimize cost since the need of labour is reduced or so does the need of animals to conduct the experiment.

In this project, the main concern will be to develop the theoretical model based toxicity descriptors such as electrophilicity index (ω), Total energy (E_t), hardness ($\boldsymbol{\eta}$) and chemical potential (μ) so that the characteristics of these descriptors can be observed and the results were compared to the toxicity values through experimentation.

1.3 OBJECTIVES and SCOPE OF STUDY

The main objective of this project is to develop a theoretical model equation to predict the toxicity of ionic liquids. The scope of this project will be to identify the theoretical model using density functional theory and compare with the experimental results found in literature which has used laboratory animals to obtain results.

i. Identifying the molecular structure of the most toxic ionic liquids.
ii. Optimizing the identified molecular structure using Material Studio 8.0 (MS) with the Dmol3 program package to obtain values for toxicity descriptors such as electrophilicity index (ω), Total energy (E_t), hardness ($\boldsymbol{\eta}$) and chemical potential (μ)
iii. Developing a correlation between toxicity of IL's (EC_{50}) and toxicity descriptors using the four parameter regression model.

CHAPTER 2

LITERATURE REVIEW AND THEORY

2.1 Toxicity

Toxicity is basically the degree of measure at which a substance can damage an organism (Salam, M. (2011). Usually the measurements for toxicity is referred to as Equimolar Concentration (EC_{50}) and its units are mg/L. The subscript EC_{50} (Equimolar Concentration, 50%) is defined as the dose required to kill half of the members of the specific animal population depending on the route of entrance of the toxic substance (Thuy Pham, Cho et al. 2010). For instance, it was found that EC_{50} are normally measured for liquids. Hence, that is the dosage that would have a toxic effect on any organism if exposed to any form of toxic substance. Besides that, toxicity is divided into three types which are chemical toxicity, biological toxicity and physical toxicity. Chemical toxicity is basically toxic exposure to chemicals which may also include inorganic substances such as lead, mercury, chlorine and etc. (Tsarpali and Dailianis 2015). On the other hand, biological toxicity is toxic effect on the living organism by bacteria and viruses. Furthermore, another most common type is the physical toxicants which means the interaction of the physical nature with the biological processes. For example, inhaling coal dust and finely defined silicon dioxide can be fatal and they may destroy tissues in the body.

In our research, the focus is on the toxicity of ionic liquids if exposed to the environment. Although the rise in the interest on ionic liquid just bloomed due to its physical properties which may be an alternative replacement for organic substances, the study on the toxicity of ionic liquid is still considered to be at its infancy stage. Hence, the exposure to the environment is basically still not thoroughly explored. It is important to know of its effect on the environment since now most industries are already using these ionic liquids to replace the volatile organic solvents. Accidental spills are bound to happen during handling of these liquids. Therefore, a better

assessment on this ionic liquids and its toxic effect should be conducted and this research is definitely focusing on this problem and possibly finding simpler ways to produce data on these ionic liquids as these ionic liquids are constantly being developed and more substances are available. In the case of environment exposure, toxicologist is primarily concerned with ionic liquids being exposed to aquatic organisms and its ecosystem.

2.2 Toxicity of ionic liquids

Ionic liquids (ILs) are organic salts with very low melting point and hence is considered as an environmental friendly substance and replacements for industrial volatile organic compounds (Thuy Pham, Cho et al. 2010). It is typically formed by large organic cation with delocalized charges and organic or inorganic anions which are not strongly attached to the organic cation. For example, the formation of 1-Butyl-3-methylimidizaloium is shown in the diagram below:-

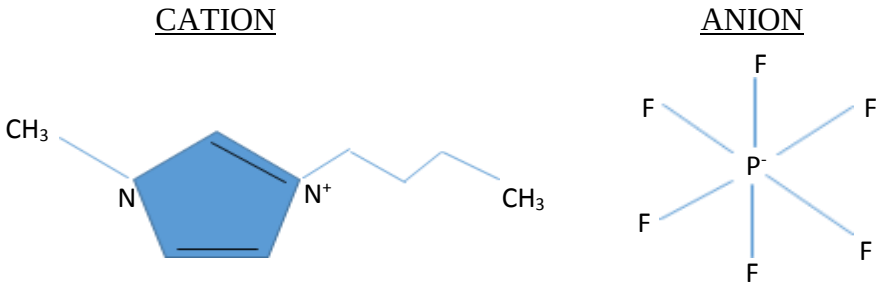


FIGURE 2.2. The formation of 1-butyl-3-methylimidazolium Hexafluorophosphate

Quite a number of study has been conducted on the toxicity of the ionic liquids to the environment. One of the most notable research was the "Investigation of toxic effects of imidazolium ionic liquids on marine mussel Mytilus gallaoprovincialis with or without the presence of conventional solvents, such as acetone". The mussels were first exposed to the different concentrations of 1-butyl-3methylimidazolium with or without the presence of acetone for 96 hours and the concentration that causes 50% toxicity (LD_{50}) was determined.

Besides that, there is also a study on the "Acute and Chronic Toxicity of Imidazolium-based ionic liquids on Daphnia Magna". Based on this research, it was found that the toxicity appeared to be related to the imidazolium cation and not the various anion. The experiment was conducted by exposing the imidazolium ionic

liquids with various anions. The lethal concentration ranged from a median lethal concentration (LC_{50}) which ranged from 8.03 to 19.91 mg L^{-1} whereas another set of experiment was conducted with salts which consist of sodium cation (Na^+X^-) where the order was of higher magnitude 9344.81 mg L^{-1} and 4765.75 mg L^{-1}. Thus the toxicity appeared to be on the cation and not the various anions.

The advantages of an ionic liquid compared to the usual organic solvents used in the industry are firstly, the volatility of these substances to the atmosphere and leads to the formation of smog, ozone depletion and global climate change. Room temperature ILs are found to be unique solvent property whereby it does not evaporate. Besides that, their structure is also bulky and asymmetrical which means the prevention of molecule packing that causes crystallization. Hence, their considered as a replacement for organic solvents in industries.

However, ionic liquids have the potential to harm the aquatic ecosystems through ways including mortality of individual organisms, altered population demographic rates and etc. This is due to the water soluble ILs being widely known only since 1992, hence their impact to the environment are unknown. More studies should be conducted on the toxicity of ionic liquids. There many types of toxicity in ionic liquids such as the cytotoxicity which is the quality of being toxic to cells and etc.

2.3 Selected ionic liquids

Based on the knowledge acquired from analysing the ionic liquids, it's very important to select the best possible samples for research purposes. The samples selected should be off high toxicity and the widely usage of that particular compound and its exposure to the environment. It would also be convenient to obtain samples which have sets of data on the experimental values of the toxicity so that it can be easier to refer for validation. The most common class of cation in ionic liquids are the imidazolium cation, pyridinium and pyrrolidinium. The imidazolium cation base is normally used in metathesis (anion exchange) reactions with compounds containing metal cation. Besides that, the compound is also used as catalyst, solvent electrolytes and etc. However, due to their low volatility makes significant air releases unlikely but releases through water is highly possible from industrial releases.

Most research study has been on the imidazolium based cation in ionic liquids. They have done research on toxicity of imidazolium based ionic liquid on Daphnia Magna, Investigation of toxic effects of imidazolium ionic liquids on marine mussels and etc. Furthermore, data of toxicity from these imidazolium ionic liquids have been recorded in a research done by via the quantitative structure-activity relationship method (Zhao, Zhao et al. 2014). Other cation such as pyridinium is also taken into consideration since the usage of these cation is also widely increasing.

Hence from the research, the five IL's selected are to be optimized once the molecular structures are sketched on the Materials Studio (MS) software. The five molecular structure includes two cation bases pyridinium and imidazolium and two anions which are chloride and bromide. The name of the structures are 1-Methyl-3-octylimidazolium Chloride, 1-Butyl-3-methyl-3-imidazolium bromide, 1-Butylpyridinium bromide, 1-Butylpyridinium chloride and 1-butyl-3-methylimidazolium Hexafluorophosphate.

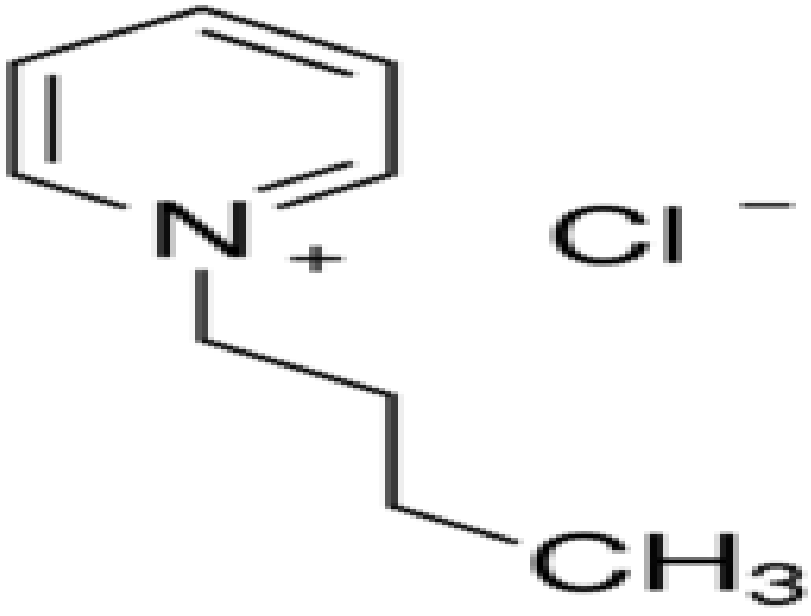


FIGURE 2.3.1. 1-Butylpyridinium Chloride

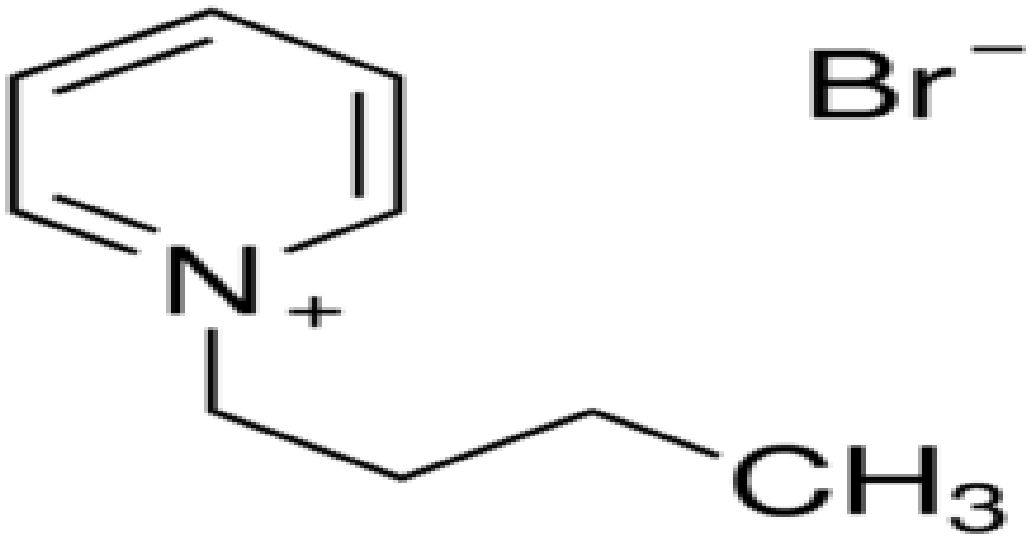


FIGURE 2.3.2. 1-Butylpyridinium Bromide

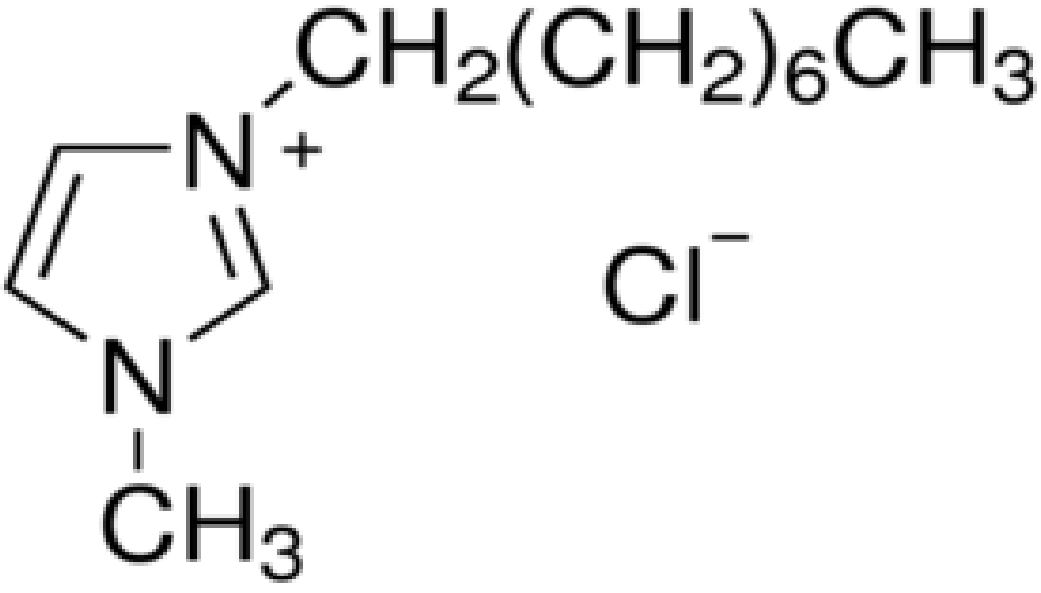


FIGURE 2.3.3. 1-methyl-3-Octylimidazolium Chloride

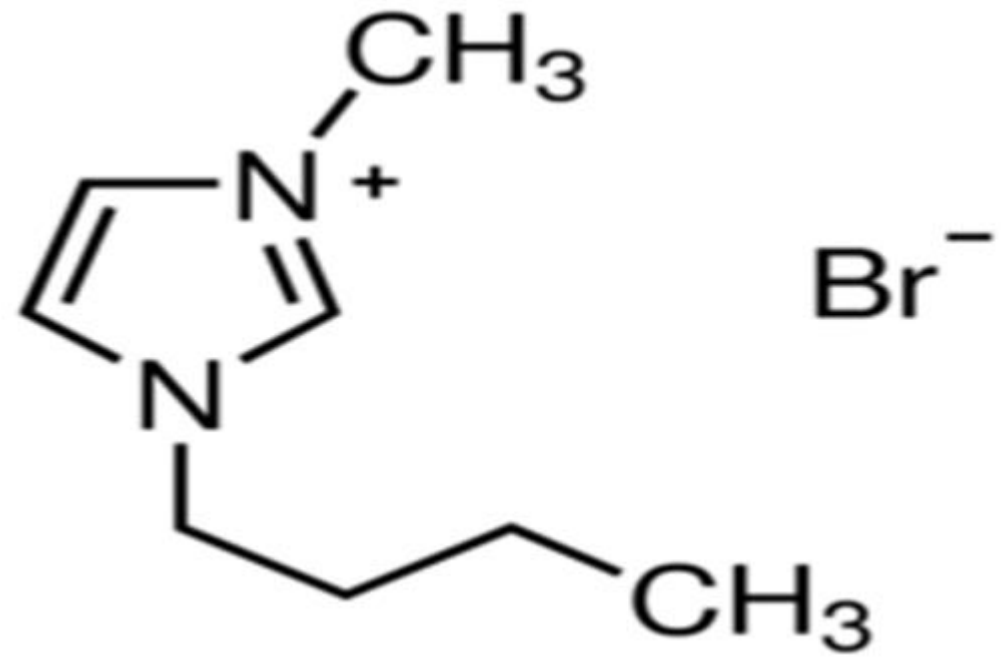


FIGURE 1.3.4. 1-Butyl-3-Methylimidazolium Bromide

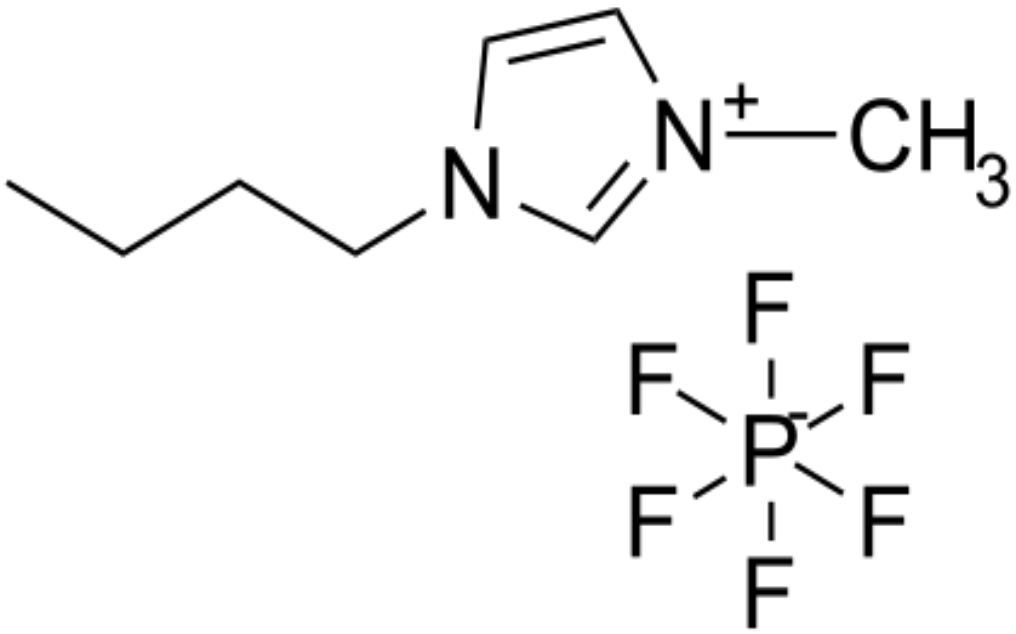


FIGURE 2.3.5. 1-butyl-3-methylimidazolium Hexafluorophosphate

The table below represents the values EC_{50} of the selected samples found in literature:-

TABLE 2.3. EC50 of selected ionic liquids

Chemical	Species	EC_{50}(mg/L)	Reference
1-Methyl-3-octylimidazolium Chloride	Leukaemia rat cell linePC-81	2691.54	(Zhao, Zhao et al. 2014))
1-Butyl-3-methyl-3-imidazolium bromide	Leukaemia rat cell linePC-81	7943.28	(Zhao, Zhao et al. 2014))
1-Butylpyridinium bromide	Leukaemia rat cell linePC-81	5888.44	(Zhao, Zhao et al. 2014))
1-Butylpyridinium chloride	Leukaemia Rat cell linePC-81	100	(Zhao, Zhao et al. 2014))
1-butyl-3-methylimidazolium Hexafluorophosphate	Leukaemia Rat cell linePC-81	118.88	(Zhao, Zhao et al. 2014))

2.4 Toxicity prediction methods

There exist a lot of prediction methods to obtain the toxicity of a certain substance or compound. These methods can either be experimental or through theoretical modelling. The objectives of all these methods are the same which is to find the toxicity of the substance by knowing its Lethal Dosage (LD_{50}) but the limitations are different. Some methods may be too complicated or time consuming to produce the desired results.

2.4.1 Quantitative-Structure Activity Relationship (QSAR)

In 2014, the toxicity of ionic liquid were predicted using the Quantitative Structure-Activity relationship method. Furthermore, Quantitative Structure–Activity relationships (QSAR) model is conducted to predict the toxicities (EC_{50} values) of various ILs toward the Leukaemia rat cell line IPC-81. Four parameters selected by the heuristic method (HM) are used to perform the studies of multiple linear regression (MLR) and support vector machine (SVM).

In recent years, QSAR method was also used to predict the cytotoxicity of ionic liquid model based on SMILES optimal descriptors. In spite of all advantages of ILs,

these compounds probably cause persistent contaminations and constitute environmental risks. Cytotoxicity (Log10 (CE50) data 225 ILs were used in the creation of quantitative structure–activity relationship (QSAR) models by COR relation And Logic (CORAL) software.

Hence from the above observation, it can be said that the usual manner in which toxicity of ionic liquids are predicted using the QSAR method. Using the density functional theory to predict toxicity is said to be more efficient and less time consumption but so far there are very little research have shown density functional theory used for predicting toxicity. Hence, it will be something more of a new

Method to predict toxicity for ionic liquids rather than using the traditional QSAR method. However, the success of predicting toxicity of water soluble arsenicals using the density functional theory was a success and hence it gives me confident that the toxicity can be predicted for ionic liquids. However, the disadvantage of this method will be that it will be expensive because of the labour intensive and time consuming to conduct.

2.4.2 Acute toxicity

Acute toxicity is a method to find the toxicity of a certain substance. It is basically experimenting the exposure to the substance on laboratory animals. They would expose the chemical to the animals and later record their observation and symptoms on the animal. It is time consuming as they would have to wait sometimes for 90 days before the symptoms can take effect. For example, in the research of Acute and chronic toxicity of Imidazolium based ionic liquids on Daphnia Magna stated that the IL-exposure bioassay were conducted as a 48-h static acute tests according to standard procedures. Hence, from this it can be conclude that the tests are run at extremely long hours and plus they would need to sacrifice a lot of animals. Hence, it is definitely much more expensive to conduct. The rule of an acute toxicity is that it should occur within 14 days of the exposure of the substance.

2.4.3 Partial Least Square Discriminant

Since the arrival of the QSAR method, it has been used widely for research purposes on the toxicity of a substance. However, Manuel Alvarez-Guerra and Angel Irabien at the University of Cantabria, in Santander have developed a model to identify

the toxicity of unknown ILs towards Vibrio Fischeri which is a standard bacterial assay for Ionic liquid toxicity. The techniques uses the toxicity data obtained form 64 unknown cation and 30 other known anions that can be combined theoretically and another 1920 unique ionic liquids can be formed. Next, they had applied the partial least squares-discriminant analysis and using toluene as the threshold value, almost all potential hazards of ILs can be predicted. Once their model was compared to a standard set of toxicity data, they were able to confirm the model's accuracy with a test set, obtaining a non-error rate of 93 per cent. However, it was also highlighted that before the model can fulfil its potential, it needs to be tested against other datasets, such as the toxicity data for cells of other species and higher organisms and in other words, it depends on the availability of the data.

Therefore, a table has been tabulated to discuss on the differences between these methods and why is it important to know the differences.

TABLE 2.4. The Advantages and Disadvantages of these prediction methods

Prediction methods	Advantages	Disadvantages
Quantitative Structure Relationship Activity (QSAR)	No sacrifice of animal needed	Time consuming and expensive
Acute Toxicity	Less complicated way to predict toxicity by only using observations and using known concentrations to predict toxicity level	Sacrifice of animals. It also time consuming as the limit for acute toxicity if when it passes 14 days from exposure. It is also expensive due to the labour cost.
Mathematical model developed using partial least square-discriminant analysis	No sacrifice of animal needed	Before the model can fulfil its potential, it needs to be tested against other data sets such as toxicity data for cells of other species and higher organism.

2.5 Quantum Structure Activity Relationship (QSAR)

Quantum Structure Activity Relationship is basically a relationship between the chemical structure of the molecule and its activity. These descriptors are also called toxicity descriptors. It comprises of vibrational frequencies, ionization potential, atomic charges, and electrophilicity and electron affinity of those ionic liquids. These descriptors are obtained through the understanding of Density functional Theory. In a research conducted on the toxicity of Water Soluble Arsenicals using density functional theory, it was found that the structure are optimized for the minimum energy of the Schrodinger equation (Salam, M. (2011). Once the structures are optimized using the Dmol3 program package, the descriptors are calculated.

These quantum structure activity relationship is not only used on chemical related toxicity but also used on the pharmaceutical applications. Usually these Quantum Structure Activity Relationship uses the corresponding descriptors calculated at a semi-empirical PM3 level. However, in a research done on the cyclic imide derivatives of protoporphyrinogen oxidase inhibitors, it was found that the DFT –based QSAR approach produced a much better result and in the future it is expected to help facilitate to obtain toxicity in potentially higher biological activity.

2.5.1 Density functional theory

Density functional theory allows us to have a powerful tool for computation of the quantum state of atoms, molecules and solids, and of ab-initio molecular dynamics (Eschrig 2003). It was discovered during the early foundation of quantum mechanics, in 1927. In the middle of the sixties, Hohenberg, Kohn and Sham on the one hand established a logically rigorous density functional theory of the quantum ground state on the basis of quantum mechanics.

Density functional theory can be built in several versions such as through a theory with particle densities (summed over spin variables) and spin independent external potentials only, irrespective whether the quantum state is polarized or not. Besides that, there is also a theory with spin-up and spin-down densities and external potentials which possibly act differently on spin-up and spin-down particles for collinear polarization situations with one global spin quantization direction and finally it can also be developed through general theory with (spatially diagonal) spin-density matrices and general doubly indexed spin-dependent potentials.

However, the two former cases are considered in parallel throughout by consequently using a combined variable x = (r s) of spatial position r and z-component of spin s. Previously, there were already research carried out using the density functional theory for various reasons including to predict toxicity for water soluble arsenicals and also an assessment of the electronic structure and properties of trichothecene toxins by Michael Appell and Wayne B.Bosma, 2015. The toxicity descriptors are vibrational frequencies, ionization potential, atomic charges, and electrophilicity and electron affinity. With these, the density functional theory is proven to calculate these descriptors and the toxicity.

In order for us to obtain the computational details, the need to understand the theory behind the calculation is important. The number of electrons (N) and v® fix the Hamiltonian of the system as it will create the wave function as well as the related properties. Through research, it has been found that the number of atoms in molecule (N) has been used as a model for physiochemical properties, biological activity (EC_{50}) or toxicity of various molecules.

Hence, the need to identify the descriptors which would be of help in calculating the results is given most importance. Electrophilicity index (w) is a measure of the decrease in energy due to maximal transfer of electron from a donor to an acceptor system as follows (Roy, Parthasarathi et al. 2005):-

$$\omega = \frac{\mu^2}{2\eta}$$

μ And $\boldsymbol{\eta}$ are the chemical potential and hardness, respectively. μ and $\boldsymbol{\eta}$ can be expressed in terms of ionisation potential (I) and electron affinity (A) as follows:

$$\mu = -\left(\frac{\delta E}{\delta N}\right) \approx -\frac{I+A}{2} \qquad \text{and} \qquad \boldsymbol{\eta} = \frac{1}{2}\left(\frac{\partial \mu}{\partial N}\right) \approx \frac{I-A}{2}$$

Using Koopmans's approximation I and A can be expressed in terms of highest occupied (ψ_{HOMO}) and lowest occupied (ψ_{LUMO}) molecular orbital energies as follows:

$$I \approx -\psi_{HOMO}; \qquad A \approx -\psi_{LUMO}.$$

CHAPTER 3

RESEARCH METHODOLOGY

The total energies and the atomic/molecular orbital energies (ψ_{HOMO} and ψ_{LUMO}) of all the five structures mentioned above have been calculated at the basis settings DNP and the functional GGA (PW91) using the Dmol3 program package. The calculations have been conducted for the ionic liquids, namely, 1-butyl-3-methylimidazolium bromide, 1-butylpyridinium bromide, 1-butylpyridinium chloride, 1-methyl-3-octylimidazolium chloride and 1-butyl-3-methylimidazolium hexafluorophosphate. For all of the above structures, optimum geometries have been obtained along with the electron density and the electrostatics using the Dmol3 program package. Electrophilicity index (ω) have been calculated with the help of the ψ_{HOMO} which is the ionisation potential and the ψ_{LUMO} as the electron affinity of the optimised structure. The calculation of the descriptors for 1-butylpyridinium bromide are shown below as example:-

$-\psi_{HOMO}$ = -0.124499 and $-\psi_{LUMO}$ = -0.090385

These values are obtained via the optimization on Materials studio 8.0 Dmol3.

According to Koopmans's approximation;

$$I \approx -\psi_{HOMO}; \qquad A \approx -\psi_{LUMO}$$

Hence,

$$\mu = -\left(\frac{\delta E}{\delta N}\right) \approx -\frac{I+A}{2} = -\frac{(-0.124499)+(-0.090385)}{2} = 0.107442$$

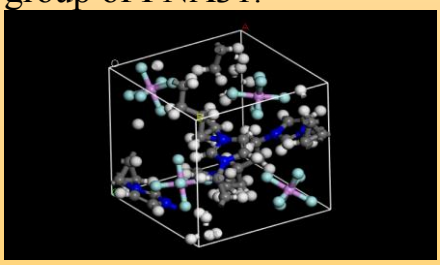
$$\boldsymbol{\eta} = \frac{1}{2}\left(\frac{\partial \mu}{\partial N}\right) \approx \frac{I-A}{2} = \frac{-0.124499-(-0.090385)}{2} = -0.017057$$

$$\omega = \frac{\mu^2}{2\eta} = \frac{0.107442^2}{2(-0.017057)} = -0.33839$$

STEP 1: **Identification** of the 5 most highly toxic IL'S molecular structure.

STEP 2: **Geometric Optimization**.
5 skeleton model sketched on Materials Studio 8.0 using Dmol3 program package to obtain $-\psi_{HOMO}$ and $-\psi_{LUMO}$ & total energy (Et).

- ***STEP 2.1***: The skeleton model of the selected IL's were optimized as a unit cell using the crystal properties with a space group of PNA31.



With a functional setting of GGA, PW91, and basis setting of DNP, basis file 3.5.

FIGURE 3.0 Unit Cell 1-Butyl-3-Methylimidazolium Hexafluorophosphate

STEP 3: **Analysis of computed results**.
Desired descriptors μ, $\boldsymbol{\eta}$, ω and Et.

$$I \approx -\psi_{HOMO}; \quad A \approx -\psi_{LUMO}$$

- $\mu = -\left(\frac{\delta E}{\delta N}\right) \approx -\frac{I+A}{2}$; $\omega = \frac{\mu^2}{2\eta}$
- $\boldsymbol{\eta} = \frac{1}{2}\left(\frac{\partial \mu}{\partial N}\right) \approx \frac{I-A}{2}$

STEP 4: **Validation**
Four parameter regression model is obtained in order for us to correlate the calculated results (EC50) with experimented results (EC50).

FIGURE 3.1. Schematic diagram of the methodology.

3.1 Identification of samples

The identification of the samples are done based on a detailed research. It's important to firstly identify the most toxic ionic liquids and identifying its structure. For this research, the ionic liquids with imidazolium and pyridinium as the cation is selected because according to most research papers, these cations are widely used for industrial applications as well as electrolytes. It has the potential to replace organic solvent in the future. Hence, the impact to environment is still not known and it's important to take these samples and test them. The five samples chosen were 1-butyl-3-methylimidazolium bromide, 1-butylpyridinium bromide, 1-butylpyridinium chloride, 1-methyl-3-octylimidazolium chloride and 1-butyl-3-methylimidazolium hexafluorophosphate.

3.2 Comparison of data obtained and validation

The comparison of the results obtained with the available experimental data in literature. The comparison are basically on the values of the calculated EC_{50} using the density functional theory and the experimental data. Finally, the validation period whereby the values are compared to see if the descriptors have an effect on the prediction of toxicity.

3.3 Gannt chart (FYP 2)

NO	DETAIL / Week	1	2	3	4	5	6	7		8	9	10	11	12	13	14	15	16
1	Sketching of the selected IL's on Materials studio																	
2	Optimization of the skeleton model of the IL's molecular structure.																	
3	Conversion of the Optimized structures to Unit cells using crystal properties									●								
4	Submission of Progress Report										●							
5	Calculating values of the descriptors based on DFT & Developing correlation between the descriptors and the toxicity value											●						
6	Pre-Sedex presentation												●					
7	Submission of Dissertation first draft and technical paper													●				
8	Submission dissertation final draft														●			
9	Viva														●			
10	Submission of Hard bound Copy of project dissertation																	●

● Planned milestone

Process of activity

CHAPTER 4

RESULTS AND DISCUSSION

4.1 Sketching of the skeleton model of the selected ionic liquids.

For this project, the selected ionic liquids must be off high toxicity based on the experimented results obtained via different research papers. Hence, four types of ionic liquids were determined from the research paper done by Zhao, Y., et al. (2014) which had their toxicity values readily available. Our objective is to match these toxicity values obtained via literature with the toxicity values which are calculated based on our methods using the Density Functional theory.

The first step into our methods would be to sketch these skeleton models of the ionic liquids selected in a software called Materials Studio. The version at which will be used is the Materials studio 8.0. As stated before, there are five ionic liquid structures to sketch on materials studio based on the skeleton model of these ionic liquid.

These structures with the imidazolium cation makes a number of ionic liquids with varying properties when it is combined with different anions. For instance, the 1-methyl-3-octylimidazolium chloride as well as the 1-butyl-3-methylimidazolium bromide are crystals at room temperature. Hence, these molecular structures were studied in depth before proceeding with the optimization. Therefore, it is necessary for us to sketch these structures in the software.

The procedure of sketching the 1-butyl-3-methylimidazolium bromide is shown and subsequently the other molecular structures are also sketched according the same procedure.

The procedure are explained as follows:-

1. Firstly, the main structure of the cation is identified and hence the appropriate pattern was selected. With the 1-butyl-3-methylimidazolium bromide, the cation is an imidazolium group having a 5 member ring like structure. Hence, the correct set of ring from the options is selected.
2. Once the option above is selected, sketch the selected ring on the 3D atomistic workspace.
3. Next, it can be observed from the skeleton structure, the ring has double bonds on the third and fifth position of the structure. Hence, modify the bond type in the options but only on those two positions.
4. So far, these structures are built only with carbon atoms since those are the basis of almost all molecular structures. However, there are presents of nitrogen atoms are the positions 1 and 3 and the nitrogen atom at the position 3 has an oxidation state of +1. Next, in cooperate these nitrogen atom into our structure.

   Once these properties have been selected, now apply them to the structure and as a reference to nitrogen atom, the particular area at which there are presents of nitrogen turns blue. Next, the nitrogen atoms have been placed at the correct position in the structure.
5. Insert the carbon atoms into the structure. There are carbon atoms from the nitrogen atom with an oxidation state of +1 and also from the nitrogen atom from position 1.

   The structure above is almost 80% complete. Now, add the anion into the structure. As stated above, this molecular structure has bromide as the anion. Hence, from the options of the periodic table, select the bromide ion from group 17.
6. In order for us to differentiate between each atom, the bromide ion is highlighted in red.
7. Now, an almost complete structure of 1-butyl-3-methylimidazolium bromide is obtained. The next step is to add the hydrogen atoms to the structure. The display style is also changed in order to give us a more detailed view of the molecular structure.

8. Next adjust the hydrogen in the settings by clicking on the icon “H”:-After adjusting the hydrogen, also “Clean” the structure so that it can be arranged at the most optimum positions for calculations in the future.

The following are the structures sketched on MS software:-

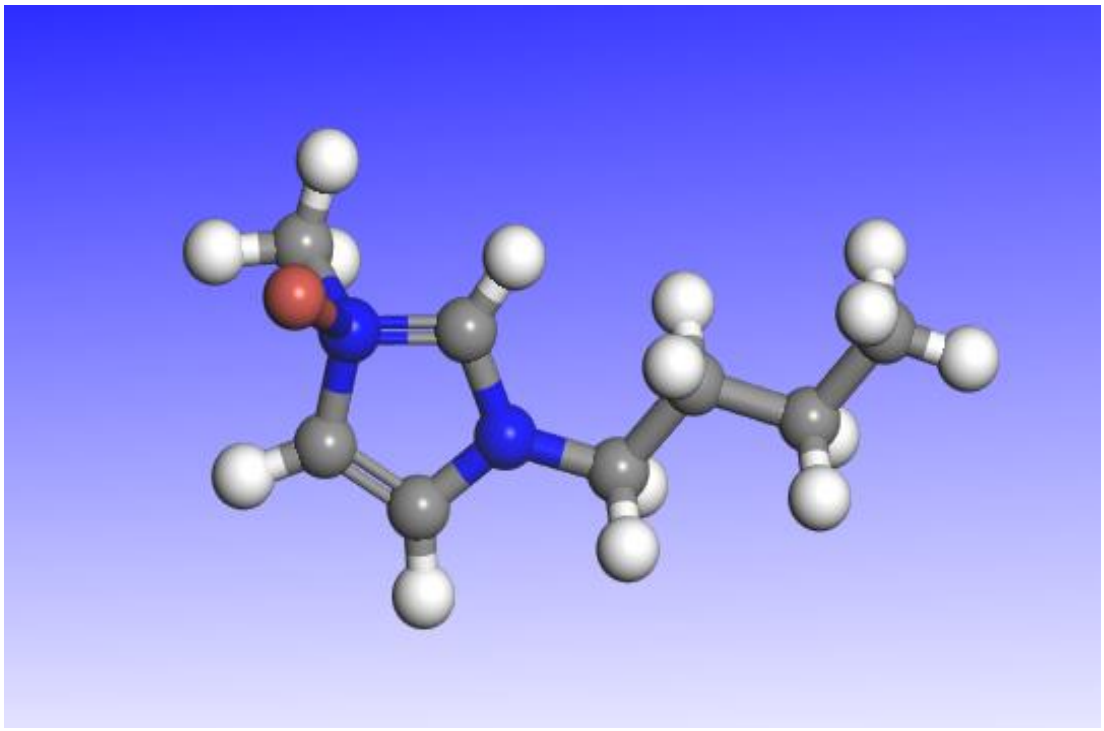


FIGURE 4.1.1. 1-Butyl-3-Methylimidazolium Bromide

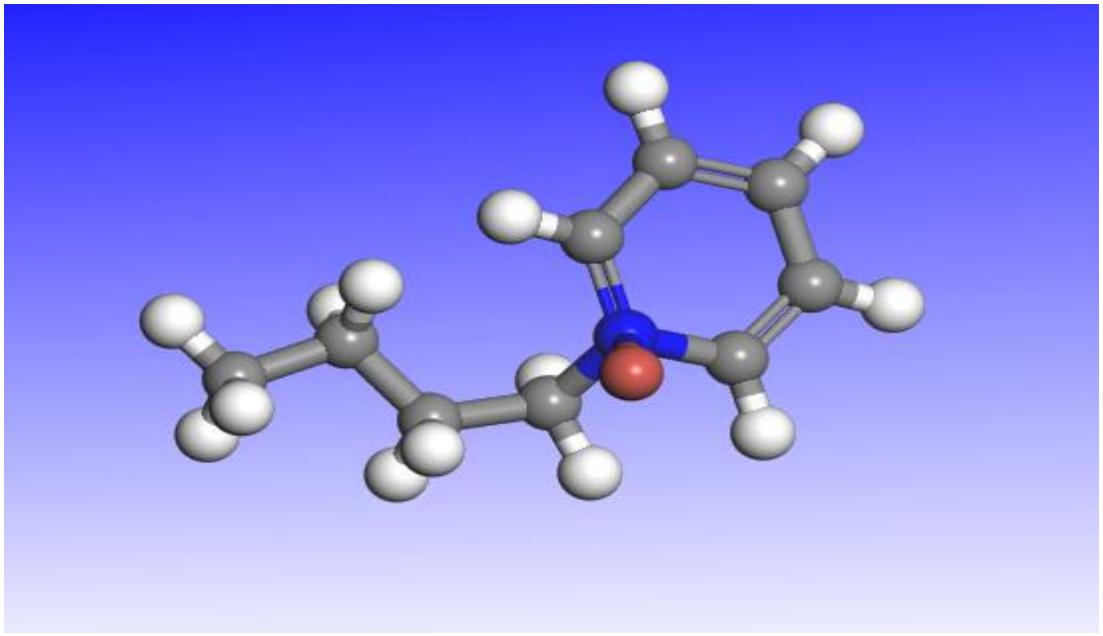


FIGURE 4.1.2. 1-Butylpyridinium Bromide

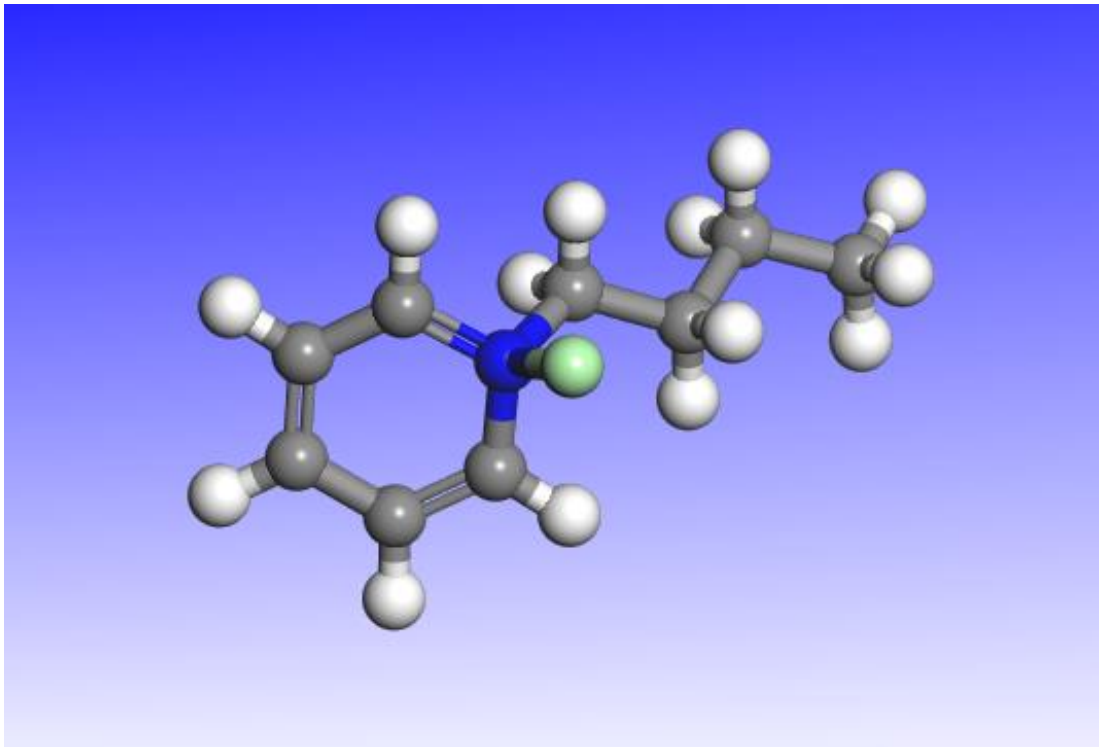


FIGURE 4.1.3. 1-Butylpyridinium Chloride

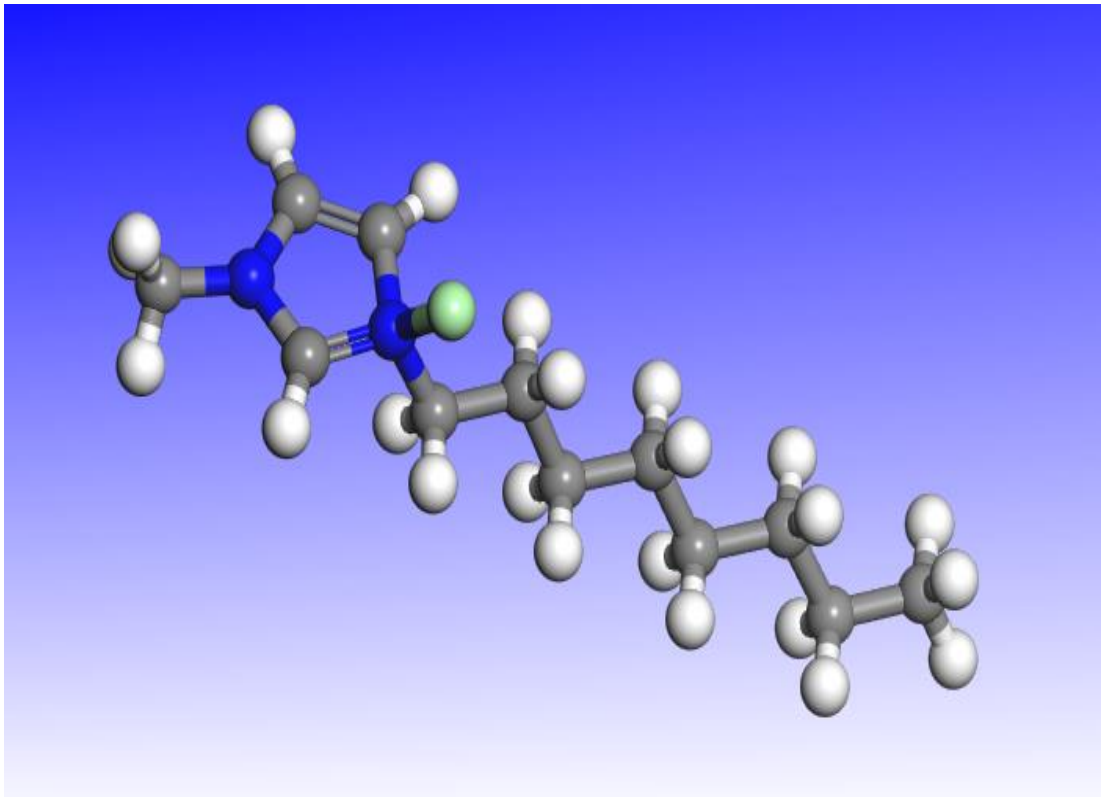


FIGURE 4.1.4. 1-methyl-3-Octylimidazolium Chloride

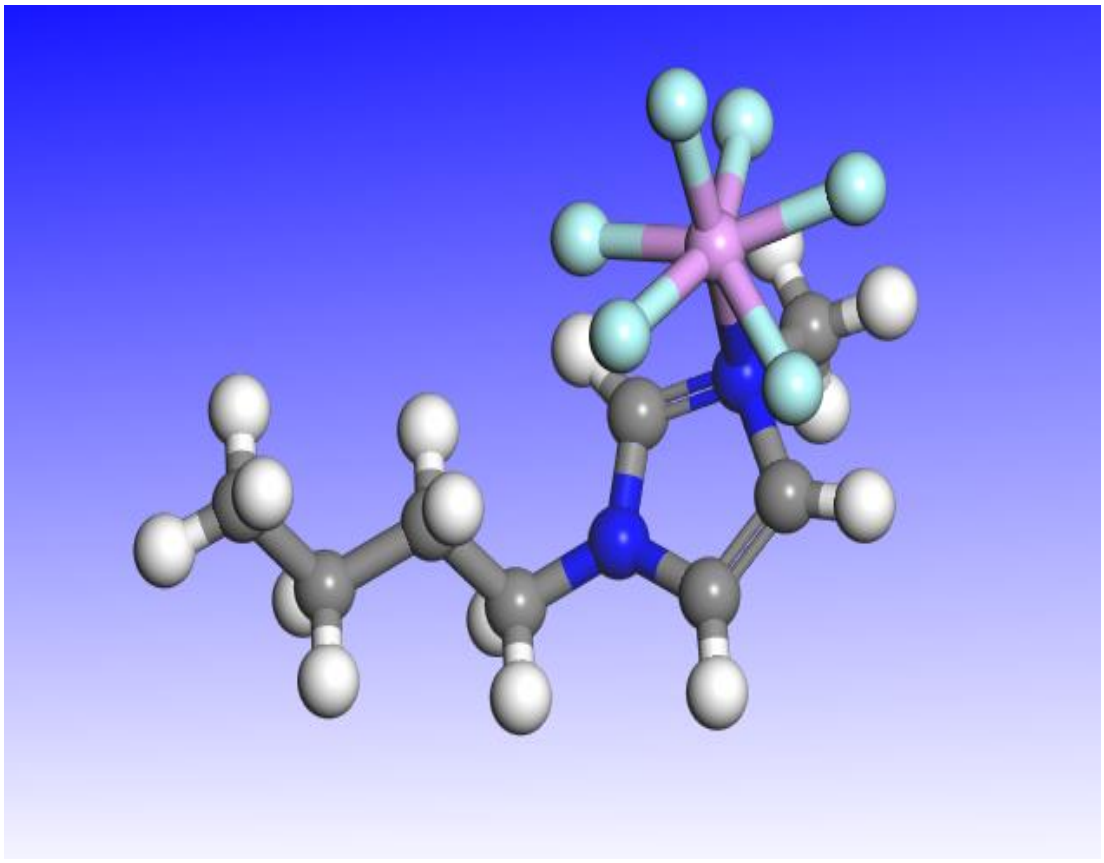


FIGURE 4.1.5. 1-butyl-3-methylimidazolium hexafluorophosphate

4.2 Geometric optimization

i. 1-butyl-3-methylimidazolium bromide: Optimized the structure above using the crystal systems since these ionic liquids are known as molten salts and hence can be considered a crystal. The space group PNA31 which is orthorhombic. From the optimization, the value for the ψ_{HOMO} and ψ_{LUMO} which are -0.16207 and -0.07442 respectively. The mulliken atomic charges are -0.043825. The total energy, E_T is -2997.7399058 Ha.

ii. 1-butylpyridinium bromide: This structure is also optimized using the crystal systems. The space group is also similar to the structure above which is PNA31. From the calculations, it was found that ψ_{HOMO} is -0.124499 and the ψ_{LUMO} is -0.090385. On the other hand, the mulliken atomic charges are based on the hardness, $\boldsymbol{\eta}$, are -0.017057. Lastly, the total energy of the optimized structure is -2890.483402 Ha.

iii. 1-butylpyridinium chloride: From the optimization of these structure using the crystal system as well and a space group of PNA31, obtained the ψ_{HOMO} and the ψ_{LUMO} values which are -0.141248 and -0.106585 respectively. The $\boldsymbol{\eta}$ values are -0.0173315. The total energy is -920.7142112 Ha.

iv. 1-methyl-3-octylimidazolium chloride: The geometric optimization of this structure was also based on the crystal structures. The space group is also PNA31. The subsequent value of ψ_{HOMO} is -0.172785 and the value of ψ_{LUMO} is -0.035845. The mulliken atomic charges are -0.06847. Lastly, the total energy of the optimized structure is -867.9049714 Ha.

v. 1-butyl-3-methylimidazolium hexafluorophosphate: Using the crystal-like system and through research, finding its space group to be PNA31, the structure was optimized. The value of ψ_{HOMO} is -0.281728 and the value of ψ_{LUMO} are -0.129627. The atomic charges are -0.152099 and the total energy is -1231.5920913 Ha.

The results were tabulated as follows:-

TABLE 4.2. Values of the descriptors obtained from the optimization.

IL's	I, ψ_{HOMO}	A, $-\psi_{LUMO}$	η	μ	ω	Et (Ha)	EC_{50}(mg/L)
1-butyl-3-methylimidazolium bromide	-0.16207	-0.07442	-0.043825	0.118245	-0.15952	-2997.74	2691.54
1-butylpyridinium bromide	-0.124499	-0.090385	-0.017057	0.107442	-0.33839	-2890.48	7943.28
1-butylpyridinium chloride	-0.141248	-0.106585	-0.017332	0.1239165	-0.44299	-920.714	5888.44
1-methyl-3-octylimidazolium chloride	-0.172785	-0.035845	-0.06847	0.104315	-0.07946	-867.905	100
1-butyl-3-methylimidazolium hexafluorophosphate	-0.281728	-0.129627	-0.152099	0.2056765	-0.13906	-1231.59	118.88

4.3 Regression model analysis

After obtaining the values for the descriptors above, the next step would be to correlate them in order to obtain an equation that could calculate the toxicity of the ionic liquids. Hence, a regression model analysis was carried out on the descriptors so that a correlation can be obtained. In this case, the general regression model equation used are:-

$$\mathbf{Y' = a + bx_1 + cx_2 + dx_3 + ex_4}$$

Where a is known as the constant or the Y-intercept, b,c,d and e are the coefficients that are required to calculate, x_1, x_2, x_3, and x_4 are E_T(Ha), $\boldsymbol{\eta}$, μ, and ω respectively which are also known as the independent variables. The dependent variable would be the Experimented values of EC50 obtained via literature.

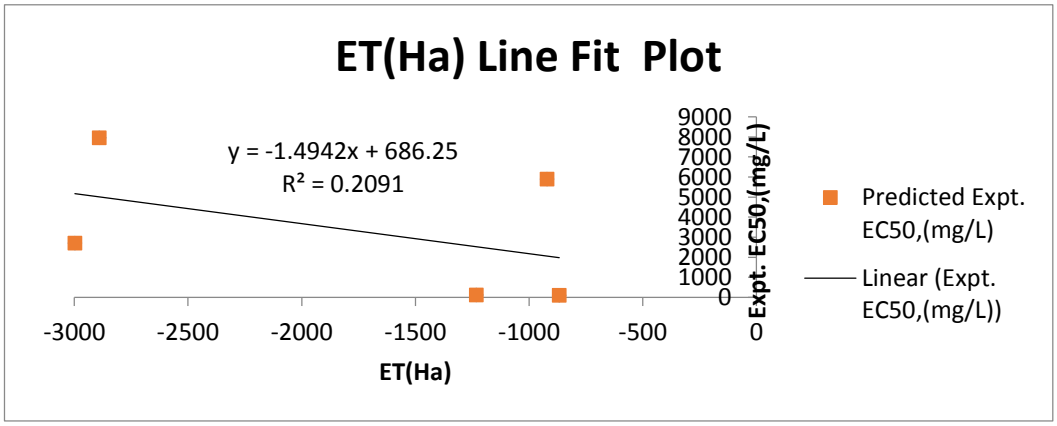


FIGURE 4.3.1. Regression for Experimented toxicity EC50 values versus Total energy (Et).

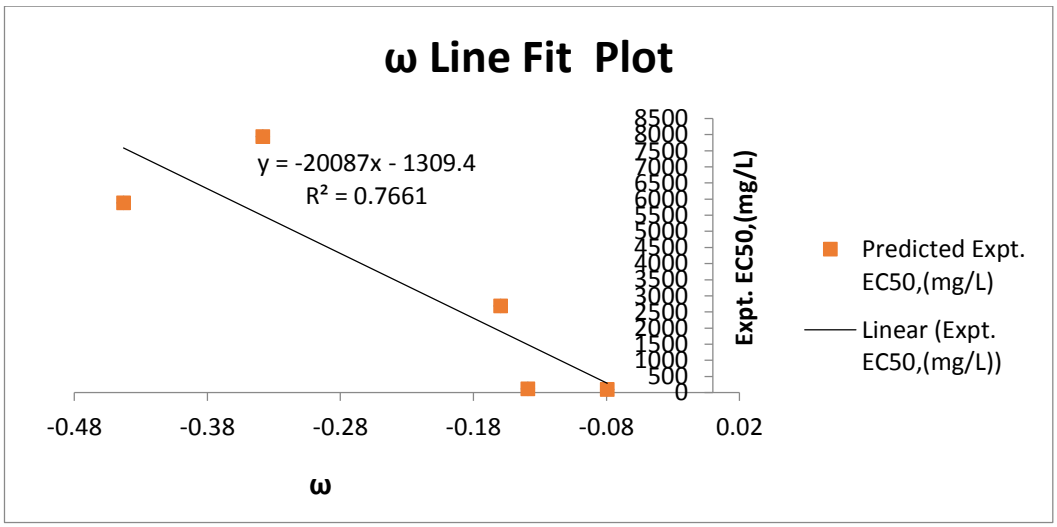


FIGURE 4.3.2. Graph of Electrophilicity index versus Experiment values of EC50 obtained via literature.

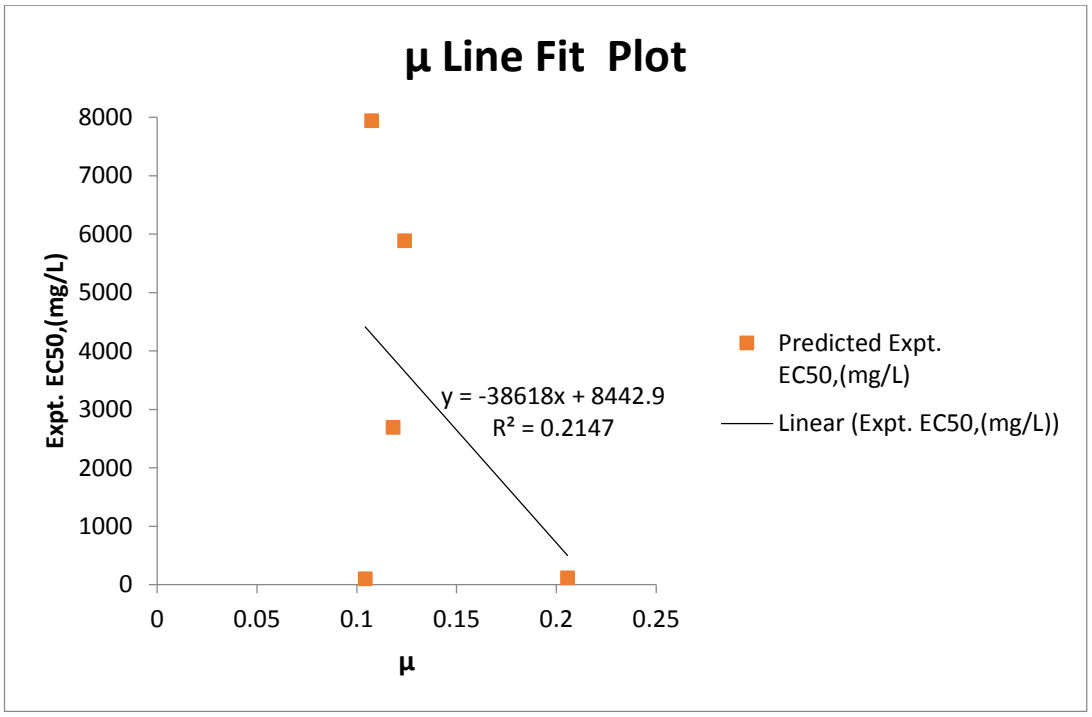


FIGURE 4.3.3. Graph of Chemical Potential Versus Experiment values of EC50 obtained via literature.

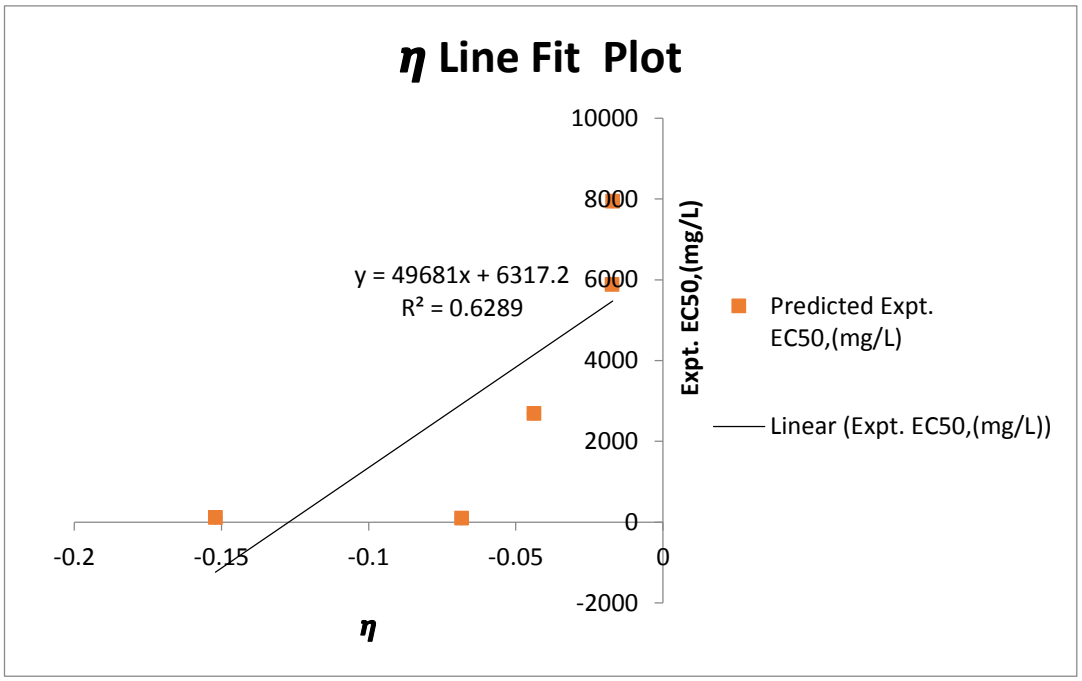


FIGURE 4.3.4. Graph of Hardness versus Experimented values of EC50 obtained via literature.

As it can observed from the graphs obtained above, the correlation between electrophilicity index (ω) and the toxicity values are reasonable high which is 0.7661. Besides that, it can also be observed that the correlation between hardness (***η***) and the toxicity also produced a reasonably high value which is 0.6289. Hence, it can be said that the descriptors of ω and ***η*** have good relationship with the toxicity values. However, in our calculation, a four parameter regression model including all four descriptors ω, ***η***, **μ** and E_t were performed.

$$\text{Calc.EC}_{50} = -2.105 \times Et - 132935.408 \times \eta - 141715.595 \times \mu - 41957.548 \times \omega + 620.330$$

TABLE 1.3. Calculated EC50 values versus Experimental values of EC50 obtained via literature.

Calc.EC50,(mg/L)	Expt. EC50,(mg/L)
2693.34	2691.54
7944.48	7943.28
5888.43	5888.44
100.25	100
119.28	118.88

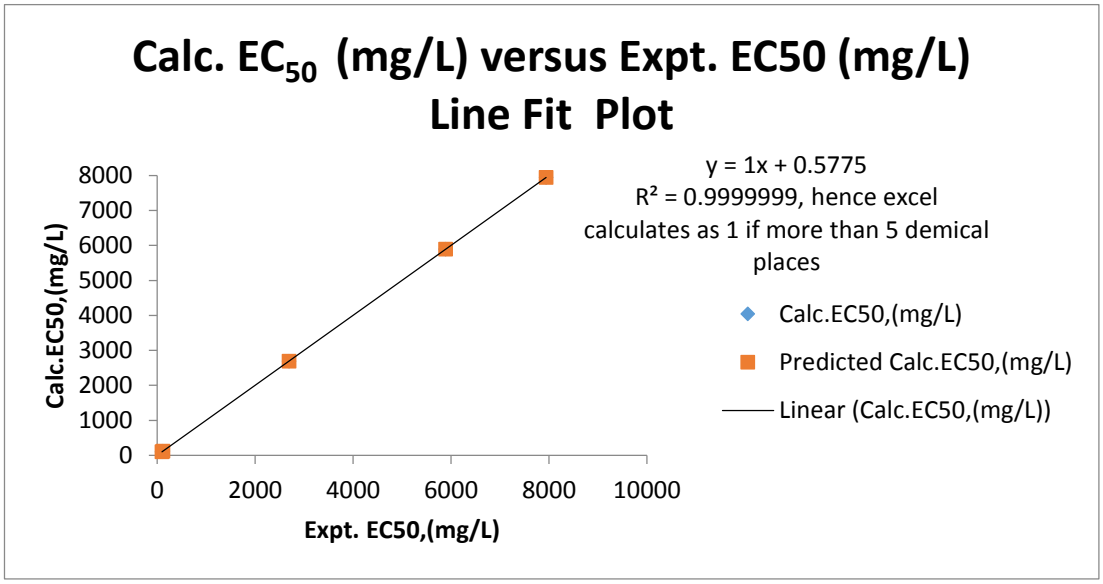


FIGURE 4.3.5. Graph of Calculated EC50 versus Experimental values of EC50 obtained from literature

As mentioned earlier, a correlation of the descriptors, ω, $\boldsymbol{\eta}$, $\boldsymbol{\mu}$ and E_t with the experimental values of EC_{50} obtained via literature which uses the Acute toxicity method on leukaemia rat cell line is carried out. Hence, the objective would be to match these values obtained via literature to the predicted EC_{50} values using the regression analysis involving the mentioned descriptors. If the correlation is positive and gives us a high value, it can be said that the regression model equation can predict the toxicity of ionic liquids. However, bearing in mind that a regression model analysis needs almost 100 samples to prove of its true correlation and the feasibility of the regression model. Due to time constraint, only five highly toxic ionic liquids were selected for our study. With these five samples, a correlation will be obtained via the regression analysis.

There are basically four parameters which are ω, $\boldsymbol{\eta}$, $\boldsymbol{\mu}$ and E_t. Hardness is basically a measure of the resistant solid matter to change shape when a force is applied. Hence, these could be a good descriptors for us to predict the toxicity. Chemical potential, $\boldsymbol{\mu}$, is the form of potential energy that is absorbed or released during a chemical reaction. These parameters were correlated with the experimented values obtained via literature, Expt. EC_{50}. Four linear plots were obtained and their correlations were also given. From figure 4.3.1, we could observe that the relation between the total energy, E_t and the Expt. EC_{50} did not produce a good results. Their correlation are observed to be $R^2 = 0.2091$. However, these descriptors are included in our parameter regression model. Besides that, the correlation between the chemical potential, $\boldsymbol{\mu}$ and the experimented values of EC_{50} obtained via literature Expt. EC_{50} also did not produce a good results. It give a correlation of only $R^2 = 0.2147$. These descriptor will be included in the regression model as well although having low correlation. This could be due to its effect on the toxicity is low. Chemical potential probably gave a low reading due no reaction because chemical potential was defined to a potential energy released only during a chemical reaction. However, the descriptors ω, and $\boldsymbol{\eta}$ gave good correlation of $R^2 = 0.7661$ and $R^2 = 0.6289$ respectively.

After obtaining these values, a correlation was performed between these descriptors being the independent variable, x and the Expt. EC_{50} to be the dependent variable, y.

Next, excel had calculated a correlation of $R^2 = 0.9999999 \approx 1$ (excel calculates as 1 if more than 5 decimal places) between the calculated EC_{50} value and the Expt. EC_{50} values.

Hence, the regression model equation obtained via the regression analysis proves to be a good equation that can be used to predict toxicity of ionic liquids.

CHAPTER 5

CONCLUSION AND RECOMMENDATION

5.1 Conclusion

The purpose to carry out this project was to enable us to meet our objectives and to produce good results so that it can prove further of the theory if it's functional or dysfunctional. A good set of samples was first selected in order for us to continue to the next level of sketching these structures on the MS software. Once the sketching was complete, these five samples were optimized to a unit cell following the properties of a crystal system since these ionic liquids have somewhat similar structure due to them being a molten salt. After optimization, a set of $-\psi_{HOMO}$ and $-\psi_{LUMO}$ values were obtained from each IL's. These values were then fitted into the equations stated by Paar and co on the relationship of the electrophilicity index and using koopman's approximation to get the values of the other descriptors. Using the regression analysis method, a correlation was formed between the descriptors which acts as the independent variables, x and experimented values of EC_{50} obtained via literature.
With these correlations being formed, a four parameter regression model equation was developed.
Regression model:-

$$\text{Calc.}EC_{50} = -2.105 \times Et - 132935.408 \times \eta - 141715.595 \times \mu - 41957.548 \times \omega + 620.330 \quad (1)$$

These equation basically gives the values of the toxicity of ionic liquids if the descriptors are known. After carefully analysing the results, it can be said that the correlation obtained between the predicted values, Calc. EC_{50} and the existing experimented values of EC_{50} is high with almost a value of $R^2 = 0.9999999 \approx 1$. In a nutshell, the regression model equation obtained above can somewhat be a good predictor for toxicity of ionic liquids. Besides that, all of the objectives above are met. The sample is identified, then a correlation is obtained between descriptors and the existing EC_{50} values and finally the regression model equation is obtained.

5.2 Recommendations

As a future recommendation, with more time, a lot more structures can be optimized and probably obtain more samples. With these large sample group, to develop a regression model and compare to see if the correlations are still high is even more accurate. Hence, a more solid foundation to the equation can be obtained and further prove of the accuracy of the Quantum Structure Activity Relationship (QSAR).

Besides that, a different set of descriptors in the future can be used such as the charge transfer, N and etc. to correlate them to the toxicity of ionic liquids.

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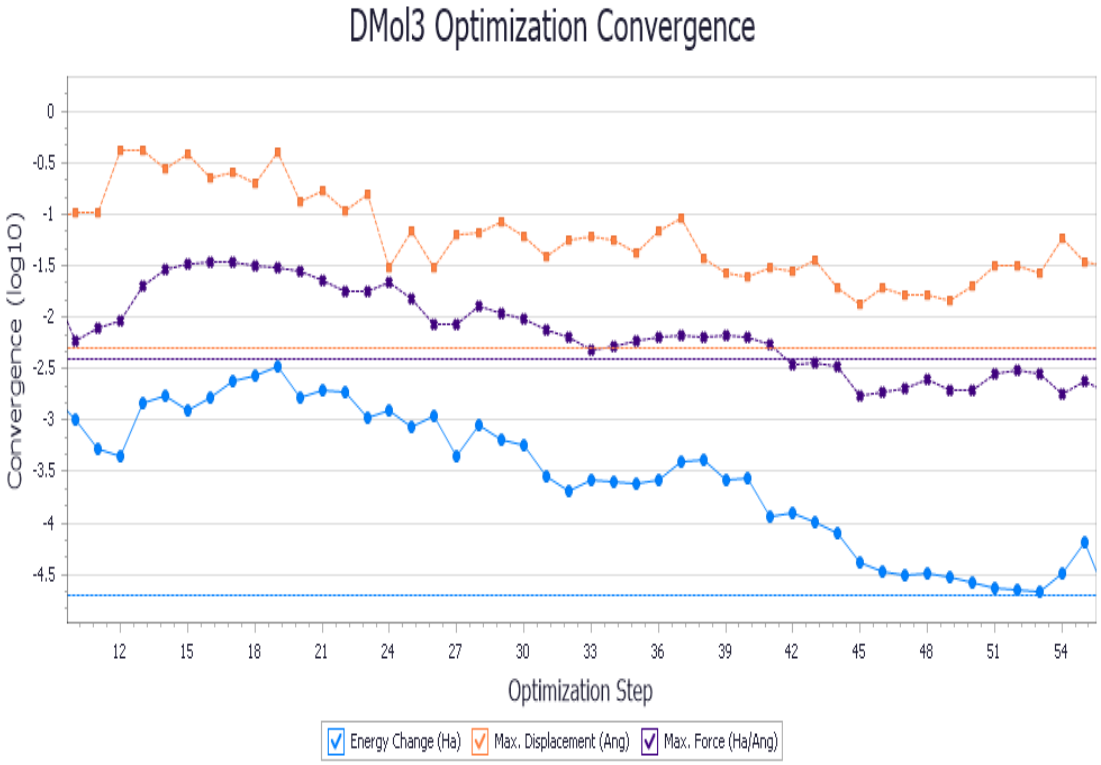
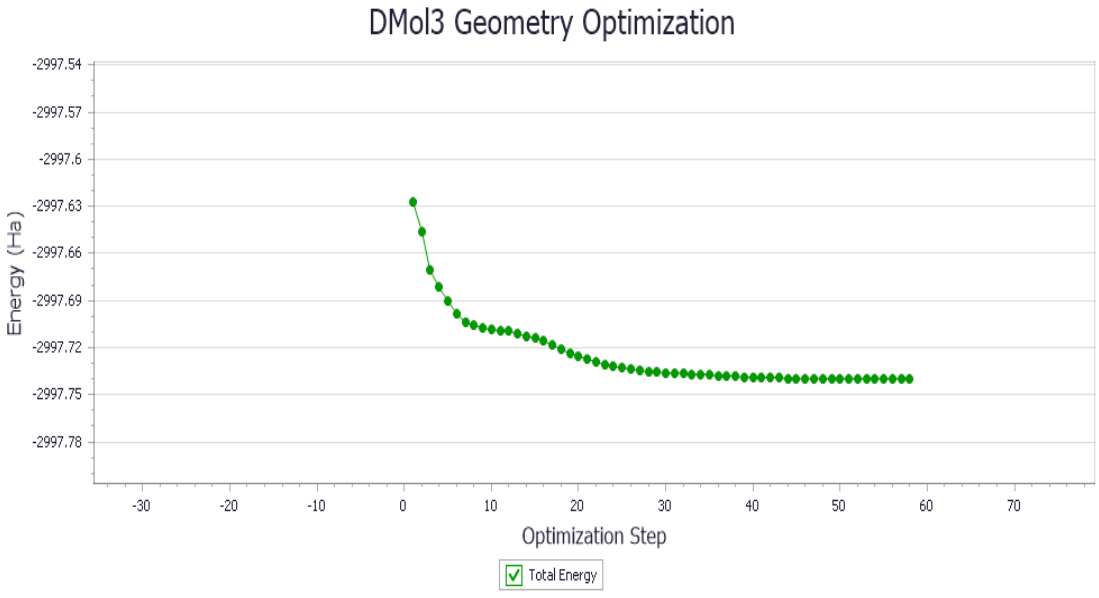
APPENDICES

a. General Regression Equation

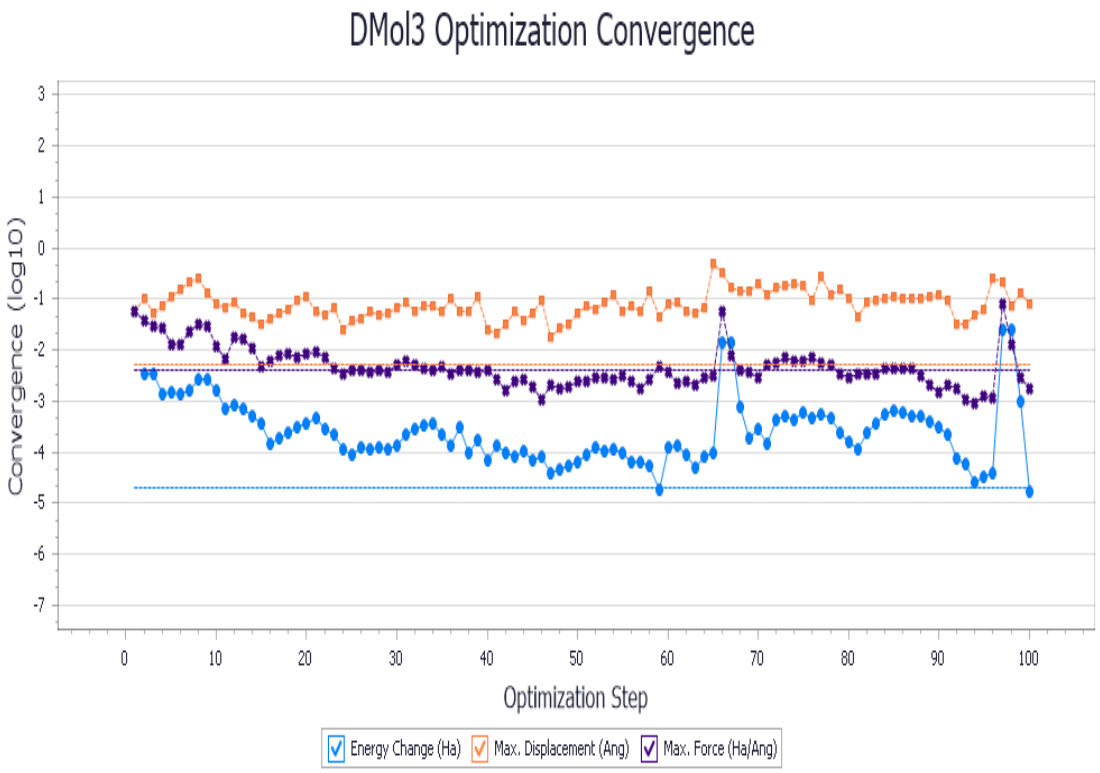
$Y' = a + bx_1 + cx_2 + dx_3 + ex_4$

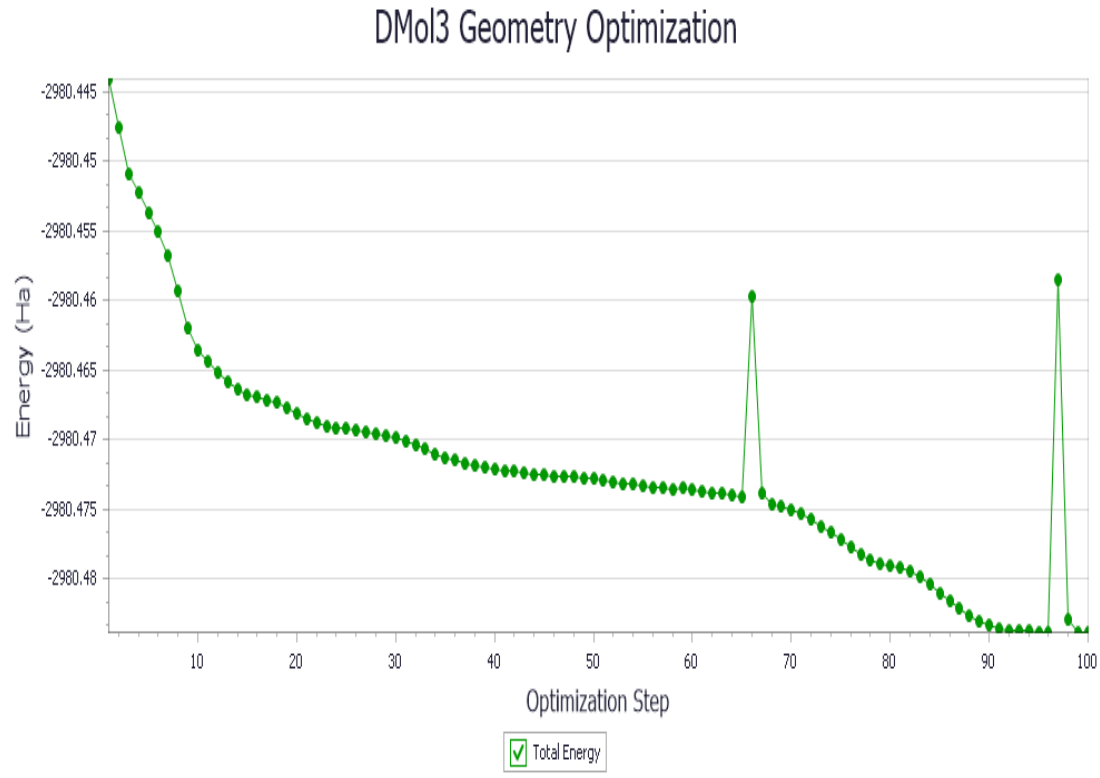
b. Geometric optimization graph of 1-Butyl-3-methyl-3-imidazolium bromide



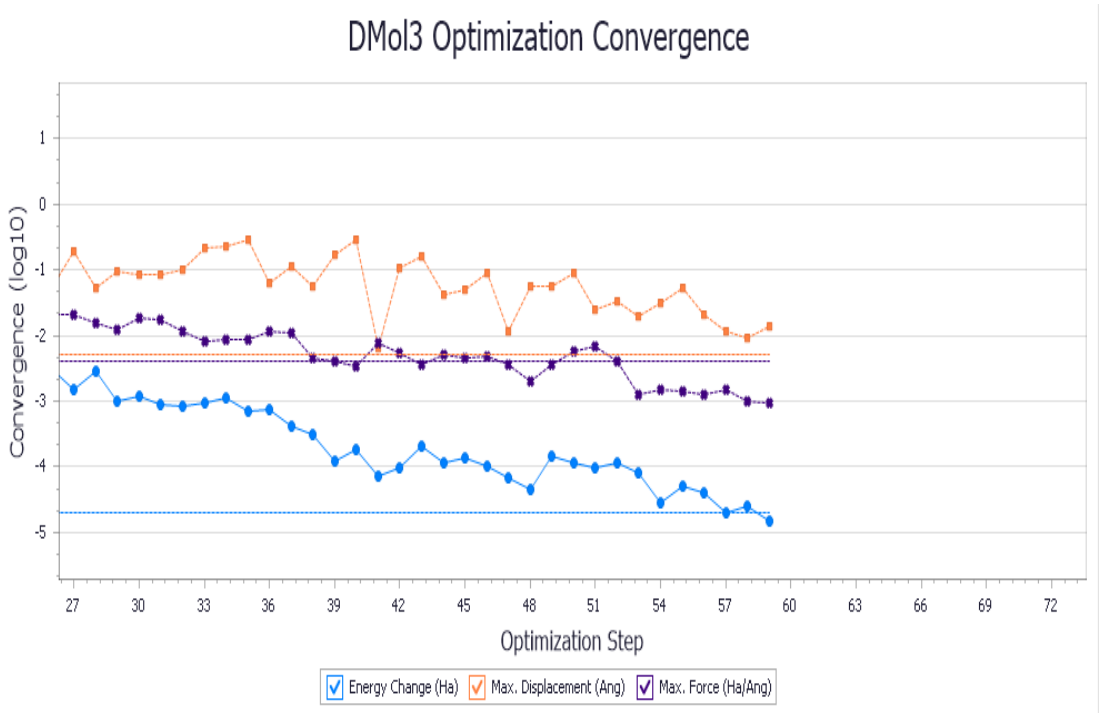


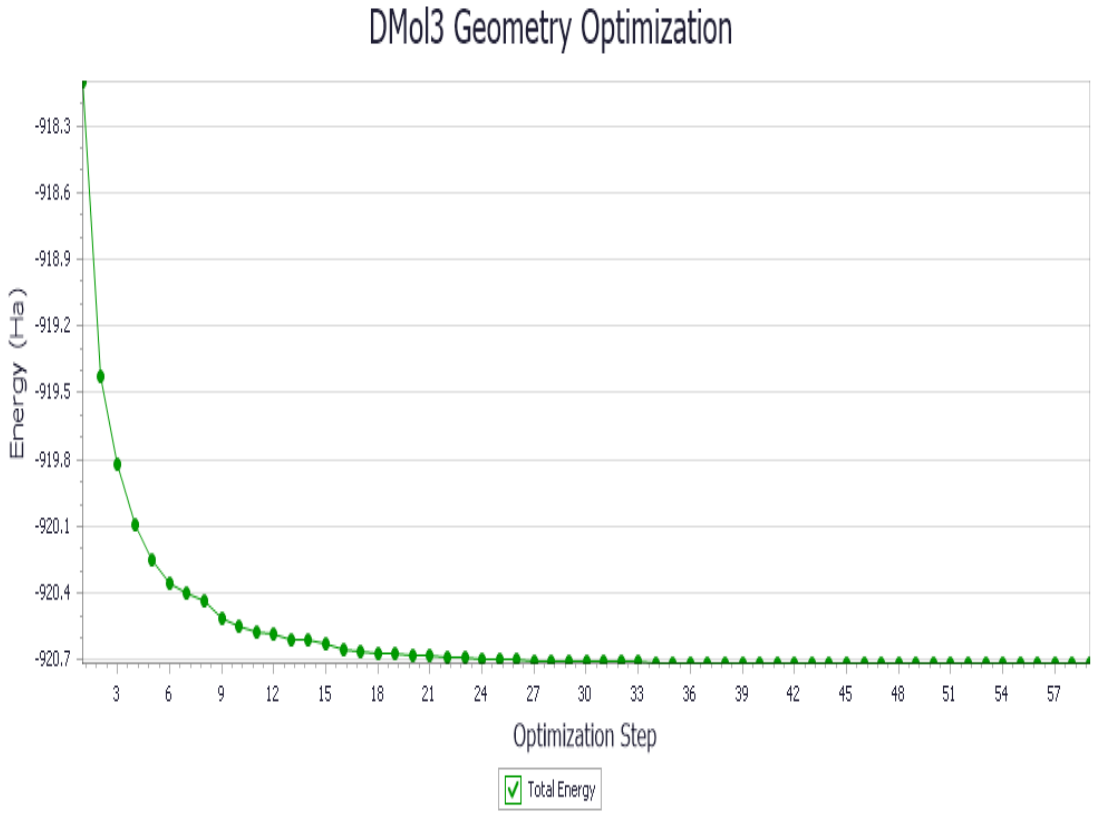
c. Geometric optimization graph of 1-butylpyridinium bromide



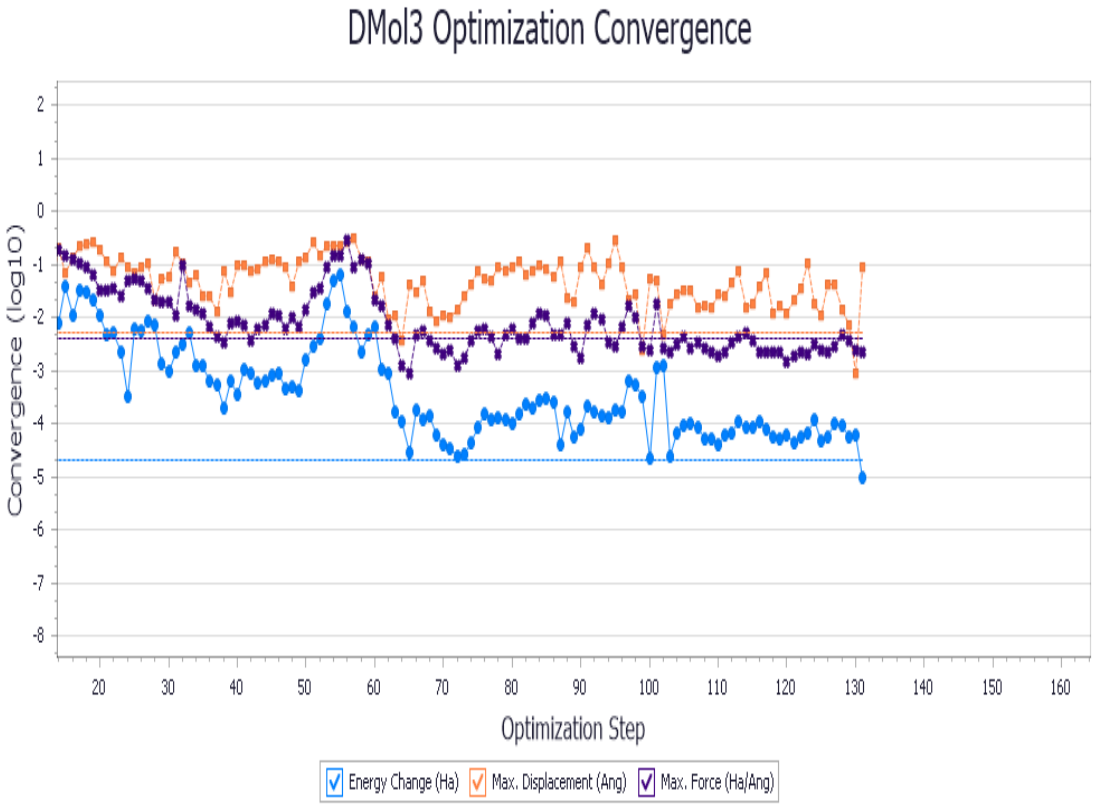


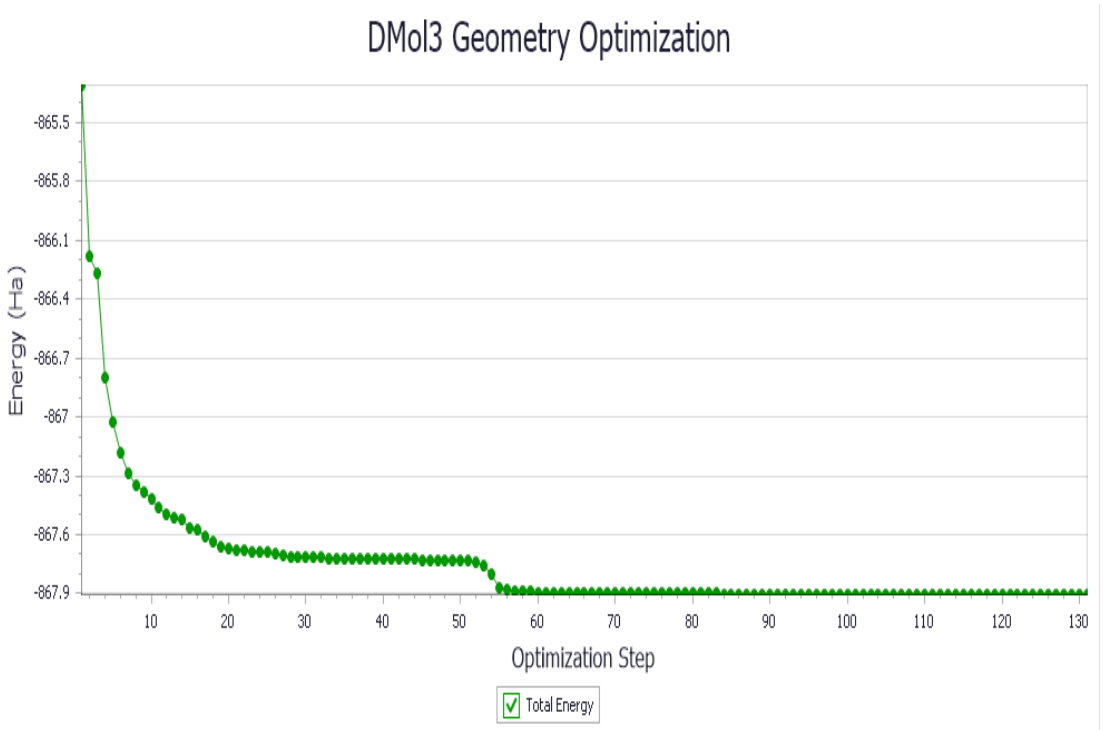
d. Geometric optimization graph of 1-butylpyridinium chloride





e. Geometric optimization graph of 1-Methyl-3-octylimidazolium Chloride





f. Geometric optimization graph of 1-butyl-3-methylimidazolium hexafluorophosphate