# Synthesis and characterization of ammonium ionic liquids and their antimicrobial and computational overview

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## Abstract
The ammonium acetate and formate ionic liquids (ILs) were synthesized in the neutralization reaction with different cations of alkanol amine and alkyl amine with formic and acetic acid as an anion. Thin layer Chromatography monitored the reaction and the synthesized six ILs was characterization by FT-IR spectroscopy and $^{1}H$, $^{13}C$ Nuclear Magnetic Resonance (NMR) spectroscopy. The spectroscopic analysis of the synthesized ILs gives the excellent agreement for the confirmation of their synthesis and purity. These compounds were tested in computing program of computational chemistry on two basis sets, Mm+ and PM3 for the calculation using molecular mechanics by Hyperchem$^{R}$ Release 8.0 (Hypercube, Inc.). Single point calculations were carried out to evaluate the data profile on the physical properties, such as total energy, binding energy, hydration energy, dipole moment, heat of formation and energies for HOMO and LUMO. Furthermore, the synthesized ILs was screening antimicrobial activity against both of gram positive and gram negative bacteria such as *Bacillus cereus*, *Staphylococcus aureus*, *Escherichia coli*, *Salmonella typhi*, *Pseudomonas aeroginosa* and *Shigella dysenteriae* and two phytopathogenic fungi such as *Aspergillus niger* and *Rhizoplus azzahra* to establish as a green and environmentally benign solvent.

Keywords: Ethanolamine, FT-IR, NMR, HyperchemR, Antimicrobial activity.

## 1. INTRODUCTION
Ionic Liquids (ILs) was introduced almost a century ago by the observation of Paul Walden in a neutralization reaction of ethanolammonium nitrate [1]. They belong to a novel class of low temperature (typically <100°C) molten salts, consisted of discrete anions and cations [2]. They exhibit, in most cases, relatively low viscosities, thermally stabilities, high thermal conductivities, a large electrochemical window, highly polar and nonvolatility [3-4]. Millions of ILs can be synthesized and characterized with a variety of cations and anions in the theoretical study which is also known as the designer solvent due to their tunable physical and chemical properties [5]. Due to their unique properties [6], ILs have become one of the most important research targets in chemistry, physics, engineering and material science and technology [7].

There is a strong interest in ILs as alternatives for volatile organic solvents in the organic synthesis [8-9].They can act as solvents for chemical reactions, including catalytic reactions [10-14]. ILs are found as the use in electrochemical applications [8-9, 15-16], e.g. electrolytes in batteries [17], absorption of carbon dioxide[18], in photovoltaic devices [19], and a medium for electrodeposition of metals. ILs can also find applications in separation sciences as solvents for in depolymerization process of lignin[20], extraction processes [21], as a stationary phase for gas chromatography [22], as well as in mass spectrometry [23]. To explain the Molecular interactions between ammonium-based ionic liquids and molecular solvents, the thermodynamic parameters can be effectively used to gain valuable insights which would be most useful in various industries[24]. In other hands the fascinating physio-chemical properties of alkanolammonium based ILs including ion conducting, viscous, non-volatile and non-flammable, they are now used in various fields both of academia and industries as a solvent, catalyst, reaction media, pharmaceutical ingredient and solvent, polymerization reaction [25]. In addition, the biodegradability of ammonium cations is considerably enhanced when hydroxyl groups are incorporated. To make in utilizing as the green and promising eco-friendly solvents in the separation process, ILs can serve as nonvolatile entertainers to break zoetrope and enable a more energy efficient. [26]. Ammonium-based ILs especially ethanolammonium ILs are easy to prepare and the raw materials are mostly available with cheapness. To investigate, the study of alkanolammonium based ILs, three cations such as different types of a cation such as ethanolamine, diethylamine and triethanolamine were used to form ILs with anion as formate and acetate. To ensure their use as a solvent, the physiochemical properties are the greatest goal. Estimation the physio-chemical properties are a process of the time consuming and waste of chemical materials. To save the time and chemicals, the computer programming e.g. Hyperchem$^{R}$ Release 8.0 (Hypercube, Inc.) was used to calculated the total energy, binding energy, Heat of formation , HOMO, LUMO, LUMO~HOMO and different vibrational stretching of bonds [27-29]. Different cations and anions were used to make a comparison effects in physio-chemical properties through computer programming and antimicrobial toxicity. Most of these are highly polar and well soluble in water. So there is a large scope to pollute water body and environment [30].The antimicrobial toxicological profile of these ILs

was not well documented and the information are not enough for further applications. Due to the short lifespan, bacteria and fungi are the best starting point to investigate and estimate the toxic bio-data of synthesized formate and acetate ammonium ILs. The bacterial toxicity level in the form of antimicrobial activity against gram positive and gram negative bacteria was screened and evaluated for confirming their use in specific fields [31-32].

## 2. EXPERIMENT

### 2.1 Materials and reagents

All the chemicals were of research grade and used without further purification unless otherwise stated. All the solvent were obtained upon distillation before use. Formic acid (Merck KGaA), Acetic acid (Merck KGaA), 2-aminoethanol (Merck KGaA), Ethylamine (Merck KGaA), Triethanolamine (Merck KGaA) was analytical grade commercial products. Thin Layer chromatography powder (Merck KGaA), standard antibiotic Gentamycin were purchased for the reaction workup FT-IR spectrophotometer, SHIMADZU, Japan, range 600 -4500 cm-1) was used with KBr disc technique. The synthesis, computer programming and the characterization were done at the department of chemistry in University of Chittagong, and Chittagong, Bangladesh. The antimicrobial activity was at Department of Microbiology in University of Chittagong, and Chittagong, Bangladesh. The $^{1}H$, $^{13}C$ NMR Spectroscopy was done at Iwate University, Japan.

### 2.2 Ionic Liquids Synthesis and Purification

The synthesis of alkanol or alkyl ammonium ILs consists in an acid-basic neutralization reaction[33]. The base, in this case, ethanolamine, diethylamine and triethanolamine were added under stirring in a slow dropwise about 20-25 minute maintaining the temperature using ice bath from exothermic heat release on a glass flask with the acid (formic and acetic acid respectively).

Then the mixture was stirred for 24 hours at room temperature, to obtain a viscous clear liquid. The reaction was monitored by Thin layer chromatography (TLC).The reaction products are an ester and a salt of ethanolamine [33-35] (The ILs purification process consists in a strong agitation and slight heating, at 323.15K, for the vaporization of impurities (residual non reacted and water) under vacuum of 20 kPa. Humidity below 0.1% was obtained after this purification process[33, 35] (Alvarez et al., 2010; Iglesias et al. 2010), and the liquids presented a limpid and viscous appearance. The ammonium salt formation was proved in by FT-IR spectroscopy by using a Shimadzu IR. The structure of synthesized IL is shown in figure 1.

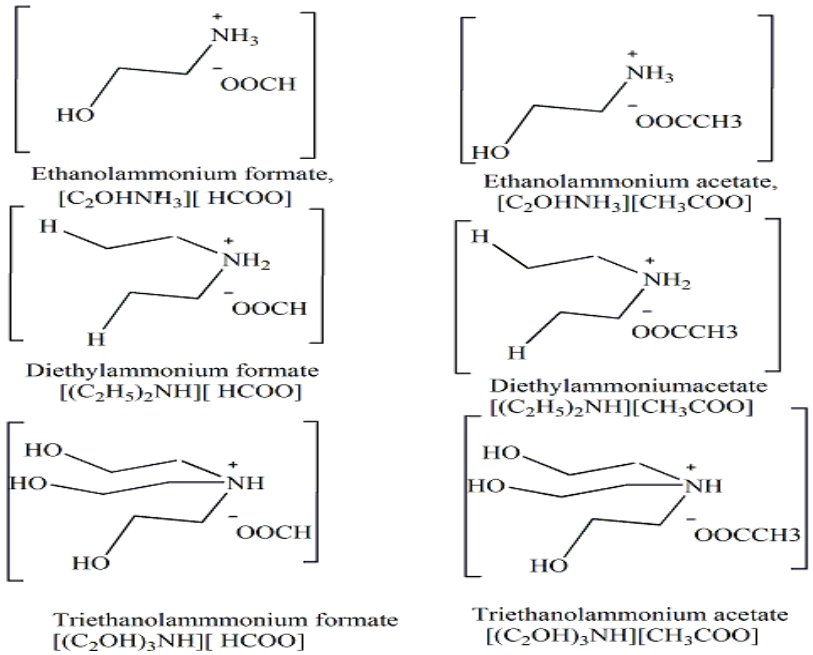


**Figure 1: synthesized ammonium based IL**

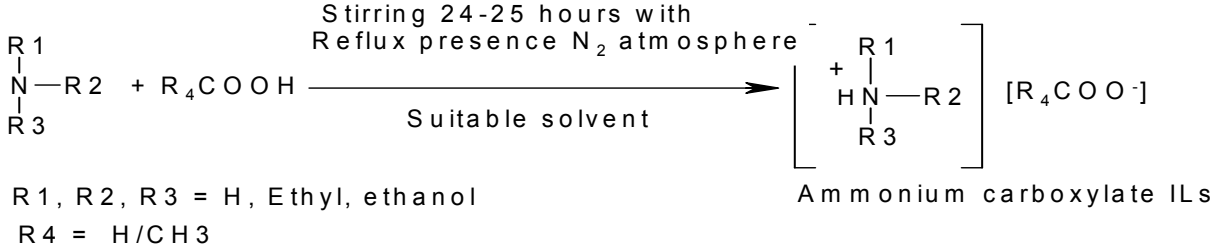


**Figure 2: Reaction scheme for synthesis**

3. **RESULT AND DISCUSSION:** The NMR, Elementary analysis, FTIR and Molecular Weight of synthesized ILs are listed in table 01

| Name | Abbreviation | Yield %, | Elementary analysis | Calculated | | | Molecular Weight |
|---|---|---|---|---|---|---|---|
| | | | | $^{1}$H NMR | $^{13}$C NMR: | FTIR | |
| Ethanolammonium formate (IL01) | [$C_2OHNH_3$] [HCOO] | 92 % | Found: %C = (33.62 34.02), %H= 8.47-8.23; %N= 13.08-12.96 | 2.0 (s, 1H, OH), 3,52(t, 2H, $NCH_2$), 4,27 (t, 2H, $OCH_2$), 7.0 (bs, 3H, $NH_3$), 9.6 (s, 1H, CH) | 63.3 (-$CH_2$), 42.9 (-$CH_2$), 47.0 -$CH_3$). FTIR: 3572 (-OH), 3147 (N-H) | Asymmetry, 3012 (C=C) in benzene ring, 2966 (N-H) symmetry, 2931 (C-H) asymmetry, 2897 (C-H) symmetry, 2360, 1589 (C-O) asymmetry, 1485 (-CO) symmetry cm$^{-1}$ | 107.11gm |
| Ethanolammoniumacetate (IL02) | [$C_2OHNH_3$] [$CH_3COO$] | 83.0 % | %C = 39.64-39.43, %H= 9.16- 9.51, % N= 11.56-11.27 | 2.0 (s, 1H, OH), 2.20 (s, 3H, CH3), 3,52 (t, 2H, NCH2), 4.21 (t, 3H, OCH2), 7.0 (bs, 3H, NH3) | 63.3 (-CH2), 42.9 (-CH2), 56.0 (CH2), 17.0 (-CH3) | 3437 (-OH), 3414(N-H asymmetry), 3390 (N-H) asymmetry, 2939 (C-H) asymmetry, 2893 (C-H) symmetry, 1697 (C-O) symmetry, 1723 (C-O) asymmetry, cm$^{-1}$ | 121.09 gm |
| Diethylammonium formate (IL03) | [$(C_2H_5)_2$ NH] [HCOO] , | 93.0 %, | %C = 50.38 – 50.22, %H= 11.00-11.76, % N= 11.75-12.03 | 1.61 (t, 6H, 2×$CH_3$), 3,27 (q, 4H, 2×$CH_2$), 7.0 (bs, 2H, $NH_2$), 9.2 (s, 1H, CH) | 11.08 (-$CH_3$), 42.2 (-$CH_2$), 30.6 (-$CH_3$), 47.0 (-$CH_3$), 6.8 (-$CH_3$), 6.8 (-$CH_3$) | FTIR: 3433(N-H) asymmetry, 3059(N-H) symmetry, 2993 (C-H) asymmetry, 2951 (C-H) symmetry, 2885 and 2796 alkyl C-H stretch asymmetry and symmetry of ethylamine, 1650 (C-O) asymmetry , 1589 (C-O) symmetry, cm$^{-1}$ | 119.10 gm |
| Diethylammonium accetate (IL04) | [$(C_2H_5)_2$ NH] [$CH_3COO$] | 96.0 %, | %C = 54.09-53.69, %H= 11.36-10.96, % N= 10.52-10.28 | 1.61 (t, 6H, 2×$CH_3$), 2.20 (s, 3H, $CH_3$), 3, 37 (q, 4H, 2×$CH_2$), 7.20 (bs, 2H, $NH_2$) | 11.08 (-$CH_3$), 42.2 (-$CH_2$), 30.6 (-$CH_3$), 56.0 (-$CH_3$), 17.0 (-$CH_3$), 6.8 (-$CH_3$), 6.8 (-$CH_3$) | 3437(N-H) asymmetry, 3414 (N-H) symmetry, 3035 (C-H) asymmetry, 2893 (C-H) symmetry, 2885 and 2796 alkyl C-H stretch asymmetry and symmetry of ethylamine, 1535 (C-O) asymmetry , 1500 (C-O) symmetry, cm$^{-1}$ | 133.12 gm |
| Triethanolammonium formate (IL05) | [$(C_2OH)_3$ NH][ HCOO] | 81 %, | %C = (45.90-45.70), %H= (9.44-9.66), % N= (7.65-7.40) | 2.0 (s, 3H, 3×OH), 3.43 (t, 6H, 3×$NCH_2$), 3.77 (t, 6H, 3×$OCH_2$), 7.08 (s, 1H, NH), 9.62 (s, 1H, CH) | 58.6 (-$CH_2$), 59.5 (-$CH_2$), 38.1 (-$CH_3$), 59.5 (-$CH_2$), 58.6 (-$CH_2$), 47 (-$CH_3$), 16.9 (-$CH_3$), 55.8(-$CH_2$) | 3417 for N-H bond of ammonium ion, 2947 (C-H) asymmetry, 2893 (C-H) asymmetry, 1666 (C-O) symmetry, 1593(C-O) symmetry | 195.52 gm |
| Triethanolammonium formate(IL06) | [$(C_2OH)_3$NH][$CH_3COO$] | 84.0 %, | %C= (45.60-45.28), H%= (9.74-9.89), % N= (6.65-6.52) | 2.05 (s, 3H, 3×OH), 2.26 (s, 3H, CH3), 3.21 (t, 6H, 3×$NCH_2$), 3.87 (t, 6H, 3×$OCH_2$), 7.0 (s, 1H, NH) | 58.6 (-$CH_2$), 59.5 (-$CH_2$), 38.1 (-$CH_3$), 59.5 (-$CH_2$), 58.6 (-$CH_2$), 56.0 (-$CH_2$), 17.0 (-$CH_3$), 55.8 (-$CH_2$). | 3363(N-H asymmetry), 3151(N-H asymmetry), 3016 (C-H) asymmetry, 2970 (C-H) symmetry,2931, and 2897, alkyl C-H stretch asymmetry and symmetry of ethanolamine, 2839.22 C-H asymmetry and C-H symmetry, 1566 (C-O) asymmetry,1485 (C-O) symmetry cm$^{-1}$ | 210.52gm |

**Table 1: Data of NMR, Elementary analysis, FTIR and Molecular Weight of synthesized ILs**

## 4. COMPUTATIONAL OVERVIEW OF THE SYNTHESIZED ILS

Computational chemistry is a branch of chemistry that uses computers to assist in solving chemical problems uses the results of theoretical chemistry, incorporated into efficient computer programs, to calculate the structures and properties of molecules and solids[36]. There are two broad areas within computational chemistry devoted to the structure of molecules and their reactivity as molecular mechanics (MM) and quantum mechanics (QM) [37]. They both perform the same basic types of calculations. Molecular mechanics uses the laws of classical physics to explain and interpret the structure and properties of molecules. Molecular mechanics methods are available in many computer programs, including MM3, HyperChem, Gaussian, Quanta, Sybyl and Alchemy[24, 38].
The tested compounds were constructed using Hyperchem$^R$ Release 8.0 (Hypercube, Inc.) drawing platform and are geometrically optimized for further calculations. For the calculation using molecular mechanics, these compounds were tested in two basis set, Mm+ and PM3. Single point calculations were carried out to create the data profile on the physical properties, such as total energy, binding energy, hydration energy, dipole moment, heat of formation and energies for HOMO and LUMO[27].
In the calculation, total energy and free energy for the compounds are same due to zero value of entropy. According to Gibb's equation, A = E – TS (where, A=Free energy, T=Temperature, E=Total energy, S=Entropy); If S=0, then A=E–0 and A=E.

Table01: Properties calculation using optimized Mm+ set.

| Chemicals | Calculation type | Total energy (kcal/mol) | Dipole moment (debyes) | Hydration energy (kcal/mol) |
|---|---|---|---|---|
| IL01 | Mm+ | 0.50945 | 3.50600 | 3.900 |
| IL02 | Mm+ | -0.69262 | 1.53409 | -11.670 |
| IL03 | Mm+ | 3.74434 | 2.18200 | -2.620 |
| IL04 | Mm+ | 7.14584 | 4.75700 | -14.100 |
| IL05 | Mm+ | 14.66050 | 2.40697 | -8.660 |
| IL06 | Mm+ | 17.62987 | 6.28500 | -17.940 |

Table02: Properties calculation using optimized PM3 set.

| Chemicals | IL01 | IL02 | IL03 | IL04 | IL05 | IL06 |
|---|---|---|---|---|---|---|
| Calculation type | PM3 | PM3 | PM3 | PM3 | PM3 | PM3 |
| Total energy (kcal/mol) | -34720.769 | -38168.335 | -48382.8 | -51832.872 | -61706.87 | -65498.4 |
| Binding energy (kcal/mol) | -1262.9246 | -1542.4632 | -1916.36 | -2198.4278 | -2533.345 | -2855.42 |
| Heat of formation (kcal/mol) | -93.863591 | -98.308165 | -137.554 | -144.52623 | -196.9071 | -191.771 |
| HOMO (eV) | -10.54155 | -10.06375 | -10.41844 | -10.24847 | -3.97568 | -8.92506 |
| LUMO (eV) | 1.13607 | 0.90474 | 0.93535 | 0.94809 | 0.83742 | -1.65566 |
| LUMO~HOMO (eV) | 11.677621 | 10.968487 | 11.353785 | 11.196557 | 4.813104 | 7.269402 |

The properties estimated by the basis set Mm+ and PM3, can be used to explain the molecular properties and variations can be attributed to the hydrogen bonding, binding energy calculations. According to the results found by the software, it is obvious that the total energy is more for higher alkyl chain ILs compare to the lower alkyl chain ILs. Standard methods were employed for all the test and experiments. Lower carboxylate such as formate and acetate ILs were tested in molecular programming software Hyperchem$^R$ Release 8.0 and found a good result for properties calculation.

## 5. EXPERIMENT WITH ANTIMICROBIAL ACTIVITY

### 5.1 Preparation of IL solutions in different concentrations

The required amount of the sample was measured in Digital balance with highly carefully so that no impurities were obtained. Then the required 1.5 mL distilled water was added and well shake for well soluble.

### 5.2 Antimicrobial assay

A preliminary investigation of the antibacterial activities of pure ILs was performed through measurements of primary screening both the gram-positive and gram-negative bacteria. Antibacterial screening of the test ILs were carried out with six bacterial pathogens, such as *Bacillus cereus*, *Staphylococcus aureus*, *Escherichia coli*, *Salmonella typhi*, *Pseudomonas aeroginosa* and *Shigella dysenteriae*. This method was carried on via well diffusion method[39-40]. The bacterial inhibition zone (subtracting the well diameter 5.0 mm) was measured in mm scale with consideration ±1.0 with all taking value. All the measurements were done in triplicate and the averages were listed in table 03. The initial concentration was maintained for all ILs in 1000 mM/L, 500 mM/L and 100 mM/L in distilled water or methanol. A control plate is always observed for the ILs if there is any significant inhibition occurred for the solvent. The results showed that all compounds had antimicrobial activity against bacterial pathogens used in this study.

Table03: Zone of inhibition (in mm) observed in three different concentrations (X=1000mM/L, Y=500mM/L, Z=100mM/L).

| pathogens and ILs | *B. Cereus* | | | *S. aureus* | | | *E.coli* | | | *S.typhi* | | | *P. aeroginosa* | | | *S. dysenteriae* | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | X | Y | Z | X | Y | Z | X | Y | Z | X | Y | Z | X | Y | Z | X | Y | Z |
| IL01 | 6.0 | 0 | 0 | 3.0 | 0 | 0 | 6.0 | 0 | 0 | 4.0 | 0 | 0 | 2.0 | 0 | 0 | 3.0 | 0 | 0 |
| IL02 | 11.0 | 0 | 0 | 6.0 | 0 | 0 | 10.25 | 6.0 | 0 | 11.0 | 5.2 | 0 | 3.0 | 0 | 0 | 4.0 | 0 | 0 |
| IL03 | 6.0 | 0 | 0 | 4.0 | 0 | 0 | 8.0 | 0 | 0 | 6.0 | 5.0 | 0 | 5.0 | 0 | 0 | 7.0 | 0 | 0 |
| IL04 | 8.0 | 0 | 0 | 6.0 | 0 | 0 | 9.0 | 0 | 0 | 7.0 | 0 | 0 | 7.0 | 0 | 0 | 9.0 | 0 | 0 |
| IL05 | 10.0 | 6.0 | 0 | 8.0 | 6.0 | 0 | 12.0 | 8.0 | 0 | 12.0 | 0 | 0 | 10.0 | 0 | 0 | 10.0 | 14.0 | 0 |
| IL06 | 11.0 | 0 | 0 | 9.0 | 0 | 0 | 13.75 | 0 | 0 | 14.0 | 0 | 0 | 11.0 | 0 | 0 | 12.0 | 0 | 0 |

From table 03, a graph is plotted different ILs vs Zone of inhabitation in fig 3.

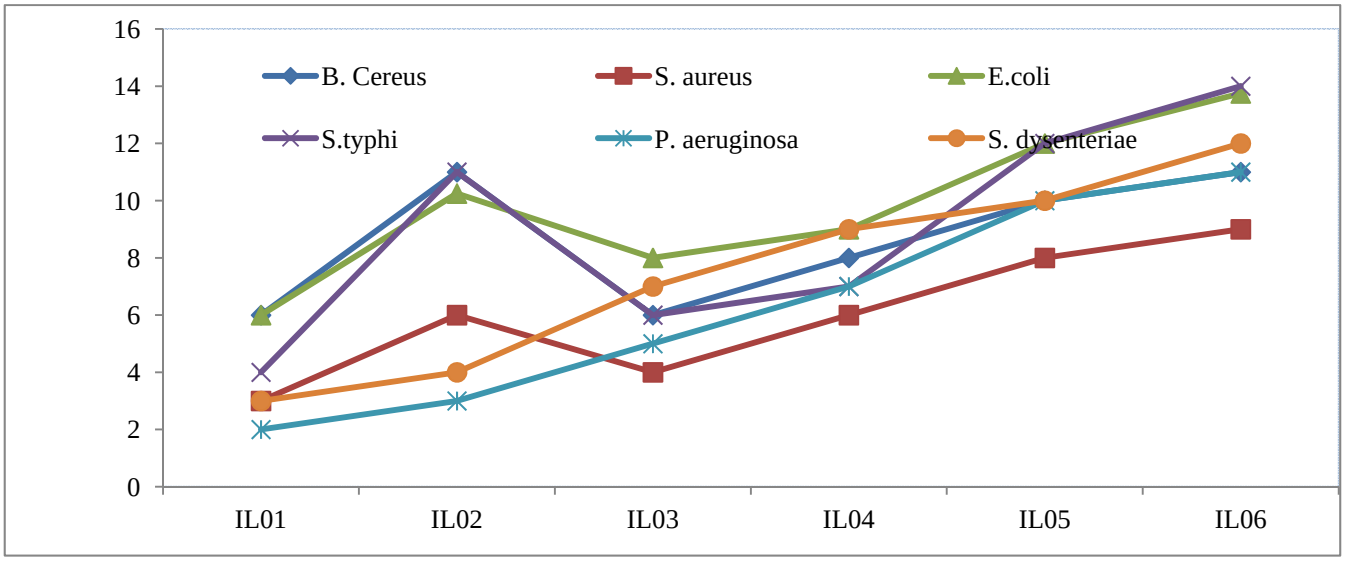


**Figure 3: A comparison study of toxicity for ammonium bases ILs.**

From the above graph, it was shown that the antimicrobial toxicity of primary, secondary and tertiary is not same. In all case, primary ammonium based ILs has lower toxicity than secondary ammonium based ILs. Again secondary ammonium based ILs has higher toxicity than tertiary ammonium based ILs. But in all case, anion also has an activity on the toxicity. The ammonium formate has lower toxicity than ammonium acetate.

**5.3 Antifungal Screening Test**

*Aspergillus niger* and *Rhizoplus azzahra* were used for evaluating the antifungal activity of all synthesized compounds. The antifungal activity was evaluated by Well diffusion method [41]. The media was altered Potato dextrose broth (abbreviated "PDB") is formulated identically to PDA, omitting the agar. Common organisms that can be cultured on PDB are molds such as *Aspergillus niger and Rhizoplus azzahra*. All synthesized compounds were dissolved in water or methanol basis on their solubility for making the concentration 1000 mM/L. The 100 µL solution of ILs were taken in

Petri-plate. The Media of PDB was dispersed and solidified. A well of 5 mm were made in the middle of Petri-plate using cork-borer and the fungal lead were place there. The plates were then kept in incubator for 96 h at 37°C. After 3 days, the fungal growth in presence of ILs, were measured and analyzed.

The antifungal test was completed and calculated the growth percentage compared with the control where the growth of control is 100% percent. The growth percentage is deduced as the following equation:

$$\%\text{Growth} = \frac{\text{Growth of fungi with ILs solution}}{\text{Growth of fungi without ILs solution as control}} \times 100$$

*Table no 04: Result and data for antifungal test.*

| Chemicals tested | Zone of growth (in mm) | | Percentage of Growth | | Percentage of Inhabitation | |
|---|---|---|---|---|---|---|
| | *Aspergillus niger* | *Rhizoplus azzahra* | *Aspergillus niger* | *Rhizoplus azzahra* | *Aspergillus niger* | *Rhizoplus azzahra* |
| Control | 28 mm | 41mm | 100.0% | 100.0% | -- | -- |
| IL01 | 20.5±1 | 29.2±1.0 | 78.57% | 71.34% | 21.25% | 28.66% |
| IL02 | 19.0±1.0 | 25.2±1.0 | 67.80% | 62.43% | 32.20% | 37.53% |
| IL03 | 23.0±1.0 | 28.5±1.0 | 82.14% | 69.51% | 17.86% | 30.49% |
| IL04 | 19.7±1.0 | 27.0±1.0 | 70.53% | 65.85% | 29.47% | 34.15% |
| IL05 | 24.0±1.0 | 30.5±1.0 | 85.00% | 74.39% | 15.00% | 25.61% |
| IL06 | 16.2±1.0 | 24.0±1.0 | 57.85% | 58.53% | 42.15% | 41.75% |

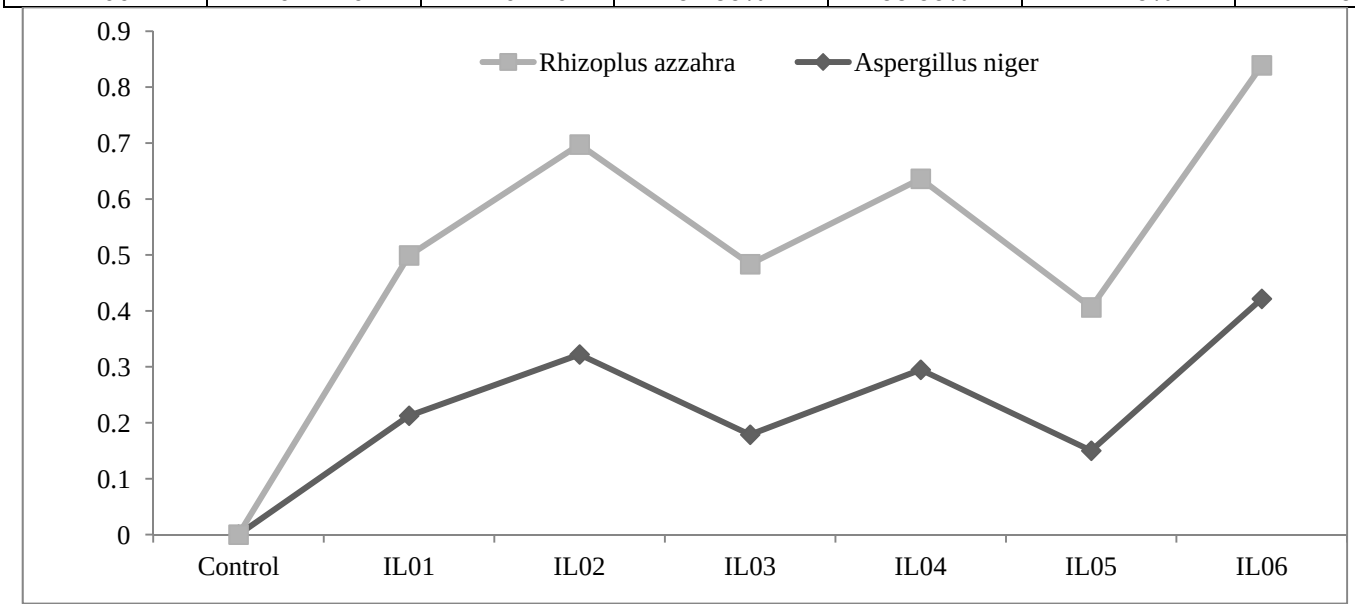


**Figure 4: Comparative study of 1°, 2 °and 3º ammonium based ILs**

From the above data, it is found that the antifungal activity against *Aspergillus niger and Rhizoplus azzahra* is a similar relation as like bacteria. The increasing order of antifungal activity is as of 1°< 2°< 3º ammonium based ILs. The anions have an effect on antimicrobial toxicity where acetate has higher antifungal activity than formate. The antifungal activity of *Rhizoplus azzahra* is higher than *Aspergillus niger.*

## 6. Conclusion

The synthesized ILs was characterized by Fourier Transform Infrared Spectroscopy (FT-IR), Nuclear Magnetic Resonance (1H-NMR and 13C NMR) Spectroscopy and elemental analysis. The synthesized quaternary ammonium formate and acetate based ILs is the room temperature ionic liquids so it has a great goal to use in chemical, pharmaceutical, textile industries, organic synthesis, organic extraction and separation method as solvent. Using HyperchemR computer programming, the HOMO-LUMO grap of IL01 to IL06 is 11.677621, 10.968487, 11.353785, 11.196557, 4.813104 and 7.269402 respectively that indicates the low reactivity of ILs. The low reactivity of ILs is the good agreements to use as solvents in chemical process. To safe use in chemical industries, the antimicrobial toxicity profile is the most imperative key point of chemical substance. To evaluate the antimicrobial toxicity profiles, bacteria is the first choice due to having short life. Antibacterial screening was conducted using the well-diffusion technique with ILs solutions of three different concentrations. The results were then used to select the initial higher concentration for dilution method, to obtain the concentrations as 1000, 500 and 100 mM/L and the antifungal activity was tested against two phytogathogenic fungi using well diffusion susceptibility test. All of synthesized ILs showed poor inhibition as potential antimicrobial agents, can proceed to future study. It also shows the information about the comparative

study of primary, secondary and tertiary-based ILs. They have very near toxicity. It offers a new scope to evaluate the physi0-chemical properties and application in chemical process as an alternative to organic traditional solvent.

## 7. Reference


1. Walden, P., *"Molecular weights and electrical conductivity of several fused salts".* Bull. Russian Acad. Sci, 1914. **8**: p. 405-422.
2. Seddon, K.R., *ionic Liquids for Clean Technology.* J. Chem. Technol. Biotechnol, 1997. **68**: p. 351–356.
3. Hasse, B., Lehmann, J., Assenbaum, D., Wasserscheid, P., Leipertz, A., Fröba, A., *Viscosity, interfacial tension, density, and refractive index of ionic liquids [EMIM][MeSO3], [EMIM][MeOHPO2], [EMIM][OcSO 4], and [BBIM][NTf2] in dependence on temperature at atmospheric pressure.* Journal of Chemical and Engineering Data, 2009. **54(9),** : p. 2576-2583.
4. Ghassan M JAl Kaisy, M.I.A., J MLeveque, T V V L NRao,, *Novel low viscosity ammonium-based ionic liquids with carboxylate anions: Synthesis, characterization, and thermophysical properties.* Journal of Molecular Liquids, 2017. **230**: p. 565-573.
5. Freemantle, M., *Designer solvents – Ionic Liquids may boost clean technology development. .* Chem. Eng. News,, 1998. **76,** : p. 32.
6. Wang X., H.F., Yang M., Melcher B., Fekete M., Mudring A. J., Wasserscheid P., Meyer, K., *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**: p. 7405-7407.
7. Greaves T.L., W.A., Fong C. ,Krodkiewska I. , Drummond C.J. ,, *Protic ionic liquids: solvents with tunable phase behavior and physicochemical properties.* Journal of .Physical Chemistry, B, 2006. **110**(45): p. 22479–22487.
8. J. S. Wilkes, R.D.R., and K. R. Seddon, and *'Ionic Liquids: Industrial Applications to Green Chemistry',.* American Chemical Society, 2002. **818,pp. 214–229**: p. 73–80.
9. Plechkova, N.V.S., . *Applications of ionic liquids in the chemical industry.* K. R. Chem. Soc, 2008. **37**: p. 123-150.
10. F. van Rantwijk, R.A.S., . , *Biocatalysis in ionic liquids.* Chem. Rev., 2007. **107 (6)**: p. 2757–2785.
11. J. Shen, H.W., H. Liu, Y. Sun, Z. Liu,. , *Brønsted acidic ionic liquids as dual catalyst and solvent for environmentally friendly synthesis of chalcone.* J. Mol. Catal. A: Chem, 2008. **280 (2008)**: p. 24–28.
12. Zhao H, B.G., Song Z, Olubajo O, Zanders L, Campbell SM., *Effect of ionic liquid properties on lipase stabilization under microwave irradiation.* Journal of Molecular Catalysis B 2009. **57**: p. 149–157.
13. Hussain W, P.D., Truppo M, Lye G, *Enzymatic ketone reductions with co-factor recycling: Improved reactions with ionic liquid co-solvents. J.* journal of Molecular Catalysis B: Enzymatic, 2008;. **55** p. 19–29.
14. Yang Z, P.W., *Ionic liquids: green solvents for nonaqueous biocatalysis.* Enzyme Microbiology and Technology, 2005: p. 19–28.
15. Tavares APM, R.O., Raquel Cristóvão, Macedo EA. , *Applications and Perspectives Ionic Liquids: Alternative Reactive Media for Oxidative Enzymes. In: Kokorin A (Ed) Ionic Liquids: .* Rijeka: InTech;, 2011: p. 449-516.
16. Rodríguez O, C.R., Tavares APM, Macedo EA. , *Effect of the alkyl chain length on enzymatic kinetic with imidazolium ionic liquids.* Applied Biochemistry and Biotechnology, 2011. **164**: p. 524-533.
17. H.F. Xiang, B.Y., H. Wang, H.W. Lin, X.W. Ge, S. Xie, C.H. Chen, and *Improving electrochemical properties of room temperature ionic liquid (RTIL) based electrolyte for Li-ion batteries.* Electrochim.Acta, 2010(55): p. 5204–5209.
18. S.Stevanovic, A.P., L.Mour, C.C.Santini, A.A.H.P .M.F.Costa Gomes,, *Absorption of carbon dioxide by ionic liquids with carboxylate anions.* International Journal of Greenhouse Gas Control, 2013. **17**: p. 78-88.
19. C. Lagrost, D.C.ı., M. Vaultier, P. Hapiot, , *Reactivities of some electrogenerated organic cation radicals in room-temperature ionic liquids: toward an alternative to volatile organic solvents?* J. Phys. Chem, 2003. **A 107**: p. 745–752.
20. Leta Deressa, T.B., S.Gupta, Ming-Jer Lee,, *The chemistry of ammonium-based ionic liquids in depolymerization process of lignin.* Journal of Molecular Liquids, 2017. **248**: p. 227-234.
21. Wei G.-T., Z.Y., C.J. Chen, , *Room temperature ionic liquid as a novel medium for Liquid/liquid extraction of metal ions,.* Anal. Chim. Acta, 2003. **488**: p. 183–192.
22. Waichigo M.M., D.N.D., *Comparison of ethylammonium formate to methanol as a mobile-phase modifier for reversed-phase liquid chromatography.* Journal of Sep. Society. , 2006. **29**(5): p. 599-606.
23. Wilkes J. S., J.A.L., R. A. Wilson and C. L. Hurrey *Dialkylimidazolium Chloroaluminate Melts: A New Class of Room-Temperature Ionic Liquids for Electrochemistry, Spectroscopy and Synthesis" Inorganic Chemistry, .* Scientific Research Publishing Inc. (SCIRP) 1982. **21**(3): p. 1263-1264.
24. Varadhi Govinda, P.V., Indra Bahadurb, *Molecular interactions between ammonium-based ionic liquids and molecular solvents: current progress and challenges.* Phys. Chem. Chem. Phys., 2016. **18**(12): p. 8278-8326

25. Saravanababu Murugesan , J.M.W., Rex X. Ren , Robert J. Linhardt, *Benzoate-based room temperature ionic liquids—thermal properties and glycosaminoglycan dissolution.* Carbohydrate Polymers, 2006. **63**: p. 268–271.
26. Earle M. J. , E.J.M.S.S., Gilea M. A., Lopes J. N. C., . Rebelo L. P. N, Magee J. W., Seddon K. R.,, *The distillation and volatility of ionic liquids.* Nature. , 2006. **7078**(394): p. 831-834.
27. Arunprakash T. K., R.F., Kamila D., *Ionic Liquids: Applications by Computational Design*. 2016, Inorganic and Bioinorganic Chemistry: University of Colorado Denver, Denver, CO, USA.
28. Ballone, R.J.B.a.P., *Computational Study of Room-Temperature Ionic Liquids Interacting with a POPC Phospholipid Bilayer.* The Journal of Physical Chemistry 2012. **116**(36): p. 11205–11216.
29. Bhargava B. L. , Y.Y., Michael L. Kleina, , *Computational studies of room temperature ionic liquid–water mixtures.* Chemical Communication, 2011. **47**(22): p. 6228-6241
30. Varma, S.R., *Solvent-Free Organic Syntheses Using Supported Reagents and Microwave Irradiation.* Green Chemistry, 1999. **1**(1): p. 43-55.
31. Coleman, D., Spulak, M., Garcia, M.T., Gathergood, N., , *Antimicrobial toxicity studies of ionic liquids leading to a 'hit' MRSA selective antibacterial imidazolium salt. .* Green Chemistry, 2012. **14**: p. 1350–1356.
32. Docherty, K.C.F.K.J., *Toxicity and antimicrobial activity of imidazolium and pyridinium ionic liquids.* Green Chemistry, 2005. **7**: p. 185–189.
33. Alvarez V.H., D.N., Gonzáles-Cabaleiro R., Mattedi S., Martin-Pastor M., Iglesias M., Navaza J.M., , *Brønsted ionic liquids for sustainable process: synthesis and physical properties.* Journal of Chemical &
Engineering Data, 2010. **55**: p. 625-632.
34. Bicak N., *A new ionic liquid: 2-hydroxy ethylammonium formate.* Journal of Molecular Liquids, 2004. **116**: p. 37-44.
35. Iglesias M., G.-O.R., Cota I.; Medina F.,, *Brønsted ionic liquids: Study of physico–chemical properties and catalytic activity in aldol condensations.* Chemical Engineering Journal, 2010. **162**: p. 802-808.
36. T., C., *A Handbook of Computational Chemistry*. 1985, Wiley: New York.
37. Ramachandran K.I., D.G., Krishnan Namboori. P.K. -Verlag GmbH ISBN -3-540-, *Computational Chemistry and Molecular Modeling Principles and applications.* Springer: p. 77302-3.
38. Pannuru Kiran Kumar, A.R., Lukman O. Olasunkanmi, Indra Bahadur, Pannuru Venkatesu , Eno E. Ebenso,, *Probing Molecular Interactions between Ammonium-Based Ionic Liquids and N,N-Dimethylacetamide: A Combined FTIR, DLS, and DFT Study.* J. Phys. Chem. B, , 2016. **120**(49): p. 12584–12595.
39. Bonev, B., Hooper, J., Parisot, J. , *Principles of assessing bacterial susceptibility to antibiotics using the agar diffusion method,.* Journal of Antimicrobial Chemotherapy, 2008. **61**(6): p. 1295-1301.
40. Standards, N.C.f.C.L., *Methods for dilution antimicrobial susceptibility tests for bacteria that grow aerobically;.* approved standard, 5th edn. NCCLS document M7-A5, Wayne, PA, 2000.
41. Standards, N.C.f.C.L., *Reference method for broth dilution antifungal susceptibility testing of yeasts;.* approved standard. NCCLS document M27-A, Wayne, PA., 1997.