American Journal of Heterocyclic Chemistry
2016; 2(1): 13-19
http://www.sciencepublishinggroup.com/j/ajhc
doi: 10.11648/j.ajhc.20160201.13

Aqueous Synthesis and Biological Studies of Indole Derivatives

Sudhakara Aralihalli[1, *], Raghavendra Ramappa[2], Harish Basavanthappa Gowdru[3], Nataraja Gummanar[1], Ramesha Sonnappa[4], Mahadevan Kittappa Malavalli[5]

[1]Department of Chemistry, Jain Institute of Technology, Davanagere, India

[2]Department of Micro Biology & Cell biology, Indian Institute of Science, Bengaluru, India

[3]Department of Biotechnology, M. S. RIT, Bengaluru, India

[4]R&D Center Department of Chemistry, Rajarajeswari College of Engineering, Bengaluru, India

[5]Department of Chemistry, Kuvempu University, Karnataka, India

Email address:

suda.sagar@gmail.com (S. Aralihalli)

[*]Corresponding author

To cite this article:

Sudhakara Aralihalli, Raghavendra Ramappa, Harish Basavanthappa Gowdru, Nataraja Gummanar, Ramesha Sonnappa, Mahadevan Kittappa Malavalli. Aqueous Synthesis and Biological Studies of Indole Derivatives. *American Journal of Heterocyclic Chemistry*. Vol. 2, No. 1, 2016, pp. 13-19. doi: 10.11648/j.ajhc.20160201.13

Received: October 26, 2016; **Accepted**: November 12, 2016; **Published**: December 27, 2016

Abstract: One pot with expedient approach to the synthesis of 2,3-disubstituted indole derivatives using indium (III) trichloride through Fisher indole method has been developed, in aqueous media. The synthesized compounds were screened for their anti analgesic and antibacterial activity. All the six indole derivatives exhibited significant antibacterial activity against *Pseudomonas aeruginosa* when compared to standard drug Ciprofloxacin. Based on the results of antibacterial activity, the molecular docking of all six indole derivatives were performed against *Pseudomonas* elastase a matrix metalloproteinase from *Pseudomonas aeruginosa* were presumed as an infectious wound healer via MMP dependent pathway. The active pocket docked with indole derivatives at the torsional degree of freedom 0.5 units with Lamarckian genetic algorithm. The inhibitors binding is facilitated by direct hydrogen bond interactions with the residues residing in the catalytic motif of *Pseudomonas* elastase consisted of Ala113, His140, Glu141, His144, Glu164, Arg198 and His223. In addition, the inhibitors make many hydrophobic interactions with both the enzyme and the co-factor Zinc ion. In view of the possibility that the elastase is an important determinant in *Pseudomonas* infection, it is conceivable that inhibitors of the enzyme will reduce its destructive effects that may lead to new therapeutic intervention.

Keywords: Fischer Indole, Indium (III) Chloride, EMK, Phenyl Hydrazine Hydrochloride, Molecular Docking, *Pseudomonas* Elastase

1. Introduction

Choice of solvent is one of the problems to face in order to perform eco-efficient processes. The use of water as a solvent in organic chemistry was rediscovered in the 1980s by Breslow [1], previously the scant solubility of the reactants was the main reason that ruled this solvent out from studies. Who showed that hydrophobic effects can strongly enhance the rate of several organic reactions and further reasons that make water is unique among solvents meanwhile it is cheap not inflammable and more significantly, it is not toxic. The indole nucleus is a common and important feature of a variety of natural products and medicinal agents [2]. The traditional approach for preparing the indole nucleus is the Fischer indole reaction [3], as this reaction has shortcomings, the palladium-catalyzed coupling of *o*-halo anilines is becoming an excellent alternative [4, 5]. A combination of such a palladium-catalyzed reaction with ketones and aldehydes would be a tremendously straightforward approach [6] for the synthesis of 2, 3-dialkyl indoles. In spite of diverse synthetic approaches developed so for, we thought it is perhaps hoped that environmental friendly one pot options seems possible with the use of soft Lewis acid which is stable to water. As part

of our research concerning the use of water as an eco-compatible solvent and herein we wish to disclose a new and efficient method for the synthesis of indoles using a indium (III) chloride catalyzed, the annulation between phenyl hydrazine hydrochlorides and Ethyl Methyl Ketone (EMK).

In recent years there has been a rising interest in the discovery of new antimicrobial compounds due to alarming increase in the rate of infections with multi-drug resistant microorganisms [7]. The increased prevalence of antibiotic resistance bacteria due to the extensive use of antibiotics may render the current antimicrobial agents insufficient to control bacterial diseases [8]. *Pseudomonas aeruginosa* is a ubiquitous bacterium that is responsible for many nosocomial infections such as pneumonia [9] surgical wound infections [10], and respiratory tract infections in cystic fibrosis patients [11]. *Pseudomonas* elastase is a zinc metalloendo peptidase, probably responsible for the tissue destruction observed during infections and enhances the growth and invasiveness of the organisms. Elastase-producing *P. aeruginosa* isolates were shown to significantly degrade human wound fluid as well as human skin proteins. Molecular docking (or binding) is an essential process in biochemical processes and is the main challenge in drug design since, the development of a pharmaceutical drug is a long, incremental process, typically requiring years of research and experimentation [12]. Hence, the present work reports anti-analgesic, antibacterial activity and molecular docking of zinc metalloendopeptidase *Pseudomonas* elastase with synthesized bioactive 2, 3-dimethyl indoles derivatives. The advantages of this method are low cost, finer selectivity, easy isolation of the product with good yield along with used water as an environment friendly solvent is the additional advantage in this etiquette.

2. Experimental

2.1. Materials and Measurements

All chemicals and reagents used in the current study were of analytical grade. Products were identified by their physical and spectroscopic data all the melting points were determined on an XT4 MP apparatus. The purity of the compounds was checked by TLC on silicagel. ^{1}H NMR spectra were recorded on a Bruker-400Hz spectrometer using DMSO-d_6 and $CDCl_3$ as an internal standard. Chemical shifts were reported in ppm (δ). IR spectra were obtained using a FTS-135 spectrometer instrument. Mass spectra were recorded on a JEOL SX 102=DA-6000 (10kV) FAB mass spectrometer. Elemental analyses were performed on a CHN-O-Rapid instrument and were within 0.4% of the theoretical values. Solvents, Chemicals and reagents were purchased from Merck chemical company in high-grade quality.

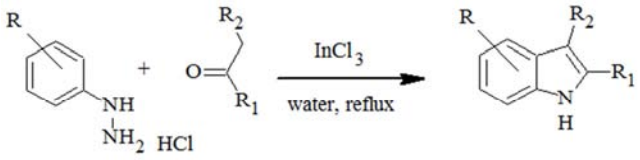
2.2. General Procedure for the Synthesis of 2,3-Di Substituted Indoles

The phenyl hydrazine hydrochloride [1gm (0.0069156mol)] and ethyl methyl ketone [0.49gm, (0.0069065mol)] were dissolved in water. Indium (III) chloride [(20 mole %) 0.6701 gm], was added to the reaction mixture and refluxed on water bath for appropriate time after the completion of the reaction as indicated by TLC the reaction mixture was poured into cold water (100mL) and the crude product was extracted with ethyl acetate washed with $NaHCO_3$ and brine, dried and evaporated under reduced pressure to provide a crude solid. The solid was further recrystal lised in acetone get pure product. The compound entries (1d in Table 1) are known, their identity were proven by means of melting point (Mp), IR, ^{1}H NMR, and MASS spectra [13].

3. Results and Discussion

3.1. Chemistry

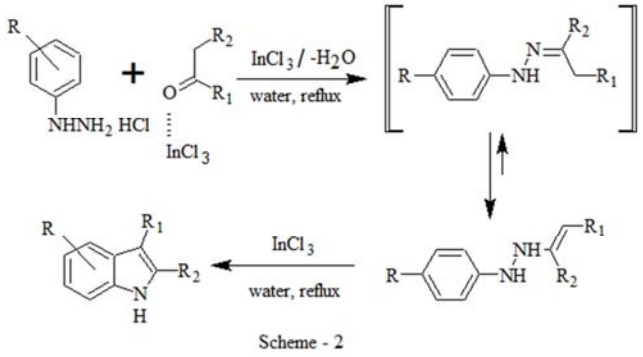
To study the generality of this process, it was demonstrated with wide variety of functionalized phenyl hydrazine hydrochlorides with EMK, (Scheme-1) the results are summarized in Table 1. An important issue of the Fischer indole synthesis in the cyclization of the phenyl hydrazones derived from unsymmetrical ketones is that the direction of cyclization is governed by the acidity of the reaction medium. The lower acid concentration or weaker acids promote cyclization towards the more branched carbon and higher acid concentrations, another way to explain stronger acids enhance the extent of cyclization at the less branched positions. As indium (III) chloride ($InCl_3$) provides a weaker acid system direction of cyclization occurred to produce more branched carbon during indole synthesis.



Scheme - 1

We felt that the phenyl hydrazine hydrochloride contains HCl could alone able to produce a hydazone that would undergo Fischer indolization under the acidic reaction conditions; hence the reaction was performed in the absence of catalyst. The reaction proceeds slowly in the beginning and after the prolonged reaction, the poor yield and undesired side products such as part of hydrazone and unreacted aryl hydrazine hydrochloride remains in almost 85-90%. Consequently, involvement of the catalyst in the reaction of aryl hydrazine hydrochloride with ethyl methyl ketone have been found to be significant in terms of clean reaction, high inter conversion and easy work up. Thus the indium (III) chloride was found essential to activate carbonyl group to react with phenyl hydrazine hydrochloride even in the presence of HCl in single step reaction condition. This single step reaction condition would be advantages from two perspectives; first it would obviate the need to prepare or isolate potentially sensitive aryl hydrazones and secondly it would provide a potentially very general means to the requisite N-aryl hydrazones for

fisher indolization from a single commercially available precursor, after examining a number of acids commonly employed fisher indolization procedures [13] and our previous work [14] we have discussed the synthesis of indole derivatives by using various organic solvents, however, using water as a solvent in the current procedure we found that the best results were obtained using indium (III) chloride.



Scheme - 2

Finally we found probably $InCl_3$ helps during hydrazone formation, followed cyclization occurs via thermal [3, 3] sigma tropic rearrangement at reflux temperature. Although water was found to be suitable solvent in this reaction procedure. The possible mechanism of Fischer indole synthesis catalyzed by indium (III) chloride as shown in Scheme-2.

Biological review

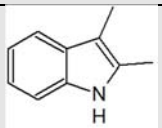
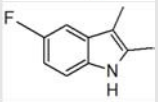
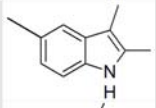
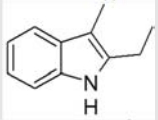
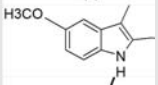
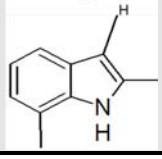
3.2. Anti Analgesic Activity

The analgesic activity of compounds a-f was performed by the acetic acid induced writhing test in mice by using previous method [15]. The compounds 5-methoxy-2,3-di methyl-1H-indole,5-ethyl-2,3-dimethyl-1H-indole,5-fluro-2,3-dimethyl-1H-indole,5-methyl-2,3-dimethyl-1H-indole, 5-Nitro-2,3-dimethyl-1H-indolefand2,3-dimethyl-1H-indo le, exhibitedexcellent analgesic activity (Table 2) compared with acetylsalicylic acid (aspirin) as a strandered analgesic agent.

3.3. Antibacterial Activity of the Synthesized Compounds

The 2, 3-dimethyl indole derivatives showing significant antibacterial activity against 5 clinically isolated strains of both Gram-positive and Gram-negative bacteria (Table 3). All the 2,3-dimethyl indole derivatives showed a significant zone of inhibition for Gram negative *Pseudomonas aeruginosa* isolated from the burn wound infected patients, when compared to other clinically isolated strains. Several recent reports have demonstrated that proteases of *P. aeruginosa* play a role in the virulence of this microorganism and are part of the pathogenic process in these infections. Generally Gram-positive bacteria should be more susceptible having only an outer peptidoglycan layer which is not an effective permeability barrier [16] whereas Gram-negative bacteria, having an outer phospholipidic membrane, carry structural lipopoly saccharide components. These make the cell wall impermeable to drug constituents. As earlier authors reported that most clinical isolates displayed multiple antibiotic resistances to various antibiotics clinically used against both Gram-positive and Gram-negative strains [17]. The present study indicates these indole derivatives can be used as potent antibacterial agents against *P. aeruginosa* infections.

Table 1. *Synthesis of 2, 3-dimethyl indoles.*

Entry	Product	Time (min)	Yield (%)[b]
1a		50	95
1b		45	80
1c		45	88
1d		120	65
1e		20	90
1f		80	80

Reaction was carried out at reflux temperature in Water in the presence of 20 mol% of $InCl_3$; [b] Isolated yields.

3.4. Molecular Docking of Pseudomonas Elastase

The current drug discovery is based on interaction of drug molecules that will bind to therapeutic target proteins. Inhibition of such protein gave the relief from the respective ailments. A major goal of rational drug designing is to generate novel drug molecules that can fulfill the pharmacophoric pattern and therefore, may act as new lead compounds, which may be considered further for the drug development process. Based on the results of *in vitro* antibacterial activity the molecular docking of 2, 3-dimethyl indole derivatives was performed. Before performing docking studies all derivatives were subjected to predict the Log P, Human intestinal absorption capacity, *In vitro* skin permeability and *In vitro* plasma protein binding capacity which indicates the bioavailability of these derivatives. The data was depicted in the Table 4. The log P value within 5 to 8 indicates the drug likeliness property of the derivatives.

Table 2. *Analgesic activity of Synthesized compounds.*

compound	Streptococcus haemolyticus	Staphylococcus aureus	Salmonella typhi	Pseudomonas aeruginosa	Klebsiella pneumonia	F- value
1a	11.20±0.47	19.57±0.35	8.97±0.18	22.87±0.19	12.17±0.15	19.7
1b	13.63±0.24	18.53±0.12	9.63±0.24	20.03±0.18	10.60±0.26	67.2
1c	13.90±0.23	19.67±0.23	11.67±0.12	21.50±0.15	10.33±0.15	124
1d	10.40±0.21	19.93±0.09	8.07±0.15	21.63±0.09	11.57±0.18	136
1e	11.40±0.06	17.67±0.12	8.33±0.22	22.08±0.11	10.27±0.12	65.4
1f	12.60±0.23	20.47±0.20	10.63±0.15	21.30±0.12	11.40±0.06	80.0
Ciprofloxacin	30.07±0.22	30.60±0.31	30.43±0.12	31.70±0.26	31.87±0.15	71.3

Table 3. *Antibacterial activity of the Synthesized compounds and their zone of inhibition (in mm).*

Compound	*Dose mg/kg*	*Mean no. of writhing Before Drug*	*After Drug*	% Protection
1a	100	28.00 ±061	13.33±0.32	52.39
1b	100	34.16±0.70	15.5±0.447	54.63*
1c	100	24.00±1.41	11.00±1.26	54.20*
1d	100	40.6±2.1	15.8±1.43	61.1*
1e	100	26.4±1.5	10.1±0.89	62.1*
1f	100	35.33±0.57	16.33±0.51	53.77*

Table 4. *Results of druglikliness and ADMET prediction using PreADMET server.*

Entry	Log_p	Human intestinal Absorption (%)	*In vitro* skin permeability (logKp, cm/hour)	*In vitro* plasma protein binding (%)
1a	2.758	100	-2.15	100
1b	3.159	100	-2.01	100
1c	3.183	100	-1.99	100
1d	3.331	100	-1.89	100
1e	2.791	93.92	-2.91	71.86
1f	4.105	100	-3.03	92.42

Mammalian skin serves for a number of vital physiological functions to maintain homeostasis. The functional properties of skin are often underappreciated until substantial loss of the skin occurs. The existence of undifferentiated cells in the skin suggests that skin has the potential to regenerate, but the context of molecular signals after tissue injury promotes scar repair. Bacterial protease have recently been evidenced in the interaction between host and the invading microorganisms: interruption of cascade activation pathways, disruption of cytokine network, excision of cell surface receptors, and inactivation of host protease inhibitors. The MMPs are involved in critical processes such as colonization and evasion of host immune defenses, acquisition of nutrients for growth and proliferation, facilitation of dissemination, or tissue damage during infection [18]. In the present study, *Pseudomonas* elastase was used as important therapeutic drug target in the search of potent and selective inhibitors using computer-aided molecular modeling and docking techniques. The docking accuracy and reliability of the 2, 3-dimethyl indole derivatives were evaluated along with the docking energy, inhibition constant, H-bond interaction and bond length. Comparative docking of *Pseudomonas* elastase with 2,3-dimethyl indole derivatives revealed that the docked energy and estimated inhibition constant for compounds was gradually decreasing with introducing the new group or changing the position of atoms (Table 5). The orientation of compounds 1a, 1b, 1c, 1d molecule was towards the catalytic motif of *Pseudomonas* elastase consisted of Ala113, His140, Glu141, His144, Glu164, Arg198 and His223. The reactive amide hydrogen of compounds 1a, 1c and 1d hydrogen bonded with the backbone amide nitrogen of His140 with a bond distance of 2.21Å, 2.22Å and 2.09Å respectively (Fig 1a, 1b, 1c). The amide hydrogen of compound b hydrogen bonded with the backbone reactive oxygen of Glu141 with a bond distance of 2.08Å (Fig 1d). The compound e formed two H-bonds with amino acid residues of the protein elastase, the first location of the H-bond formed was between amide hydrogen of 1e and the reactive oxygen of Glu141 with bond distance being 2.03Å. The second location of the H-bond formed was between the hydroxyl group of the 1e and the amine hydrogen of Arg198 with its bond distance being 2.03Å (Fig 1e). Earlier reports showed that a series of amino acid and peptide derivatives containing the metal-chelating moieties hydroxamate, phosphoryl, or thiol were synthesized and tested as potential inhibitors of the enzyme.

Table 5. *Results of molecular docking studies of Pseudomonas elastase with 2, 3-dimethyl indole derivatives.*

Entry	Docking Energy (Kcal/mol)	Inhibition constant (µM)	Amino acid residue involved in H-bond	Bond Length (Å)
1a	-5.96	4.31	His140	2.21
1b	-6.26	2.58	Glu141	2.08
1c	-6.39	2.06	His140	2.22
1d	-6.71	2.62	His140	2.09
1e	-7.01	0.72	Glu141	2.03
			Arg198	2.03
1f	--	--	--	--

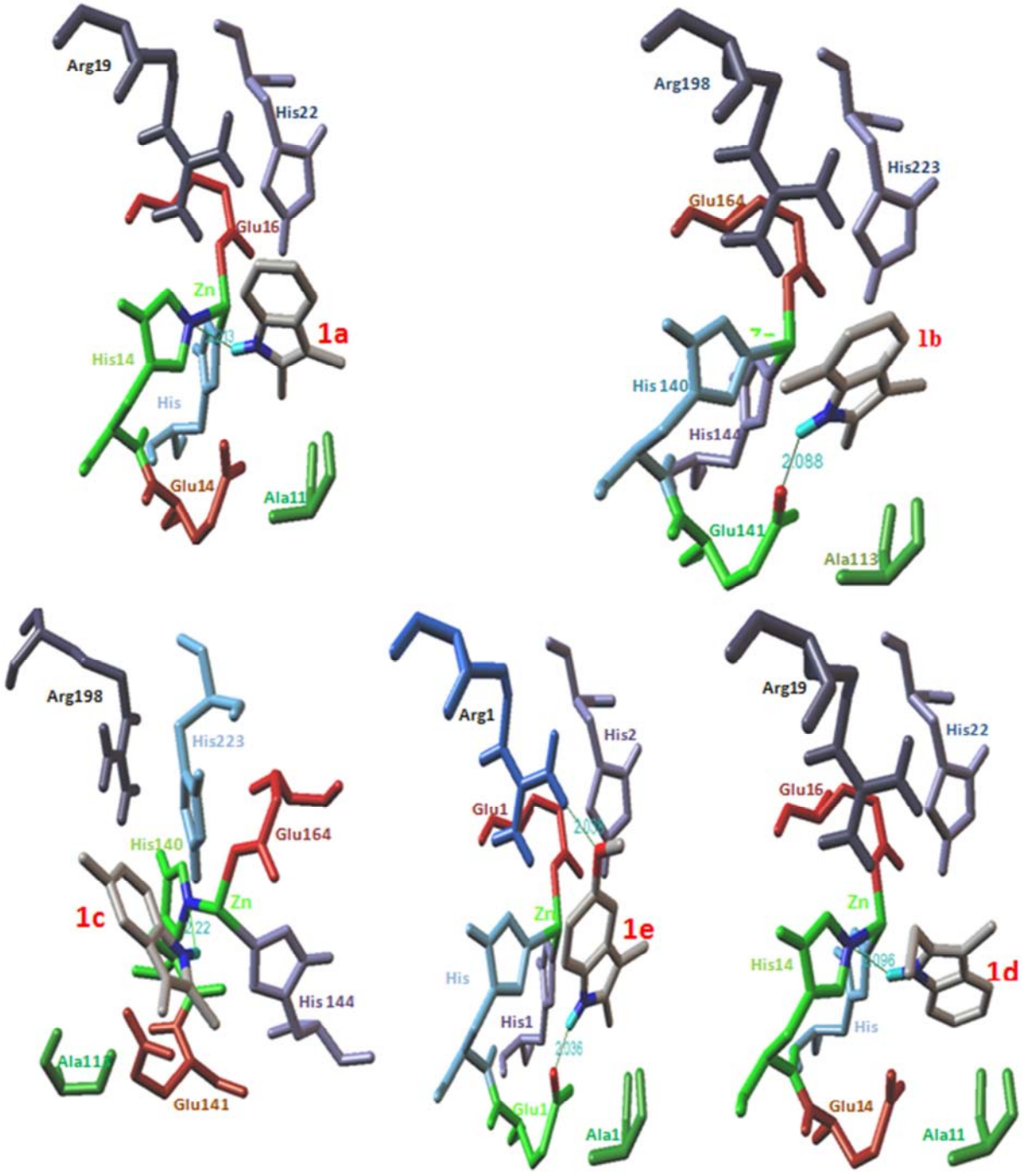


Figure. 1. *The orientation and H-bond interaction of 2, 3-dimethyl indole derivatives within the catalytic motif of the Pseudomonas elastase along with zinc ion (stick model). a) compound 1a, b) compound 1c, c) compound 1d, d) compound 1b, e) compound 1e.*

3.5. Antibacterial Activity of Compounds

The antibacterial activity of the 2, 3-dimethyl indole derivatives was screened by the agar well diffusion method [19] against 5 clinical isolate of bacterial strains belonging to Gram-positive *Staphylococcus aureus, Streptococcus haemolyticus* and Gram-negative *Pseudomonas aeruginosa, Salmonellatyphi* and *Klebsiella pneumonia*. The bacterial strains used for screening antimicrobial activity were collected from different infectious statuses of patients who had not taken any antibacterial drugs for at least two weeks with the help of an authorized physician, in the district health center of Gulbarga, Karnataka State, India. The clinical isolates were identified following a standard method [20]. The bacterial suspensions were diluted in 10^{-1} to 10^{-8} phosphate buffered saline. The fluoroquinolone antibiotic Ciprofloxacin (BioChemika, ≥98.0% (HPLC) (Fluka) was used as the standard (50µg/100µL of sterilized distilled water) concomitantly with the test samples. The minimal inhibitory concentrations (MIC) of the 2, 3-dimethyl indole derivatives were determined by micro dilute ion techniques in nutrient broth, according to National Commi ttee for Clinical Laboratory Standard, USA guidelines (NCCLS 1990). The inoculates were prepared in the same medium at a density adjusted to a 0.5 McFarland turbidity standard colony forming units and diluted 1:10 for the broth micro dilution procedure. The micro titer plates were incubated at 37°C and the MIC was determined after 24h of incubation. The results of these experiments are expressed as mean ± SE of six replicates in each test. The data were evaluated by one-way ANOVA followed by Tukey's pair-wise comparison test and the results were conside red significant when $p<0.05$.

3.6. Molecular Docking of Pseudomonas Elastase

Automated docking was used to determine the orientation of inhibitors bound in the active site of metallo endopeptidase elastase. A genetic algorithm method, implement ted in the program AutoDock 3.0, was employed [21]. The 3D structure files of 2, 3-dimethyl indole derivatives were designed and the structure was analyzed by using Chemsketch. The structure files are loaded on to prodrg server [22] and PreADMET server for energy minimization and drug likeliness prediction respectively. The protein structure file 3DBK was downloaded from Protein Data Bank (www.rcsb.org/pdb) was edited by removing the heteroatom, adding C-terminal oxygen [23]. For docking calculations, Gasteigere- Marsili partial charges [24] were assigned to the ligands and non-polar hydrogen atoms were merged. All torsions were allowed to rotate during docking. The grid map was centered at the residues of the protein predicted from the CASTp server [25]. The Lamarckian genetic algorithm and the pseudo-Solis and Wets methods were applied for minimization, using default parameters. The number of docking runs was 50, the population in the genetic algorithm was 250, the number of energy evaluations was 100,000, and the maximum number of iterations 10,000.

4. Conclusion

In summary we have developed eco-friendly reaction of phenyl hydrazine hydrochloride with EMK in aqueous media using soft catalyst a series of 2, 3-dimethyl indole derivatives have been synthesized and their antibacterial activity was done against clinically isolated strains of bacteria. All derivatives showed a significant zone of inhibition against *Pseudomonas aeruginosa* when compared to other bacteria. Based on this molecular docking of all derivatives was performed aginest pseudomonas elastase. In addition, the inhibitors make many hydrophobic interactions with both the enzyme and the co-factor Zinc ion. In view of the possibility that the elastase is an important determinant in *Pseudomonas* infection, it is conceivable that inhibitors of the enzyme will reduce its destructive effects that may lead to new therapeutic intervention.

5. Spectral Data

2,3-dimethyl-1H-indole (1a): Crystalin solid Mp 104 -106°C. IR (KBr): 3381 (NH). ^{1}H NMR (400 MHz; $CDCl_3$): δ 7.68 (br, s, NH), 7.46 (1H, d, J=8.05Hz), 7.24 (1H, d, J=4Hz), 7.11 (2H, m, J=8Hz), 2.3 (s 3H), 2.1 (s, 3H). ^{13}C NMR (75 MHz, $CDCl_3$): δ 156.6, 128.1,132.2, 131.6, 110.3, 108.9, 107.5, 103.1, 102.9, 11.6, 8.4; MS (EI 70 eV): m/z (%): 146 (M+1). Anal. Calcd (%): C (80.89), H (6.81), N (8.79);

5-fluoro-2,3-dimethyl-1H-indole (1b): crystalline solid Mp 60-61°C. IR (KBr):3385 (NH), ^{1}H NMR (400 MHz, $CDCl_3$): 7.6 (br s, NH), 7.1 (dd, 2H, J = 4.38),7.2 (dd, 1H, J = 4.38) 6.8 (t, 1H, J = 2.41 Hz), 2.3 (s 3H), 2.1 (s, 3H). ^{13}C NMR (75 MHz, $CDCl_3$):δ 156.6, 128.1,132.2, 131.6, 110.3, 108.9, 107.5, 103.1, 102.9, 11.6, 8.4; IR 3381. MS (EI 70 eV): m/z (%): 145 (M^+). Anal. Calcd (%): C (72.50), H (5.89), N (8.01).

2,3,7-trimethyl-1H-indole (1c): crystalline solid Mp 98-99°C. IR (KBr):3376 (NH). ^{1}H NMR (400 MHz, $CDCl_3$): 10.4 (br s, 1H, NH), 7.1 (dd, 2H, J = 8.9 Hz), 6.7 (d, 1H, J = 8.08 Hz), 2.3 (s, 3H), 2.2 (s, 3H), 2.1 (s, 3H). ^{13}C NMR (75 MHz, $CDCl_3$): 168.1, 155.4, 131.8, 131.5, 112.3, 109.3, 104.3, 97.8, 16.1, 11.8, 8.7. MS (EI 70 eV): m/z (%): 159 (M^+). Anal. Calcd (%): C (81.22), H (8.05), N (8.25);

5-methoxy-2,3-dimethyl-1H-indole (1e): crystalline solid: Mp 89-90°C IR (KBr): 3382 (NH), ^{1}H NMR (400 MHz, $CDCl_3$): 10.4 (br s, NH), 7.1 (d, 1H, J = 8.6 Hz), 6.8 (s, 1H), 6.6 (d, 1H, J = 2.24 Hz), 3.7 (s, 3H, -OCH_3), 2.2 (s, 3H), 2.1 (s, 3H). ^{13}C NMR (75 MHz, $CDCl_3$): 154.7, 131.7, 131.6, 108.1, 106.9,104.5, 102.5, 98.8, 20.4, 11.7, 8.9. MS (EI 70 eV): m/z (%): 176 (M^+). Anal. Calcd (%): C (74.93), H (7.30), N (6.90).

7-nitro-2,3-dimethyl-1H-indole (1f): crystalline solid: Mp 95-96°C. IR (KBr): 3368 (NH). ^{1}H NMR (400 MHz, $CDCl_3$): 7.6 (br s, NH), 7.5 (d, 1H, J = 7.6 Hz), 7.4 (d, 1H, J = 8.2 Hz), 7.3 (t, 1H, J = 8.0 Hz). ^{13}C NMR (75 MHz, $CDCl_3$): 135.2, 131.1, 129.5, 120.9, 119.4, 117.9, 110.3, 107.8, 11.54, 8.4. MS (EI 70 eV): m/z: 174 (M^+). Anal. Calcd (%): C (62.12), H (5.11), N (13.90).

References

[1] R. Breslow, D. C. Rideout, J. Am. Chem. Soc. 1980, 102, 7816–7817. (b) R. Breslow, Acc. Chem. Res. 1991, 24, 159-164.

[2] (a) J. E. Saxton, Indoles; Wiley-Interscience: Part 4, New York, 1983. (b) Saxton, J. E. Nat. Prod. Rep. 1994, 493; 1995, 387. (c) M. Ihara, Fukumoto, K. Nat. Prod. Rep. 1995, 277.

[3] For reviews on the Fischer indole reaction: (a) B. Robin son, The Fischer Indole Synthesis; Wiley-Interscience: New York, 1982. (b) D. L. Hughes, Org. Prep. Proc. Int. 1993, 25, 607. (c) G. W Gribble, Contemp. Org. Synth. 1994, 1, 1.

[4] (a) R. Odle, B. Blevins, M. Ratcliff, L. S. Hegedus, J. Org. Chem. 1980, 45, 2709. (b) A. Kasahara, T. Izumi, S. Mura kami, H Yanai, M. Takatori, Bull. Chem. Soc. Jpn. 1986, 59, 927. (c) R. C. Larock, S. Babu, Tetrahedron Lett. 1987, 28, 5291. (d) R. C. Larock, E. K. Yum, J. Am. Chem. Soc. 1991, 113, 6689. (e) R. C. Larock, J. M. Zenner, J. Org. Chem. 1995, 60, 482. (f) E. Desarbre, J.-Y. Merour, Tetrahedron

[5] Cheng-yi, Chen. David, R. Lieberman.; Robert, D. Larsen. Thomas, R. Verhoeven. and Paul, J. Reider. J. Org. Chem. 1997, 62, 2676-2677.

[6] For examples of couplings with 1, 3-dicarbonyl systems, see: (a) H. Iida, Y. Yuasa, C. Kibayashi, J. Org. Chem. 1980, 45, 2938-2942. (b) T. Sakamoto, T. Nagano, Y. Kondo, H. Yamanake, Synthesis. 1990, 215. (c) K. Koerber-Ple, G. Massiot, Synlett 1994, 759.

[7] Abu-Shanab Bassam, Adwan Ghaleb, Jarra Naser, Abu-Hijleh, Kamel, Adwan. Awni, Turk. J. Biol, 2006, 30, 195-198.

[8] Nelson, Cowan; Lara D. Nugent.; Emily M. Elliott.; Igor. Ponomarev; J. Scott Saults. Child Development. 1999, 70 (5), 1082–1097.

[9] R. T. Wirblin, In Prevention and Control of Nosocomial Infections, R. P. Wenzel, eds 3rd Ed. Williams & Wilkins, Baltimore,1997, 807–819.

[10] Kluytmans Jan, Belkum Van Alex, and Henri, Verbrugh. Nasal Carriage of Staphylococcus aureus: Epidemiology, Underlying Mechanisms, and Associated Risks, CLINICAL MICRO BIOLOGY REVIEWS. 1997, 505-520.

[11] H. Mark, M. D. Pollack. Social phobia: Clinical and research perspectives Depression and Anxiety. 1998, 4 (5), 256.

[12] E. C Lavelle, G. Grant, A Pusztai, U. Pfüller, D. T. O'Hagan, Mucosal immunogenicity of plant lectins in mice. Immunology. 2000, 99 (1), 30.

[13] D-Q. Xu, W-L. Yang, S-P. Luo, B-T. Wang, J Wu; Z–Y Xu, Eur. J. Org. Chem. 2007, 1007, 1012.

[14] WagawSeble, H. Yang Bryant, L. Buchwald Stephen, J. Am. Chem. Soc. 1998, 120, 6621-6622.

[15] A. Sudhakara, H. Jayadevappa, H N Harish Kumar, and K. M Mahadevan, Letters in Org anic Chemistry. 2009, 6, 159-164.

[16] B. M Basavaraja, L. P Srikrishna, H. M. Vagdevi, and V. P vaidya. IJGP. 2009, 13 (3), 75-77.

[17] R Scherrer and G Gerhardt, Journal of Bacteriology. 1971, 107, 718-735.

[18] O. Zaborina, J. E Kohler, Y. Wang, C. Bethel, O Shev chenko, L. Wu, J. R. Turner. and J. C. Alverdy, Annals of Clinical Microbiology and Antimicrobials (Bio Med Central) 2006.5:14.

[19] J. Travis, & Potempa, J. Biochim. Biophys. Acta. 2000 1477 (1-2), 35-50. Infect. Immun. 1997, 65 (5), 1830-1835.

[20] R. A. Carron, J. M. Maran, L. Montero, L. Fernandozaigo And A. A. Dominguez. 1987. Pl. Med. Phytother, 21: 195-202.

[21] S. T Cowen, S. Steel, Manual for the identification of Medical Bacteria. Barrow, G. I. and R. K. A. Feltham (Eds), Cambridge University, 1993; Press No. 32.

[22] M. A Bhat, S. Izaddoost, Y Lu, K. O Cho, K. W Choi, Bellen, H. J. Cell. 2003, 115 (6), 765-766.

[23] K. Ghose Arup and M. Gordon. Crippen. J. Chem. Inf. Comput. Sci. 1987, 27, 1.

[24] S. Francis, Binkowski. Shawn, J. Roselle.; Journal of Geophysical Research. 2003, V (108), no. D6, 4183.

[25] T Reya, H. Clevers. Nature. 2005, 434 (7035), 843-50.