Synthesis, Characterization, Molecular Docking of Sulphamethoxazole Schiff Base Metal Complexes and Its Antibacterial, Anti-Inflammatory and Anti Depressant Activity

R.Gandhimathi[1], Magdivari Sangeetha[2]

[1]Professor, [2]Research Scholar, Department of Pharmaceutical Sciences, Vels University, Chennai, Tamil Nadu, India

Abstract

In our study we have a tendency to synthesized schiff' base of bactericide drug sulphamethoxazole on treating with aromatic aldehydes like p-diethyl amino benzyldehyde and p-dimethyl amino benzyldehyde. The synthesized schiff's bases were regenerate to its ion Schiff bases by treating with methyl group halide. The ion Schiff bases were regenerate to metal complexes by treating with metals like CuCl2, ZnCl2 and CdCl2. All the synthesized compounds were characterised by Elemental analysis, IR and ^{1}H proton magnetic resonance. Docking study was performed to know the interaction of binding sites with protein receptor using MAO-B enzymes (PDB ID: 2BK5) and COX-2 enzyme (PDB ID: 5IKR) by Virtual Screening software for Computational Drug Discovery. Synthesized metal complexes were evaluated for antibacterial, anti inflammatory and antidepressant activity. Copper metal complexes showed potent antibacterial and anti-inflammatory activity. Significant anti-depressant activity was shown for 2A2 and 2B2 zinc metal complexes

Key words: *Sulphamethoxazole, antibacterial, anti-inflammatory, antidepressant activity*

Introduction

Schiff bases area unit the vital compounds within the field of healthful chemistry due to their wide selection of biological activities and industrial applications. Many studies showed that the presence of a lone combine of electrons during a sp2 hybridized orbital area unit of biological importance. Development of a replacement chemotherapeutical Schiff base could be a new space of analysis. Several studies are according relating to the biological activities of Schiff base viz. Anticancer, antibacterial drug antifungal and herbicidal activities. They conjointly function a back base for the synthesis of assorted heterocyclic compounds[1, 2]. We all need iron, copper and atomic number 30 for traditional brain perform however metal metabolism becomes dysregulated during a sort of neurodegenerative diseases. Metals accumulate in Alzheimer's {disease|Alzheimer's|Alzheimers|presenile insanity} dementia and shaking palsy and are deficient in Menkes disease. Whether or not excess metals seem as a cause or a consequence of the illness method isn't sure, however accumulation of metals have the potential to trigger cellular harm. During a healthy brain, metals ar tightly regulated. Through a sublime system of copper chaperones that taxi copper from the cell surface to specific living thing destinations,

Corresponding Author:
R.Gandhimathi,
Professor, Department of Pharmaceutical Sciences, Vels University, Chennai, Tamil Nadu, India
Email id: gandhimathi.sps@velsuniv.ac.in

there's primarily no free copper in cells. Cellular iron isn't regulated by chaperones however rather by iron regulative proteins that orchestrate and synchronize iron uptake with iron storage to cut back the supply of free iron. In distinction to copper and iron, atomic number 30 is that the 'wild card' of brain metal metabolism as a result of not solely will it contribute to the site of key antioxidative metalloenzymes, like the Cu-Zn SOD, however it additionally exists as free atomic number 30 in some colligation vesicles and acts as a neurochemical. Once brain tissue is broken, like following a stroke, free atomic number 30 will flood the abraded areas leading to necrobiosis thanks to atomic number 30 excitotoxicity. Transition metals perform an outsized vary of biological functions at intervals the brain. a standard feature is their ability to exist during a sort of chemical reaction states and participate in reaction reactions; therefore copper, iron, and metal ar all catalytically active metals during a category of enzymes that sequester free radicals. it's helpful to appear at the common and ranging functions of transition metals within the brain to higher perceive what mechanisms ar noncontinuous in metal dyshomeostasis and the way this could cause necrobiosis in diseases of the central nervous system[3-5].

Literature review reveals that synthesized schiff's base metal complexes possess sensible medicine property. Antibacterial medicines are accepted antimicrobial agents, which are wide utilized in varied diseases. up to now antibacterial drug metal advanced has not been synthesized. Thus we have a tendency to aim to synthesize schiff's base of antibacterial medicine with aromatic organic compound like p-diethyl amino benzyldehyde and p-dimethyl amino benzyldehyde and to make metal complexes higher than schiff's base with metals like copper, metal and metallic element.

Materials and Methods

Syntheis of schiff's base

The schiff's base 2A has been synthesized by refluxing the reaction mixture of hot ethanolic solution (30ml)of Sulphamethoxazole (mol wt. 253.28) with hot ethanolic solution (30 ml) of (0.01 mole) Para diethyl amino benzaldehyde " (mol wt 177.24) for about 2 – 3 hours at 60 – 70^0C. The resulting solution was concentrated, allowed to stand overnight and recrystallized with ethanol. The schiff's base 2B has been synthesized by refluxing the reaction mixture of hot ethanolic solution (30 ml0.01 mole) Sulphamethoxazole (mol wt 253.28) with hot ethanolic solution of para dimethyl amino benzyldehyde (mol wt 149.19) for about 2 – 3 hours at 60 – 70^0c The resulting solution was concentrated, allowed to stand overnight and recrystallized with ethanol (Figure-1)[6, 7].

Synthesis of transition metal complexes

Anhydrous $CuCl_2$, $ZnCl_2$ and $CdCl_2$ (0.0005 mol) was dissolved in 50ml ethanol added with cationic derivative of Schiff base refluxed for 6 hours. The reaction mixture was left overnight and recrystallized with C_2H_5OH. (Figure-2)[8].

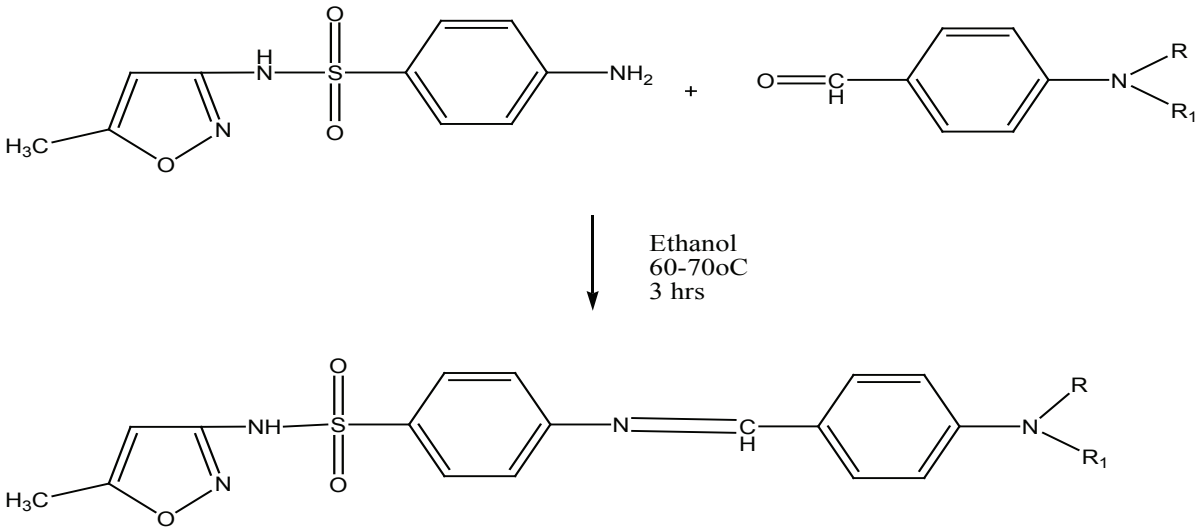

Figure 1- Scheme-1 (Synthesis of Sulphamethoxazole Schiff base)

Compound 2A R-C_2H_5, R_1-C_2H_5

Compound 2B R-CH_3, R_1-CH_3

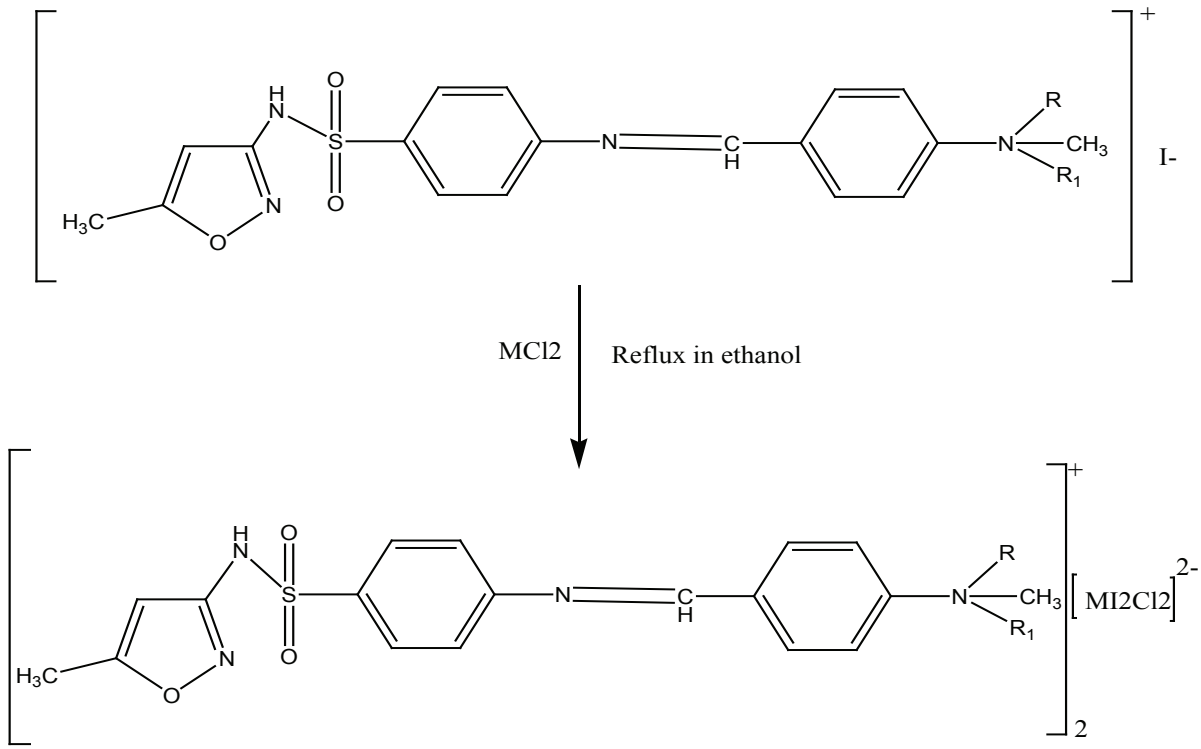

Figure 2-Scheme-2 (Synthesis of cationic Schiff base metal complex)

Compound 2A R-C_2H_5, R_1-C_2H_5

Compound 2B R-CH_3, R_1-CH_3

MCl_2- $CuCl_2$, $Zncl_2$ and $CdCl_2$

Docking study

Docking study was performed to know the interaction of binding sites with protein receptor using MAO-B enzymes (PDB ID: 2BK5) and COX-2 enzyme (PDB ID: 5IKR) by by Virtual Screening software for Computational Drug Discovery[9].

Anti bacterial activity

Required range of Muller agar plates were ready and divided into range of quadrant.Then the plates were inoculated with acknowledged take a look at organism. Sterile discs were placed with in every quadrant.Using small measuring device ten small cubic decimeter of saturated resolution of the derivatives is applied on the individual discs. Then the plates square measure incubated at 37°C for eighteen to twenty four hrs. Once incubation for every spinoff against completely different organisms was measured and tabulated[10, 11].

Determination of median lethal doses (LD50)

Animal's Swiss mice (20-25gm) and Male Sprague - Dawley rats (160-180) were maintained at customary diet and ad physical attraction. The experiment protocol was approved from institutional moral committee. LD50 values were calculable by the "acute toxicity test" as delineate elsewhere. The take a look at compounds were dissolved in three nothing DMSO administered orally to completely different teams with increasing doses. Six animals were taken in every cluster. Mortality make up my mind once twenty four hours of treatment. The dose, at that the fifty nothing mice survived, was thought of as LD50 worth of the compound[12].

Anti inflammatory activity

Swiss mice were divided into 5 teams of six animals every. The take a look at teams received orally twenty mg/kg of every sample. The reference cluster received diclofenac sodium (10 mg/kg, p.o) whereas the management cluster received vehicle (tween 80). After 1h, 0.1 mL, 1 Chronicles w/v carrageenin suspension in traditional saline was injected into the subplanatar tissue of the correct hind paw. The paw volume was measured at 30min. 1, 2,3 and 4hr once carageenan injection employing a micrometer screw gauge. The proportion inhibition of the inflammation was calculated[13].

Anti depressant activity

Male Sprague - Dawley rats consideration 160-180 grams are used. They are brought to the laboratory at least one day before the experiment and are housed one by one in makrolon cages with free access to food and water. Naïve rats are one by one forced to swim within a vertical Plexiglas. Rats placed in cylinders for the 1st time are at first extremely active, smartly swimming in circles, attempting to climb the wall or diving. Once 5-6 minutes immobility reaches a tableland wherever the rats stay immobile for around eightieth of the time. Once quarter-hour in the water the rats are removed and allowed to dry in a heated enclosure (32 c) before came back to their home cages. They are once more placed in the cylinder twenty four hours later and the total period of immobility is measured throughout a 5minute take a look at. Floating behaviour throughout this five minutes amount has been found to be duplicable in totally different teams of rats. Associate in Nursing animal is judged to be immobile whenever it remains floating passively in the water in a slightly round-backed however up-right position, its nose simply on top of the surface. take a look at medicine or commonplace are administered one hour before testing. Since experiments with the commonplace drug (Imipramine) showed that injections one,5 and twenty four previous the take a look at gave the most stable results in reducing floating these times are chosen for the experiment[14].

Results and Discussion

Characterization of synthesized compounds

2A1- Copper metal complex of *(E)*- N-(4-(diethyl, methyl amino) benzylidene)-4-(5-methyl isoxazol-3-sulfonamidyl) benzenamine

M.F: $C_{22}H_{27}Cl_2CuI_2N_4O_3S$. M.wt: 815.8. IR (KBr) cm-1: NH bond stretching at 3424 cm-1, stretching at 1690 cm-1 for C=N , S=O stretching at 1140 cm-1, C=C stretching at 1650 and 1475 cm-1. H1 proton magnetic resonance (CDCl3) δ values: Multiplet at 7.54-7.68 for aromatic nucleus, undergarment at 8.39 for N=CH peak, undergarment at 4.2 for aromatic NH peak, 2 triplet at 1.23 for 2 CH3 teams in N-ethyl substitution, 2 quadret at 3.39 for 2 CH2 cluster in N-ethyl substitution, undergarment at 2.42 for N-methyl substitution and undergarment at 2.35 for methyl radical hooked up in oxazole nucleus. Elem Anal Calc: C, 32.39; H, 3.34; Cl, 8.69; Cu, 7.79; I, 31.11; N, 6.87; O, 5.88; S, 3.93. Elem Anal Found: C, 32.29; H, 3.44; Cl, 8.65; Cu, 7.77; I, 31.31; N, 6.87; O, 5.88; S, 3.93.

2A2- Zinc metal complex of *(E)*- N-(4-(diethyl, methyl amino) benzylidene)-4-(5-methyl isoxazol-3-sulfonamidyl) benzenamine

M.F: $C_{22}H_{27}Cl_2I_2N_4O_3SZn$. M.wt: 817.6. IR (KBr) cm^{-1}: NH bond stretching at 3464 cm-1, stretching at 1690 cm-1 for C=N, S=O stretching at 1140 cm-1, C=C stretching at 1630 and 1475 cm-1. H1 magnetic resonance (CDCl3) δ values: Multiplet at 7.44-7.69 for aromatic nucleus, undershirt at 8.39 for N=CH peak, undershirt at 4.2 for aromatic American state peak, 2 triplet at 1.23 for 2 CH3 teams in N-ethyl substitution, 2 quadret at 3.39 for 2 CH2 cluster in N-ethyl substitution, undershirt at 2.42 for N-methyl substitution and undershirt at 2.35 for alkyl radical connected in oxazole nucleus. Elem Anal Calc: C, 32.32; H, 3.33; Cl, 8.67; I, 31.04; N, 6.85; O, 5.87; S, 3.92; Zn, 8.00. Elem Anal Found: C, 32.22; H, 3.43; Cl, 8.64; I, 31.08; N, 6.82; O, 5.89; S, 3.97; Zn, 8.00.

2A3- Cadmium metal complex of *(E)*- N-(4-(diethyl, methyl amino) benzylidene)-4-(5-methyl isoxazol-3-sulfonamidyl) benzenamine

M.F: $C_{22}H_{27}CdCl_2I_2N_4O_3S$. M.wt: 864.7. IR (KBr) cm^{-1}: NH bond stretching at 3405 cm-1, stretching at 1690 cm-1 for C=N, S=O stretching at 1140 cm-1, C=C stretching at 1640 and 1475 cm-1. H1 nuclear magnetic resonance (CDCl3) δ values: Multiplet at 7.46-7.70 for aromatic nucleus, undergarment at 8.39 for N=CH peak, undergarment at 4.2 for aromatic NH peak, 2 triplet at 1.23 for 2 CH3 teams in N-ethyl substitution, 2 quadret at 3.39 for 2 CH2 cluster in N-ethyl substitution, undergarment at 2.42 for N-methyl substitution and undergarment at 2.35 for alkyl radical hooked up in oxazole nucleus. Elem Anal Calc: C, 30.56; H, 3.15; Cd, 13.00; Cl, 8.20; I, 29.35; N, 6.48; O, 5.55; S, 3.71. Elem Anal Found: C, 30.52; H, 3.19; Cd, 13.01; Cl, 8.21; I, 29.37; N, 6.47; O, 5.56; S, 3.71.

2B1- Copper metal complex of *(E)*- N-(4-(trimethyl amino) benzylidene)-4-(5-methyl isoxazol-3-sulfonamidyl) benzenamine

M.F: $C_{20}H_{23}Cl_2CuI_2N_4O_3S$. M.wt: 787.7. IR (KBr) cm^{-1}: NH bond stretching at 3415 cm-1, stretching at 1690 cm-1 for C=N, S=O stretching at 1140 cm-1, C=C stretching at 1640 and 1475 cm-1. H1 nuclear magnetic resonance (CDCl3) δ values: Multiplet at 7.46-7.70 for aromatic nucleus, undergarment at 8.39 for N=CH peak, undergarment at 4.2 for aromatic NH peak, 2 triplet at 1.23 for 2 CH3 teams in N-ethyl substitution, 2 quadret at 3.39 for 2 CH2 cluster in N-ethyl substitution, undergarment at 2.42 for N-methyl substitution and undergarment at 2.35 for alkyl radical hooked up in oxazole nucleus. Elem Anal Calc: C, 30.49; H, 2.94; Cl, 9.00; Cu, 8.07; I, 32.22; N, 7.11; O, 6.09; S, 4.27. Elem Anal Found: C, 30.59; H, 2.98; Cl, 9.05; Cu, 8.17; I, 32.12; N, 7.01; O, 6.07; S, 4.25.

2B2- Zinc metal complex of *(E)*- N-(4-(trimethyl amino) benzylidene)-4-(5-methyl isoxazol-3-sulfonamidyl) benzenamine

M.F: $C_{20}H_{23}Cl_2I_2N_4O_3SZn$. M.wt: 789.6. IR (KBr) cm^{-1}: NH bond stretching at 3455 cm-1, stretching at 1690 cm-1 for C=N, S=O stretching at 1140 cm-1, C=C stretching at 1630 and 1475 cm-1. H1 magnetic resonance (CDCl3) δ values: Multiplet at 7.44-7.69 for aromatic nucleus, 8 undershirt at 8.39 for N=CH peak, undershirt at 4.2 for aromatic

American state peak, 2 triplet at 1.23 for 2 CH3 teams in N-ethyl substitution, 2 quadret at 3.39 for 2 CH2 cluster in N-ethyl substitution, undershirt at 2.42 for N-methyl substitution and undershirt at 2.35 for alkyl radical connected in oxazole nucleus. Elem Anal Calc: C, 30.42; H, 2.94; Cl, 8.98; I, 32.14; N, 7.10; O, 6.08; S, 4.26; Zn, 8.28. Elem Anal Found: C, 30.32; H, 2.98; Cl, 8.98; I, 32.24; N, 7.00; O, 6.18; S, 4.29; Zn, 8.24.

2B3- Cadmium metal complex of *(E)*- N-(4-(trimethyl amino) benzylidene)-4-(5-methyl isoxazol-3-sulfonamidyl) benzenamine

M.F: $C_{20}H_{23}CdCl_2I_2N_4O_3S$. M.wt: 836.6. IR (KBr) cm^{-1}: NH bond stretching at 3455 cm-1, stretching at 1690 cm-1 for C=N, S=O stretching at 1140 cm-1, C=C stretching at 1630 and 1475 cm-1. H1 magnetic resonance (CDCl3) δ values: Multiplet at 7.40-7.68 for aromatic nucleus, undershirt at 8.39 for N=CH peak, undershirt at 4.2 for aromatic American state peak, 2 triplet at 1.23 for 2 CH3 teams in N-ethyl substitution, 2 quadret at 3.39 for 2 CH2 cluster in N-ethyl substitution, undershirt at 2.42 for N-methyl substitution and undershirt at 2.35 for alkyl radical connected in oxazole nucleus. Elem Anal Calc: C, 28.71; H, 2.77; Cd, 13.44; Cl, 8.48; I, 30.34; N, 6.70; O, 5.74; S, 3.83. Elem Anal Found: C, 28.70; H, 2.78; Cd, 13.46; Cl, 8.47; I, 30.38; N, 6.71; O, 5.75; S, 3.84.

Molecular Docking Studies of synthesized Schiff's base metal complexes compounds

To understand the interaction of all the synthesized compounds (2A1-2A3 and 2B1-2B3) with MAO-B enzyme and COX-2 enzyme, the crystal structure of MAO-B enzyme and COX-2 enzyme was downloaded from Protein Data Bank (PDB ID: 2BK5 and 5IKR) and the molecular docking studies were performed.

The ligands 2A1 had a -score value of −9.3 to −8.5 against COX-2 enzyme (PDB ID: 5IKR) (Table-1 and figure-3). Out of all synthesized compounds 2A1 gave good score value than the standard. The minimum Glide energy required for the formation of complex between ligand and the receptor indicates excellent binding affinity.

Table-1 docking score values of synthesized metal complex 2A1

Ligand	Docking score binding affinity	Rmsd/ub	Rmsd/lb
5ikr_macro_prepared_2A1_ligand	-9.3	0	0
5ikr_macro_prepared_2A1_ligand	-9.1	25.864	24.43
5ikr_macro_prepared_2A1_ligand	-9	25.877	24.364
5ikr_macro_prepared_2A1_ligand	-9	54.922	52.502
5ikr_macro_prepared_2A1_ligand	-8.8	2.985	1.616
5ikr_macro_prepared_2A1_ligand	-8.7	28.398	24.268
5ikr_macro_prepared_2A1_ligand	-8.6	25.803	24.445
5ikr_macro_prepared_2A1_ligand	-8.6	25.42	23.292
5ikr_macro_prepared_2A1_ligand	-8.5	40.226	31.697

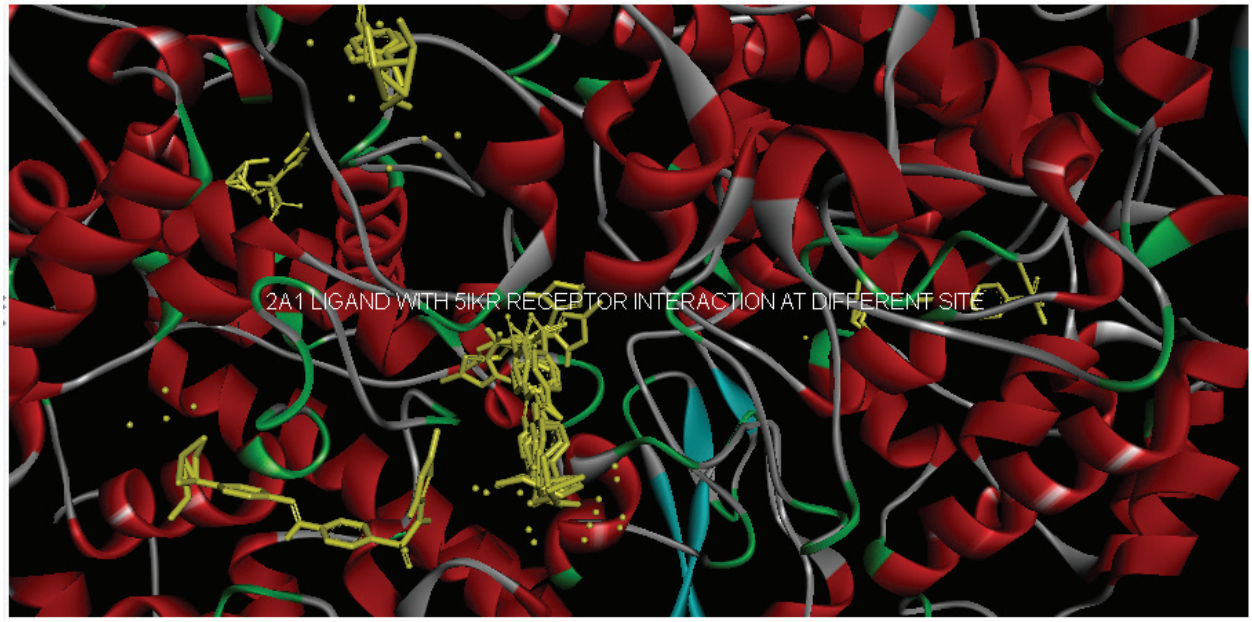

Figure 3-2A1 ligand with COX-2 (5IKR) receptor interaction at different active binding sites

The ligands 2A2 and 2B2 had a -score value of −9.0 to −7.5 against MAO-B enzyme (PDB ID: 2BK5) (Table-2 and figure-4). Out of all synthesized compounds 2A2 and 2B2 gave good score value than the standard imipramine.

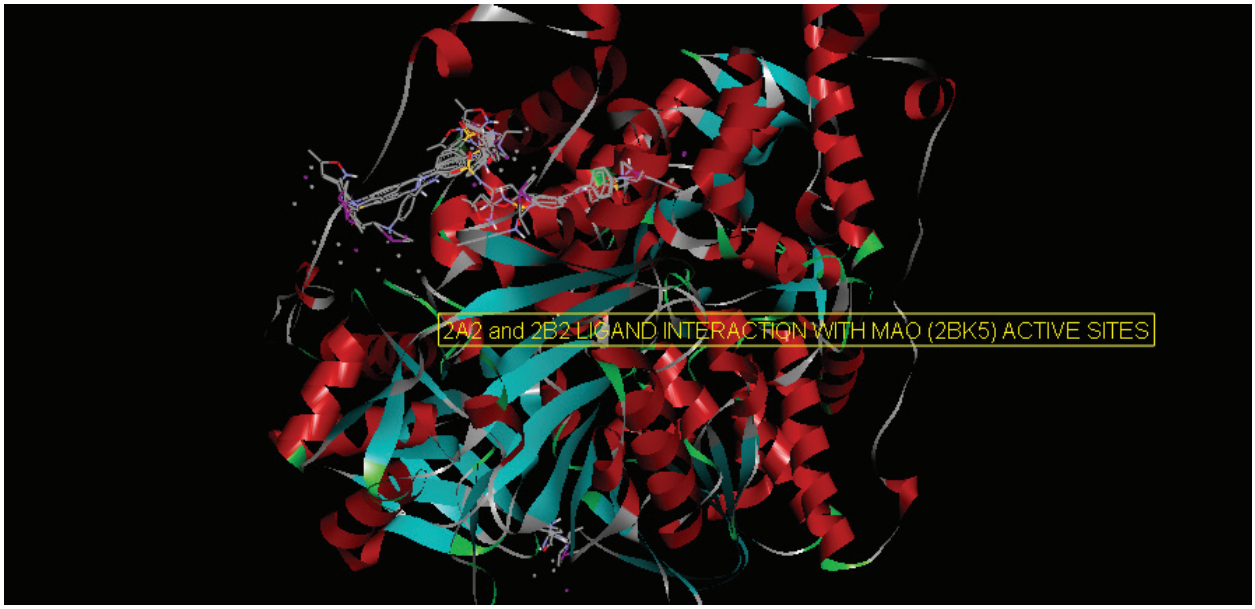

Figure 4-2A2 and 2B2 ligand with MAO (2BK5) receptor interaction at different active binding sites

Table-2 docking score values of synthesized metal complex 2A2

Ligand	Docking scoring binding affinity	Rmsd/ub	Rmsd/lb
2bk5_MACRO_PREPARED_NEW_2A2_ligand	-9	0	0
2bk5_MACRO_PREPARED_NEW_2A2_ligand	-8.3	43.145	40.927
2bk5_MACRO_PREPARED_NEW_2A2_ligand	-8.2	43.858	41.481
2bk5_MACRO_PREPARED_NEW_2A2_ligand	-8.2	45.801	42.408
2bk5_MACRO_PREPARED_NEW_2A2_ligand	-8.1	45.859	43.915
2bk5_MACRO_PREPARED_NEW_2A2_ligand	-8.1	45.765	43.917
2bk5_MACRO_PREPARED_NEW_2A2_ligand	-8.1	44.644	42.207
2bk5_MACRO_PREPARED_NEW_2A2_ligand	-7.8	43.928	42.025
2bk5_MACRO_PREPARED_NEW_2A2_ligand	-7.5	47.727	44.352

Antibacterial activity

Antibacterial activity was carried out by disc plate method using *B.cereus, E.coli, and P. aeruginosa* microorganism. The results are calculated and given in the table-3. The results of antibacterial activity revealed that the ligand with the Cd, Zn complexes does not exhibit antibacterial activities. However it is important to note that ligand with Cu complex exhibits potent antimicrobial activities. The Cu complex shows more activity than the Cd and Zn complex. This perhaps because of the upper stability of copper advanced than the Cd and metal advanced. The microorganisms take up metal ions on their cell walls and as a result respiration processes of cells square measure disturbed and macromolecule synthesis is blocked that is that the demand for more growth of organisms. The expansion inhibition effects of metal ions square measure goodly[15].

Table-3 Results of antibacterial activity of Schiff's base metal complexes of Sulphamethoxazole

S.No	Compounds	Zone of inhibition (mm)		
		B. cereus	E. coli	P.aeruginosa
1	2A1	25	24	22
2	2A2	25	18	24
3	2A3	21	20	20
4	2B1	20	24	22
5	2B2	22	17	18
6	2B3	19	17	19
7	Streptomycin	24	24	21

Anti inflammatory activity of synthesized compounds

Anti inflammatory activity was carried by paw oedema method and results shows that compound 2A1 and 2B1 at dose level of 20mg/kg.b.wt. significant activity (81 and 84 % respectively) when compared to standard diclofenac 10mg/kg.b.wt (84%). In our study results revealed that copper complexes showed potent anti-inflammatory activity but zinc complexes posses moderate anti-inflammatory activity, whereas cadmium complexes has not shown considerable activity. It was confirmed that the elevation of plasma copper-containing elements represents a physical response which can result in remission. Promotion of this physical response may be a valid approach to the treatment of the diseases with inflammatory elements. it absolutely was therefore confirmed that copper complexes, a singular category of doubtless additional therapeutically helpful for anti inflammatory medication[16].

Anti depressant activity of synthesized compounds

Anti depressant activity was evaluated by force swim test method, the animals which are immobile for less time considered as active and results showed significant percentage response such as 60%, 63% of the compound 2A2 (20mg/kg.b.wt.) and compound 2B2 (20mg/kg.b.wt.) respectively than the standard Imipramine (5mg/kg.b.wt). The most probable causes for depression area unit connected with the loss of physiological condition of the strain hormones, neurotransmitters, and disturbed trace components levels. It's rumored that youth stress could be a major risk issue for development of later depression because of affected maturation in brain, particularly in hippocampus. On the molecular level, these methods are also zinc-dependent via antioxidative activity changes and its influence on correct course of brain development process[17].

Conclusion

In conclusion we synthesized Schiff base by reacting Sulphamethoxazole with p-diethyl amino benzyldehyde and p-dimethyl amino benzyldehyde, six metal complexes of Schiff base by treating with $CuCl_2$, $ZnCl_2$ and $CdCl_2$. *In silico* studies results showed that docked complexes are the results of accumulative result of these interactions that are expected to own higher pharmacological activities such as anti inflammatory and anti depressant activity. Copper metal complexes 2A1 showed potent antibacterial activity against *B.cereus* and copper metal complexes 2A1 and 2B1 showed potent antibacterial activity against *E.coli* and copper metal complexes 2A1 and 2B1showed potent antibacterial activity against *P.aeruginosa* strains. Copper metal complexes 2A1 and 2B1 showed excellent anti-inflammatory activity. Significant anti-depressant activity was shown for 2A2 and 2B2 zinc metal complexes. The above 2A1, 2B1, 2A2 and 2B2 compounds activities are supported by results of *in silico* docking studies. Hence we tend to conclude that Schiff's base metal complexes offer a flexible platform for etymologizing numerous pharmacologically active medicines.

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Conflict of Interest: The authors declare no conflicts of interest

Ethical Clearance: All experimental protocols were approved under the Vel's University and all experiments were carried out in accordance with approved guidelines.

References

1. Arulmurugan S, Kavitha P, Venkatraman BR. Biological activities of schiff base and its complexes: a review. Rasayan Journal of

Chemistry, 2010; 3:385-410.

2. Iqbal N, Iqbal J, Imran M. Synthesis, characterization and antibacterial studies of some metal complex of Schiff base derived from benzaldehyde and sulfonamide. Journal of Scientific Research, 2009; 1:94-98.
3. Tyszka-Czochara MA, Grzywacz G, Gdula-argasi S, Librowski B, Wili Ski W, Opoka. The Role of Zinc in the Pathogenesis and Treatment of Central Nervous System (cns) diseases. Implications of zinc Homeostasis for proper CNS Function. Acta Poloniae Pharmaceutica Drug Research, 2014; 71:369-377.
4. Sanjay K, Saurabh K, Nath G, Tilak R, Sushil K. Synthesis, characterization, DNA cleavage and in vitro antimicrobial activities of Zinc(II) complexes of Schiff bases containing a 2,4-disubstituted thiazole. Transition Metal Chemistry, 2009; 4:195-198.
5. Banerjee M, Azam A, Sahu SK. Synthesis, Characterization and Biological Evaluation of Schiff'sbase Transition Metal Complexes with Celecoxib. Journal of Pharmacy Research, 2009; 2(6):1155-1158
6. Singh K, Barwa MS, Tyagi P. Synthesis and Characterization of cobalt (II), nickel(II), copper(II) and Zinc(II) complexes with Schiff base derived from 4-amino-3-mercapto-6-methyl-5-oxo-1,2,4-triazine. 2007; 42:394- 402.
7. Singh UK, Pandeya SN, Singh A, Srivastava BK, Pandey M. Synthesis and Antimicrobial Activity of Schiff's and N-Mannich Basesof Isatin and Its Derivatives with 4-Amino-N-Carbamimidoyl BenzeneSulfonamide. International Journal of Pharmaceutical Sciences and Drug Research, 2010; 2(2):151-154.
8. Negm NA, Zaki MF, Salem MAI. Cationic Schiff base amphiphiles and their metal complexes: Surface and biocidal activities against bacteria and fungi. Colloids and Surfaces B: Biointerfaces, 2010; 77:96-103.
9. Trott O, Olson AJ. AutoDock Vina: improving the speed and accuracy of docking with a new scoring function, efficient optimization and multithreading, Journal of Computational Chemistry, 2010; 31:455-461.
10. Wadher SJ, Puranik MP, Karande NA,Yeole YP. Synthesis and Biological Evaluation Schiff base of Dapsone and their derivative as antimicrobial agents. International Journal of Pharmaceutical Technology and Research, 2009; 1:22-33.
11. Uma Devi P, Aruna Lakshmi K, Taraka Ramji M, Akbar Ali Khan P. *In vitro* and in silico evaluation of metal complexes of quinazolinones incorporated with amino acids as potential antimicrobial agents Journal of Pharmacy Research, 2010;3(11):2765- 2768.
12. Usharani M, Akila E, Rajavel R. Evaluation of the pharmacological properties of schiff base mixed ligand Cu (ii), Co (ii), Ni (ii) and Zn (ii) complexes derived from 2-((e)-(4 nitrophenylimino) methyl) phenol. International Journal of Recent Scientific, 2013; 4(9): 1385-1390.
13. Hunskaar S, Hole K. The formalin test in mice, Dissociation between inflammatory and noninflammatory pain. Pain, 1987; 30:102- 103.
14. Burke A, Smyth EM, Fitzerald GA. Goodmanand Gilman's The Pharmacological Basis of Therapeutics, 5th ed. Sydney, McGraw-Hill, Pharmacotherapy of Gout, (2006), 671.
15. Raman N, Ravichandran S, Thangaraja C. Copper (II),Cobalt(II) and Zinc(II) complexes of Schiff base derived from Benzil 2,4-dinitrophenylhydrrzine with aniline. Journal of Chemical Science, 2004; 215-219.
16. Chinnasamy RP, Sundararajan R, Govindaraj S. Synthesis, characterization, and analgesic activity of novel schiff base of isatin derivatives. Journal of Advanced Pharmaceutical Technology and Research, 2010; 1(3):342–347.

17. Chandramouli C, Shivanand MR, Nayanbhai TB, Bheemachari B, Udupi RH. Synthesis and biological screening of certain new triazole schiff bases and their derivatives bearing substituted benzothiazole moiety. Journal of Chemical and Pharmaceutical Research, 2010; 4(2):1151–1159