INTERNATIONAL JOURNAL OF ADVANCES IN PHARMACY, BIOLOGY AND CHEMISTRY

Research Article

AM1 study of the electronic structure of Methicillin

Shabana Sultana, Bojja Rajeshwar Rao[1]*, Cherkupally Sanjeeva Reddy*

Department of Chemistry, Kakatiya University, Warangal, India-506 009.

[1]*Chemical Division, Kakatiya Thermal Power Project (O&M), Chelpur, India- 506 170.

Abstract

The geometry, conformation and electronic structure of methicillin have been optimized and calculated in the gas phase, usually considering an isolated molecule surrounded in a vacuum by using semi-empirical molecular orbital AM1 method. Further, the mechanism of protonation in methicillin has been studied by comparison of the different positions of net charges on nitrogen atoms in the molecule. In this connection, the heats of formation (ΔH_f^o), dipole moment (μ), ionization potential (IP), full atomic charges and energies of frontier molecular orbitals (E_{HOMO} and E_{LUMO}) have been performed and discussed. The conformational analyses of mono- and di-protonated species have also been performed for stable conformations.

Key words: AM1, methicillin, induction effect, frontier molecular orbital.

INTRODUCTION

Penicillin derivatives have been studied extensively due to their favourable absorption patterns and reduced undesirable side effects[1]. Methicillin was first penicillinase-resistant semi-synthetic penicillin in chemotherapy of bacterial infections[2]. Enzymatic splitting of natural penicillins and isolation of the important intermediate, 6-aminopenicillanic acid was led the preparation of several semi-synthetic penicillins[3]. Methicillin is a narrow-spectrum β–lactam antibiotic and it has relatively low protein binding (37 to 49%) to serum albumin[4]. In practice, penicillins had expected to assume that dipolar character of the drug could improve oral absorption[5]. A therapeutic advantage is due to a selective inhibition of the synthesis of bacterial cell walls and also the growth of both penicillin susceptible and penicillinase-producing staphylococci[6]. Austin Model-1 (AM1) is one of the semi-empirical methods with using experimental parameters and extensive simplification of the Schrodinger's equation ($H\Psi=E\Psi$) to optimize molecules for calculation of various properties to solve chemical problems[7]. It would be important to know the exact position of protonation centres' by the different positions of net charges on hetero atoms in the molecule. In this way quantum chemistry simulates chemical structure and reactions numerically and allows studying chemical phenomena by running calculations on computer rather than by examining reactions experimentally. Hence, it has been attracted that methicillin exists as anion and protonated forms and is considerably altered polarity, which are an advantage for inhibiting the synthesis of bacterial cell walls.

In view of these observations as a part of ongoing theoretical investigations[8, 9], the present study on molecular conformation and electronic properties of methicillin (**1**) in gas phase usually considering an isolated molecule surrounded by vacuum has been evaluated by AM1 method. From the obtained optimized electronic structure of methicillin, the mechanism of protonation has been studied by comparison of the relative values of net charges for nitrogen atoms at different positions of the molecule. Taking methicillin as a neutral molecule (**1**), the molecular geometry and conformations of mono-protonated (**2** & **3**) systems, di-protonated (**4**) system and anion (**5**) have been determined by full optimization calculations using semi-empirical molecular orbital AM1 method.

Computational methods[7]

Austin Model 1 (AM1) Semi-empirical molecular orbital calculations were performed on the molecules shown in Scheme-1 using the MOPAC93 in WinMOPAC ver 5.13 program by means of Intel Dualcore D102GGC2 DDR2 1GB SDRAM PC. The AM1 semi-empirical method is a modification of MNDO, offering more accurate parameterizations for polar systems and transition states. Geometry calculations in the ground state (keywords: PRECISE, equivalent to GNORM=5.0, CHARGE, GEO-OK, and MMOK to correct the increase in the barrier to rotation of the amide linkage) were completely optimized until the lowest energy conformation was found. The position of the atom in the molecule is mentioned as subscript (Figure-1). The initial molecular geometry was adopted as Pople's standard data[10], and subsequently fully optimized using an energy gradient method. The conformations were designated by Klyne-Prelog terms[8] using *s* = syn, *a* = anti, *p* = peri-planar (0 ± 30^0 & 180 ± 30^0) and all other angles *c* = clinal.

RESULTS & DISCUSSION

Electronic structure of methicillin (1) and its mono-protonated (2&3), di-protonated (4) and anion (5)

The optimized electronic structure of methicillin (**1**) and its mono-protonated (**2** & **3**), di-protonated (**4**) and anion (**5**) are shown in Scheme-1. In this context, the numbering of methicillin is shown in Figure -1. The calculated heats of formation (ΔH_f^o), ionization potential (IP), dipole moment (μ), the energies of frontier molecular orbitals (E_{HOMO} and E_{LUMO}) and net charges on hetero atoms of the molecules (**1** to **5**) are presented in Table-I. It is observed that the net charges on N_7- and N_{12}- atoms are -0.2440 and -0.3571 respectively in the case of methicillin (**1**). Usually, the nitrogen atom with larger negative value of net charge accepts proton more easily. It is also investigated that the sequence of protonation for nitrogen atoms of methicillin (**1**) is increasing in the order of $N_7 < N_{12}$. Thus, the negative charge distribution on nitrogen atoms N_{12}- atom is predicted to be main protonation site of methicillin (**1**).

It is observed that ionization potential values are increased in the order of molecules **5** < **1** < **3** < **2** < **4**. The di-protonated system (**4**) has more ionization potential. The calculated values of frontier orbital energies (E_{HOMO} and E_{LUMO}) reveal that molecules **1** and **5** have more electron-donor character whereas other molecules have electron-acceptor property. In the case of HOMO, the electron density is highest at N_{12}- atoms for molecules **1, 3, 4** and **5**. The results revealed that the electronic properties and reactivity of molecule depend on its conformational structure. The promotion of an electron from HOMO to LUMO, in a photochemical reaction, the supra-facial path way is allowed in the case of molecules **1** to **4**, due to the presence of same sign and antara-facial path way is allowed in the case of **5,** due to the presence of opposite sign[11]. The dipole moments of molecules depend on the nature of the atoms and bonds comprising the molecules and on their arrangement.

The dipole moment is increasing in the order of molecules **1** < **4** < **3** < **2** < **5**. Anion (**5**) shows higher dipole moment. The electronegative hetero-atoms cause displacement of electrons that induces an additional dipole moment in the molecule. The magnitude of the induction effect[12] (μ_{ind}) of molecules can be estimated with respect to methicillin (**1**). It is found that the induction effect is increasing in the case of $\Delta\mu_{ind}$ (**4**) 0.450D, $\Delta\mu_{ind}$ (**3**) 1.776D, $\Delta\mu_{ind}$ (**2**) 4.003D and $\Delta\mu_{ind}$ (**5**) 17.280D. According to the heat of formation (ΔH_f^o) data, the stability of compounds have decreased in the order of **5** > **1** > **3** > **2** > **4**. But geometry calculations in the ground state were completely optimized until the lowest energy conformation was found in the individual ions or molecules. It can be assumed that the electronic properties and reactivity of the molecule depend on its conformational structure. It is predicted that the protonation would take place preferably at N_{12}-atom than N_7-atom in the case of methicillin (**1**), this is due to the increased bond lengths of N_{12}-C_{11} (1.4102 Å), C_{13}-N_{12} (1.3937 Å) and C_{11}-C_9 (1.5685 Å). It is found that the stability of mono-protonated methicillin **3** (ΔH_f^o, +4.6714 kcal/mol) is more stable than **2** (ΔH_f^o, +6.3544 kcal/mol). The formation of di-protonated methicillin **(4),** from mono-protonated methicillins (**2** & **3**) is possible with the heat of formation (ΔH_f^o) of +246.9257 kcal/mol. The protonation site of methicillin **(1)** at N_{12}- atom is predicted to be the main basic centre of molecule. However, negative atomic charges are also present on the other atoms of the molecule.

In the case of neutral methicillin (**1**), the protonation at N_7-atom to form mono-protonated cation (**3**) is considered by increasing net atomic charges at N_{12}-, O_{10}-, O_{18}- and O_{20}- atoms and decreasing at N_7-, O_{32}-, O_{33}- and O_{37}- atoms. The protonation site of methicillin (**1**) at N_{12}- atom to form mono-protonated cation (**2**) is considered by decreasing net atomic charges at N_7-, N_{12}-, O_{32}-, O_{33}- and O_{37}-atoms and increasing at O_{10}-, O_{18}- and O_{20}- atoms. In the case of di-protonated cation (**4**), the negative atomic charges are decreased at N_7-, N_{12}-, O_{32}-, O_{33}- and O_{37}- atoms and increased at O_{10}-, O_{18}- and O_{20}- atoms. Anion of

methicillin (**5**) is formed by the removal of a proton from O_{10}-atom with increasing net charges at O_{10}-, O_{32}-, O_{33}- and O_{37}- atoms and decreasing at N_7-, N_{12}-O_{18}- and O_{20}- atoms.

The acid – base equilibrium of methicillin (1 to 5)

Equilibrium is normally established in polar solvents, and it is found as per Scheme-1 regarding the protonation of methicillin (**1**). N_{12}-atom is main basic centre in accordance with the negative charge distribution on N-atoms (Table-1). To determine the exact protonation centres of methicillin (**1**), the proton affinities (PA) for the different nitrogen atoms of the molecule have been calculated by means of AM1 method. The stable conformation of the cations formed by the protonation of each nitrogen atom of the molecule is determined; the heats of formation are calculated with full geometry optimization. The cations formed by the protonation at N_7- or N_{12}-atoms of methicillin (**1**) can exist in *anti*- or *syn*-conformations, as shown in Scheme-1. Its conformation can be assigned by comparison of its geometry and electronic structure. The proton affinity $(PA)^{13}$ values for the different nitrogen atoms of methicillin RH (**1**) were calculated by using the equation (1) and found to be 203.2923 kcal/mol and 205.0053 kcal/mol respectively in the case of mono-protonated methicillins (**2** and **3**). Di-protonated methicillin (**4**) was formed from either of mono-protonated methicillins (**2** and **3**) respectively with PA 126.6287 kcal/mol and 124.9157 kcal/mol.

$$PA = \Delta H_f^o(H^+) + \Delta H_f^o(B) - \Delta H_f^o(BH^+) \quad \ldots (1).$$

Where PA is the proton affinity,
$\Delta H_f^o(B)$ is the heat of formation for methicillin, $\Delta H_f^o(BH^+)$ is the heat of formation for the cation, and $\Delta H_f^o(H^+)$ is heat of formation for the proton (367.2 kcal/mol).
The proton affinity is in the order of N_7 (205.0053 kcal/mol) > N_{12} (203.2923 kcal/mol) and mono-protonated methicillin (**2**) appears to be more stable. All cations are solvated to form hydrogen bonds with the polar solvents which would affect the position of the equilibrium.

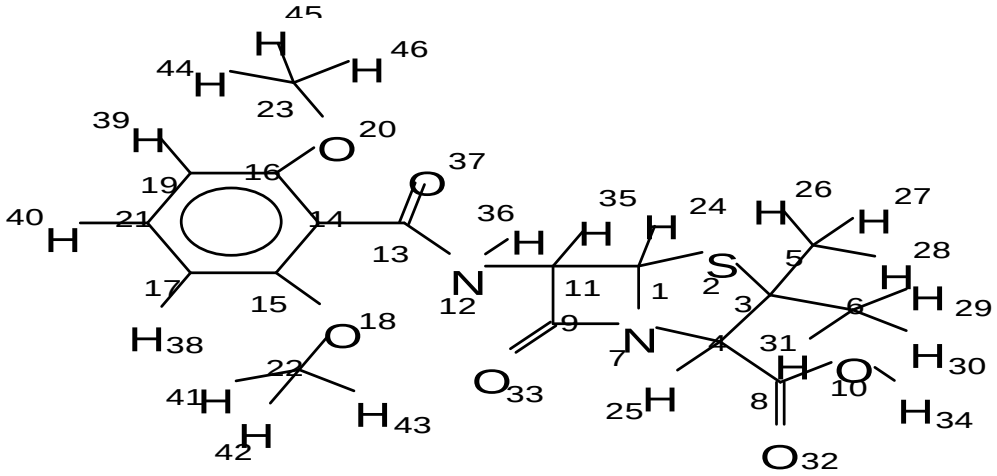
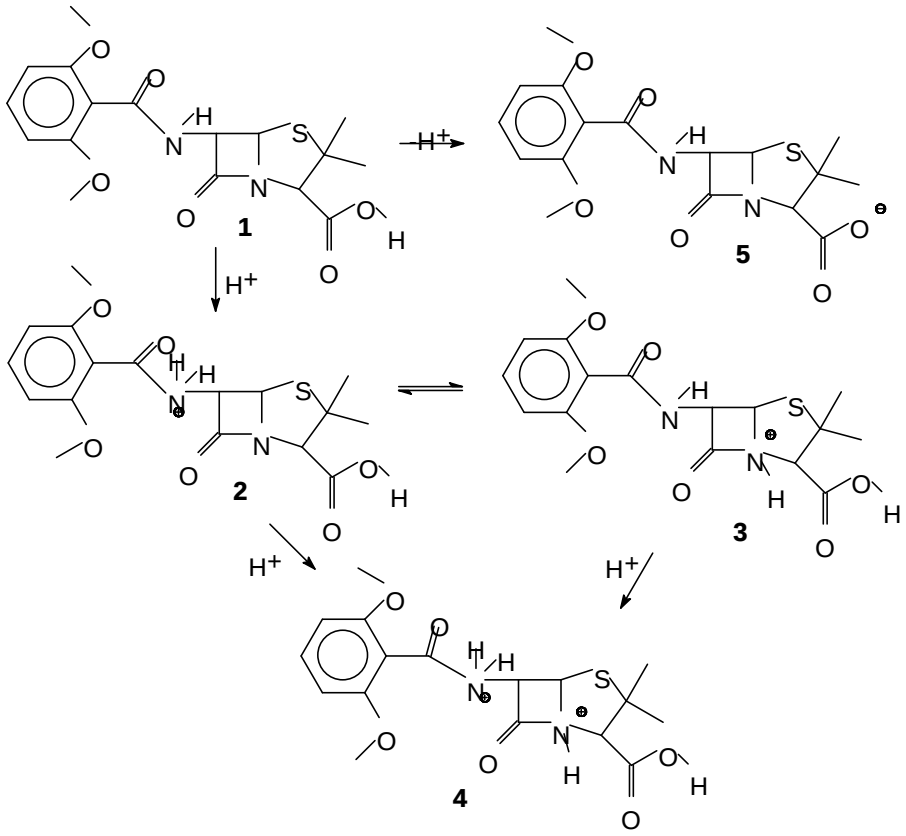


Figure - 1



Scheme - 1

The conformations of methicillin (1) and its mono-protonated (2 and 3), di-protonated (4) and anion R^- (5)

The spatial arrangement of atoms in a molecule is considered to study the conformations of methicillin (**1**), and its mono-protonated cations (**2 & 3**), di-protonated catrion (**4**) and anion (**5**) with a view to investigate molecular deformations. These can exist in *anti-* or *syn-* conformation, according to the position of atoms. In this context, the change in energy content of the protonation may depend on the changes in the parameters of dihedral angles. Figure-1 illustrates the atomic numbering of methicillin (**1**). Fully optimized AM1 calculations scrutinize only the main data of bond lengths (Table-II) and dihedral angles (Table-III) of molecules (**1** to **5**) for the sake of simplicity.

From the Table-II, Table-III and Scheme-1, mono-protonated methicillin (**2**) is formed by the addition of proton at N_{12}-atom of methicillin (**1**), with increasing bond lengths at N_{12}-C_{11} , C_{13}-N_{12} and C_{11}-C_9 and decreasing bond lengths at C_9-N_7 and O_{37}-C_{13}. The change of conformation from *–ac* of $C_{13}N_{12}C_{11}C_9$, *+ap* of $C_{14}C_{13}N_{12}C_{11}$, *+sp* of $O_{32}C_8C_4C_3$, *+ap* of $H_{34}O_{10}C_8C_4$ and –sp of $O_{37}C_{13}N_{12}C_{11}$ are changed to *-ap*, *-ap*, *+sc*, *-ap* and *+sp* conformations respectively. It is also observed that the protonation at N_{12}- atom is shown *–sc* conformation. If the mono-protonated methicillin (**3**) is formed by the addition of proton at N_7- atom of methicillin (**1**), with increasing bond lengths at C_{13}-N_{12} and C_9-N_7 and decreasing bond lengths at O_{32}-C_8, O_{33}-C_9 and N_{12}-C_{11}. The change of dihedral angle of $H_{34}O_{10}C_8C_4$ is converted *+ap* to *-ap* conformation and all other conformations are unaltered. It is observed that the protonation at N_7-atom is shown *-ap* conformation. In the case of formation of di-protonated methicillin (**4**), it is found that the dihedral angle of $C_{13}N_{12}C_{11}C_9$, $C_{14}C_{13}N_{12}C_{11}$, $O_{32}C_8C_4C_3$, $H_{34}O_{10}C_8C_4$ and $O_{37}C_{13}N_{12}C_{11}$ are changed conformations from *-ac* to *-ap*, *+ap* to *-ap*, *+sp* to *+sc*, *+ap* to *-ap* and *-sp* to *+sp* conformations respectively. It is also investigated that the protonation at N_7- atom and N_{12}-atom are shown respectively *-ac* and *-sc* conformations to form stable di-protonated methicillin (**4**). It can be concluded that

the anion (**5**) is formed with the removal of a proton from O_{10}- atom of methicillin (**1**), and the dihedral angle of $O_{10}C_8C_4C_3$ and $O_{32}C_8C_4C_3$ are changed the conformations from *-ap to +sp* and *+sp* to *-ap* respectively to form stable anion R^- (**5**) and rest of positions have moderate changes.

CONCLUSION

AM1 calculations show that protonated methicillins are nearly non-planar skeleton geometry, and the sequence of proton transfer at nitrogen atom is $N_{12} > N_7$. But, it is observed that N_7- protonated is more stable than N_{12}- protonated methicillin. All cations of methicillin are solvated to form hydrogen bonds with the polar solvents which would affect the position of the equilibrium. It is also predicted the stability of molecule in acid medium. Further, the utility of theoretical predictions is important for evaluating the ability of protein binding to serum albumin. This study reveals about the stability of conformations and molecular deformations, which is highly dependent relative upon the polarity of the medium.

ACKNOWLEDGEMENT

One of the authors (Smt. Shabana Sultana) is thankful to the Head, Department of Chemistry, Kakatiya University, Warangal – 506 009 for permission and encouragement.

Table –I

Heat of formation (ΔH_f^o in kcal/mol), ionization potential (eV), dipole moment (μ in Debye), energies of frontier molecular orbitals (in eV) and the atomic charges on S_2, N_7, N_{12}, O_{10}, O_{18}, O_{20}, O_{32}, O_{33} and O_{37} of methicillin(1) and its mono-protonated forms (2 & 3), di-protonated form (4), and anion (5) from AM1 calculations.

Parameters	1	2	3	4	5
ΔH_f^o (kcal/mol)	-157.5533	+6.3544	+4.6414	+246.9257	-183.2523
Ionization potential (eV)	8.9140	12.040	11.569	14.854	4.944
μ (Debye)	5.428	9.431	7.204	5.878	22.708
E_{HOMO} (eV)	-8.910	-12.040	-11.569	-14.854	-4.944
E_{LUMO} (eV)	-0.204	-4.233	-4.296	-8.186	+1.647
S_2 (atomic charge)	+0.0544	+0.1268	+0.2284	+0.2993	-0.0938
N_7 (atomic charge)	-0.2440	-0.2080	-0.1144	-0.0916	-0.1983
N_{12} (atomic charge)	-0.3571	-0.0909	-0.3741	-0.1405	-0.3397
O_{10} (atomic charge)	-0.2851	-0.2912	-0.3203	-0.3306	-0.5683
O_{18} (atomic charge)	-0.2126	-0.2708	-0.2369	-0.2889	-0.1919
O_{20} (atomic charge)	-0.1920	-0.2122	-0.1997	-0.2267	-0.1824
O_{32} (atomic charge)	-0.3570	-0.3235	-0.2722	-0.2355	-0.5134
O_{33} (atomic charge)	-0.2410	-0.1858	-0.0949	-0.0347	-0.2521
O_{37} (atomic charge)	-0.3335	-0.1564	-0.2950	-0.1443	-0.3536

Table –II

Bond lengths of methicillin (1) and its mono-protonated forms (2 & 3), di-protonated form (4), and anion (5) from AM1 calculations.

Bond lengths (´)	1	2	3	4	5
C_3-S_2	1.8223	1.8295	1.8078	1.8141	1.8536
C_9-N_7	1.4480	1.4338	1.5610	1.5455	1.4348
N_{12}-C_{11}	1.4102	1.4684	1.3955	1.4649	1.4183
C_{13}-N_{12}	1.3937	1.5337	1.4166	1.5849	1.3806
C_{11}-C_9	1.5685	1.5711	1.5601	1.5419	1.5776
O_{10}-C_8	1.3585	1.3556	1.3588	1.3568	1.2615
O_{32}-C_8	1.2343	1.2318	1.2256	1.2231	1.2522
O_{33}-C_9	1.2184	1.2138	1.1984	1.1995	1.2191
O_{37}-C_{13}	1.2421	1.2175	1.2358	1.2112	1.2448
H_{34}-O_{10}	0.9729	0.9751	0.9757	0.9793	- -
H_{36}-N_{12}	0.9932	1.0267	0.9914	1.0238	0.9934
H-N_7	- -	- -	1.0207	1.0242	- -
H-N_{12}	- -	1.0354	- -	1.0367	- -

-

Table – III
Dihedral angle (°) of methicillin (1) and its mono-protonated forms (2 & 3), di-protonated form (4), and anion (5) from AM1 calculations

Dihedral angle (°)	1		2		3		4		5	
	Angle	(*)	Angle	(*)	Angle	(*)	Angle	(*)	Angle	(*)
$C_4C_3S_2C_1$	-19.69	*-sp*	-19.39	*-sp*	-22.99	*-sp*	-24.58	*-sp*	-19.69	*-sp*
$C_8C_4C_3S_2$	+162.71	*+ap*	+161.64	*+ap*	+159.33	*+ap*	+159.26	*+ap*	+165.71	*+ap*
$O_{10}C_8C_4C_3$	-167.36	*-ap*	-147.94	*-ap*	-158.61	*-ap*	-152.00	*-ap*	+28.22	*+sp*
$C_{13}N_{12}C_{11}C_9$	-124.04	*-ac*	-158.90	*-ap*	-126.02	*-ac*	-172.94	*-ap*	-132.07	*-ac*
$C_{14}C_{13}N_{12}C_{11}$	+178.79	*+ap*	-170.37	*-ap*	+174.59	*+ap*	+175.40	*+ap*	+177.61	*+ap*
$C_{15}C_{14}C_{13}N_{12}$	-59.83	*-sc*	-53.75	*-sc*	-57.72	*-sc*	-54.44	*-sc*	-61.85	*-sc*
$O_{32}C_8C_4C_3$	+18.48	*+sp*	+36.08	*+sc*	+26.65	*+sp*	+32.19	*+sc*	-156.62	*-ap*
$O_{33}C_9N_7C_4$	+58.22	*+sc*	+63.15	*+sc*	+65.06	*+sc*	+73.03	*+sc*	+60.75	*+sc*
$H_{34}O_{10}C_8C_4$	+179.77	*+ap*	-177.17	*-ap*	-177.53	*-ap*	-174.41	*-ap*	- -	- -
$H_{36}N_{12}C_{11}C_9$	+45.99	*+sc*	+77.95	*+sc*	+44.28	*+sc*	+64.26	*+sc*	+38.37	*+sc*
$O_{37}C_{13}N_{12}C_{11}$	-2.71	*-sp*	+8.71	*+sp*	-5.99	*-sp*	+3.42	*+sp*	-3.44	*-sp*
$HN_{12}C_{11}C_9$	- -	- -	-38.74	*-sc*	- -	- -	-54.25	*-sc*	- -	- -
$HN_7C_4C_3$	- -	- -	- -	- -	-151.36	*-ap*	-149.19	*-ac*	- -	- -

* Conformational analyses using prefixes a = anti, s = syn, p = peri-planar, c = clinal, and + & - signs[8].

REFERENCES

1. (a) Knudsen ET, Rolinson GN, Absorption and excretion of a new antibiotic (BRL 1241), Br Med J, 1960; 2: 700. (b) Shechan JC, Henery-logan KR, The total and partial general synthesis of the penicillins, J Am Chem Soc,1962; 84: 2983-90.
2. (a) Wolfe S, Demain AL, Jensen SE, Westlake DWS, Enzymatic approach to synthesis of unnatural beta-lactams, Science, 1984; 226: 1386-92 (b) Kaiser GV, Kukolja S, Cephalosporins and Penicillins, E H Flynn, Ed. Academic Press, New York & London, 1972; 74-131.
3. (a) Fosker GR, Hardy KD, Nayler JHC, Seggary P, Siove ER, Derivatives of 6-aminopenicillanic acid. Part X. A non-enzymatic conversion of benzylpenicillin into semi-synthetic penicillins, J Chem Soc (C), 1971; 1917-22, (b) Chauvette RR, Pennington PA, Ryan CW, Cooper RDG, Jose FL, Wright IG, Van Heyningen EM, Huffman GW, Chemistry of cephalosporin Antibiotics. XXI. Coversion of penicillins to cephalexin, J Org Chem, 1971; 36: 1259-67.
4. Craig WA, Suh B, Protein binding and the antimicrobial effects: methods for determination of protein binding, In Antibiotics in Laboratory Medicine (Lorian V, ed.) 3rd edn. P367. Baltimore MD: Williams & Wilkins, 1991.
5. Allen JD, Roberts CE, Kirby WMM, Staphylococcal septicaemia treated with methicillin: Report of twenty-two cases, N Engl J Med., 1962; 266: 111.
6. (a) White A, Varga DT, Antistaphylococcal activity of sodium methicillin, Arch Intern Med, 1961; 108: 671; (b) Morin RB, Gorman M, Eds, Chemistry and Biology of Beta-lactam Antibiotics, Volumes 1-3, (Academic press, New York), 1982; (c) John H Block, John M Beak Jr, Wilson and Gisvold's Text Book of Organic Medicinal and Pharmaceutical Chemistry, 11th edn., (Lippincott Williams & Wilkins, NewYork), 2004.
7. (a) Dewar MJS, Zeobisch EG, Healy EF, Stewart JJP, AM1: a new general purpose quantum mechanical molecular model, J Am Chem Soc, 1985; 107: 3902-3909; (b) Stewart JJP, MOPAC, A general molecular orbital package, QCPE 455, 5th edn, 1988; (c) Coppola BP, Progress in practice: Organic chemistry in the introductory course, Chem Educator, 1997; 2(2), 1-8. Springer-verlag New York, INC.
8. (a) Rajeshwar Rao Bojja, AM1 study on the conformations of 6-aminopenicillanic acid, Indian J chem, 2002; 41B: 1697-1701; (b) Rajeshwar Rao B, Linga murthy S, AM1 study on the conformations of zwitterions of 6-aminopenicillanic acid (6-APA), Indian J chem, 2006; 45B: 1250-1253.
9. (a). B. Rajeshwar Rao, D. Chandra Sekhar Rao & Ch. Sanjeeva Reddy, AM1 study of the electronic structure of zwitterions of Ampicillin., Indian J. Chem, 2013; 52B (1): 164-168; (b). Bojja Rajeshwar Rao, D. Chandra Sekhar Rao & Ch. Sanjeeva Reddy, AM1 study of the Electronic structure of 7-aminodeacetoxycephalosporanic acid (7-

ADCA), Indian J. Chem., 2012; 51B(1): 307-312.
10. (a) Pople JA, Beveridge DL, Approximate molecular orbital theory, (McGraw-Hill, New York) 1970; (b) Fleming I, Frontier Orbital and Organic Chemical Reactions, (Wiley-Interscience, New York), 1976.
11. Woodward RB, Hoffmann R, The conservation of orbital symmetry, (Academic press, Inc, New York) 1970.
12. Paperno TYA, Pozdnyakov VP, Smirnova AA, Elagin LM, Physico-Chemical Laboratory Techniques in Organic and Biological Chemistry (Translated from Russian by Oleg Glebov), (MIR Publishers, Moscow) 1979; 171.
13. Saltek N, Abbasogulu R, Ikizler A, A quantum-chemical study on 3, 3'-bi(1H-1,2,4-triazole), Acta chim hung, Models in chemistry, 1996; 133: 43-51.