



## PM3 CONFORMATIONAL ANALYSIS OF THE (3S,5R,6R)-6 ACETYLAMIDOPENICILLANIC ACID. II. ELECTRONIC PROPERTIES<sup>1</sup>

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A conformational analysis with the semiempirical PM3 method was performed for the (3S,5R,6R)-6-acetylamidopenicillanic acid. 47 distinct conformers were found within 11.36 kcal/mol. Dependence of some electronic properties of these conformers (HOMO and LUMO energies, dipole moment,  $\nu_{\min}$  frequency of the minimum vibration,  $\nu_{\max}$  frequency of the maximum vibration, zero point vibration energy (ZPVE) and charge densities on atoms S1, N4, N14, O8, O12, O13 and O17) with respect to some geometrical characteristics (pseudochirality of the N14 atom, the *syn-anti* arrangement of the O17 and H28 atoms of the amidic group and the three puckering classes of the thiazolidinic ring (noted with a, b, c)) is discussed.

### INTRODUCTION

The (3S,5R,6R)-6-acetylamidopenicillanic acid is one of the simplest antibacterial compounds of the penicillin class. Its molecular structure contains two fused rings (a four-membered  $\beta$ -lactamic ring and a five-membered thiazolidinic ring) and three chiral centers,<sup>1</sup> marked with an asterisk in Fig. 1. These chiral centers generate  $2^3=8$  diastereoisomers: 3R,5R,6R, 3S,5R,6R, 3R,5S,6R, 3R,5R,6S,

3S,5S,6R, 3S,5R,6S, 3R,5S,6S, 3S,5S,6S.<sup>2</sup> The 3S,5R,6R diastereoisomer is the natural product. As a model for penicillin we have chosen the most simple penicillin: (3S,5R,6R)-6-acetylamidopenicillanic acid. In this paper we present an analysis of the dependence of the electronic properties of the (3S,5R,6R)-6-acetylamido-penicillanic acid on the geometrical characteristics of the conformers.

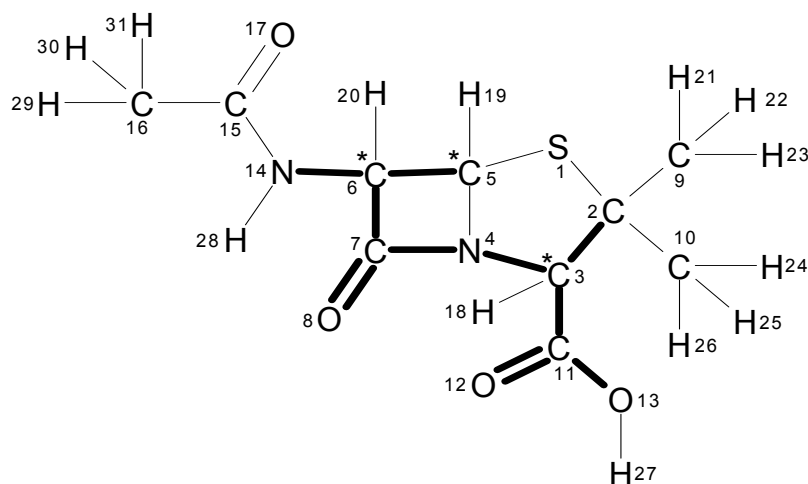


Fig. 1 – Atom numbering in the (3S,5R,6R)-6-acetylamidopenicillanic acid (\* marks the three chiral atoms: C3, C5 and C6).\*

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The same classification criteria as those used previously in the analysis of the geometrical properties are maintained: 1) pseudochirality (R,S) of the N14 atom measured by the “conicity” of the improper angle 6-15-28-1<sup>4</sup>, 2) *syn-anti* arrangement of the O17 and H28 atoms of the amidic group, and 3) the puckering classes of the thiazolidinic ring measured by the 5-1-2-3 and 5-4-3-2 dihedral angles (noted with a, b, c).<sup>1,3</sup>

## METHODS

Gas phase equilibrium geometry of conformers was obtained by semiempirical PM3 calculations using the Polak-Ribiere conjugate gradient algorithm for geometry optimization.<sup>4,5</sup> Stop criteria were: SCF convergence of 10<sup>-5</sup> and RMS gradient of 10<sup>-2</sup> kcal/Å·mol.<sup>5,6</sup> Conformational search was performed by varying (with steps of 15°) the 2-3-11-12, 5-6-14-15 and 6-14-15-16 dihedrals in the range 0 ±180°. Energy criterion was set to 30 kcal/mol above the minimum energy conformer. Calculations have been performed with

the Conformational Search module,<sup>7</sup> from the HyperChem7.52 package.<sup>5</sup>

## RESULTS AND DISCUSSION

The semiempirical PM3 method gives 47 conformers,<sup>1,7</sup> of which: 1) 23 and 24 had (R) and (S) pseudochirality at the N14 atom respectively; 2) 24 and 23 had *syn* and *anti* arrangement of the H28-N14 atoms respectively; 3) 20 conformers in class a, 16 conformers in class b and 11 conformers in class c of puckering (class a: the 5-1-2-3 and 5-4-3-2 dihedrals vary between 8.67° and 12.55° and between 13.82° and 19.58° respectively; class b: the 5-1-2-3 and 5-4-3-2 dihedrals vary between 14.56° and 21.12° and between 14.61° and 18.74° respectively; and class c: the 5-1-2-3 and 5-4-3-2 dihedrals vary between -1.09° and -7.33° and between 5.64° and 12.55° respectively).

In Table 1 these conformers are displayed in ascending order of formation enthalpies. The obtained results for some structural properties (**IP**<sub>1</sub>, **EA**<sub>1</sub>, **μ**) are presented also in Table 1.

Table 1

Numbering of the conformers of the (3S,5R,6R)-6-acetylamidopenicillanic acid arranged in the ascending order of the formation enthalpies computed with PM3 and some of their energetic, structural and electronic data

Energetic order	$\Delta H_{\text{form}}$ (kcal/mol)	HOMO (eV)	LUMO (eV)	$\mu$ (Debye)	Chirality N(14)	Position 17, 28	Vibrations (cm <sup>-1</sup> )		ZPVE (kcal/mol)
							$\nu_{\text{min}}$	$\nu_{\text{MAX}}$	
01pm3	-143.995	-9.745	-0.472	1.669	R	<i>syn</i>	29.14	3851.49	150.19
02 pm3	-143.978	-9.780	-0.472	4.623	R	<i>anti</i>	29.27	3851.90	149.65
03 pm3	-143.977	-9.779	-0.472	4.621	R	<i>anti</i>	29.10	3851.76	149.65
04 pm3	-143.501	-9.817	-0.499	3.922	R	<i>anti</i>	29.30	3847.92	149.65
05 pm3	-143.501	-9.817	-0.499	3.928	R	<i>anti</i>	29.30	3847.92	149.65
06 pm3	-143.430	-9.783	-0.500	2.138	R	<i>syn</i>	29.12	3847.96	150.20
07 pm3	-143.368	-9.726	-0.433	1.020	R	<i>syn</i>	30.46	3852.06	150.31
08 pm3	-143.363	-9.771	-0.446	4.803	R	<i>anti</i>	36.19	3851.97	149.77
09 pm3	-143.117	-9.658	-0.435	1.782	S	<i>syn</i>	32.92	3851.68	150.02
10 pm3	-143.116	-9.656	-0.433	1.776	S	<i>syn</i>	32.53	3851.48	150.02
11 pm3	-142.921	-9.812	-0.474	3.789	R	<i>anti</i>	36.70	3847.69	149.79
12 pm3	-142.831	-9.766	-0.462	2.130	R	<i>syn</i>	31.58	3847.64	150.32
13 pm3	-142.756	-9.649	-0.402	4.619	S	<i>anti</i>	31.36	3851.77	149.51
14 pm3	-142.746	-9.732	-0.443	1.098	R	<i>syn</i>	29.56	3851.74	150.02
15 pm3	-142.718	-9.686	-0.466	1.866	S	<i>syn</i>	30.89	3851.55	149.72
16 pm3	-142.712	-9.630	-0.371	1.031	S	<i>syn</i>	23.93	3851.92	150.26
17 pm3	-142.564	-9.694	-0.463	2.374	S	<i>syn</i>	33.58	3847.55	150.02
18 pm3	-142.563	-9.692	-0.460	2.385	S	<i>syn</i>	32.14	3847.86	150.02
19 pm3	-142.289	-9.682	-0.425	3.754	S	<i>anti</i>	31.15	3848.15	149.51
20 pm3	-142.200	-9.771	-0.472	2.279	R	<i>syn</i>	29.77	3847.92	150.03
21 pm3	-142.194	-9.652	-0.394	1.134	S	<i>syn</i>	31.34	3851.89	149.96
22 pm3	-142.192	-9.630	-0.357	4.796	S	<i>anti</i>	34.10	3852.13	149.69
23 pm3	-142.176	-9.670	-0.399	2.364	S	<i>syn</i>	23.86	3847.74	150.26
24 pm3	-142.165	-9.724	-0.495	2.364	S	<i>syn</i>	31.20	3847.79	149.78
25 pm3	-141.747	-9.669	-0.383	3.715	S	<i>anti</i>	34.38	3847.98	149.70
26 pm3	-141.661	-9.693	-0.423	2.284	S	<i>syn</i>	31.86	3847.72	149.96
27 pm3	-139.455	-9.567	-0.298	3.420	S	<i>anti</i>	21.19	3852.54	149.76
28 pm3	-139.384	-9.528	-0.236	3.345	S	<i>anti</i>	35.39	3852.67	150.06

29 pm3	-139.008	-9.715	-0.529	2.713	S	<i>syn</i>	24.98	3851.33	150.21
30 pm3	-138.880	-9.597	-0.320	4.769	S	<i>anti</i>	20.61	3848.38	149.75
31 pm3	-138.802	-9.562	-0.262	4.849	S	<i>anti</i>	36.09	3848.50	150.05
32 pm3	-138.765	-9.823	-0.566	4.716	S	<i>anti</i>	40.62	3851.78	150.15
33 pm3	-138.495	-9.744	-0.550	1.974	S	<i>syn</i>	25.10	3847.48	150.22
34 pm3	-138.285	-9.861	-0.594	4.068	S	<i>anti</i>	41.27	3847.62	150.17
35 pm3	-138.209	-9.289	-0.070	3.208	R	<i>anti</i>	20.59	3852.99	149.45
36 pm3	-138.157	-9.270	-0.026	3.471	R	<i>anti</i>	35.07	3852.59	149.73
37 pm3	-137.962	-9.634	-0.471	2.957	R	<i>syn</i>	20.40	3851.26	149.87
38 pm3	-137.641	-9.317	-0.089	4.576	R	<i>anti</i>	20.01	3848.39	149.45
39 pm3	-137.640	-9.317	-0.089	4.576	R	<i>anti</i>	20.15	3848.51	149.45
40 pm3	-137.638	-9.317	-0.089	4.577	R	<i>anti</i>	20.02	3848.51	149.45
41 pm3	-137.597	-9.305	-0.051	4.817	R	<i>anti</i>	35.82	3848.64	149.73
42 pm3	-137.450	-9.659	-0.489	2.645	R	<i>syn</i>	19.62	3847.03	149.85
43 pm3	-136.642	-9.683	-0.485	2.968	R	<i>syn</i>	47.95	3847.17	150.30
44 pm3	-135.386	-9.505	-0.189	3.853	S	<i>anti</i>	43.08	3852.95	149.83
45 pm3	-134.953	-9.544	-0.216	2.591	S	<i>syn</i>	43.85	3848.29	149.71
46 pm3	-133.185	-9.503	-0.296	2.152	R	<i>syn</i>	18.58	3852.10	149.71
47 pm3	-132.639	-9.541	-0.317	3.248	R	<i>syn</i>	20.81	3848.18	149.69

To be sure that the obtained conformers are minima on the potential energy surface (PES) and not transition states, for each conformer a normal vibration calculation was performed. All vibrations were positive for all conformers. The wavelength numbers:  $\nu_{\min}$  of the minimum vibration,  $\nu_{\max}$  of the maximum vibration and **ZPVE** zero point vibration energy are given in Table 1. A gap of

11.356 kcal/mol resulted between the lowest energy (01pm3) conformer and the highest energy (47pm3) one.<sup>1</sup>

The charge densities for some atoms important for biological activity: S1, N4, N14, O8, O12, O13, O17, of the conformers resulted from PM3 calculations, are presented in Table 2.

Table 2

Charge densities on some atoms of the conformers of the (3S,5R,6R)-6-acetylamidopenicillanic acid arranged by the ascending variation of the formation enthalpies computed with PM3

Energetic order	Net charge on atom (e)						
	S(1)	N(4)	N(14)	O(8)	O(12)	O(13)	O(17)
01 pm3	0.003	-0.109	-0.044	-0.250	-0.364	-0.307	-0.347
02 pm3	-0.016	-0.112	-0.037	-0.251	-0.362	-0.306	-0.349
03 pm3	-0.016	-0.112	-0.037	-0.251	-0.362	-0.308	-0.349
04 pm3	-0.018	-0.112	-0.036	-0.248	-0.389	-0.281	-0.350
05 pm3	-0.018	-0.112	-0.036	-0.248	-0.389	-0.281	-0.350
06 pm3	0.001	-0.110	-0.043	-0.248	-0.387	-0.282	-0.346
07 pm3	0.012	-0.106	-0.045	-0.245	-0.360	-0.309	-0.348
08 pm3	-0.003	-0.109	-0.042	-0.247	-0.359	-0.310	-0.348
09 pm3	0.007	-0.105	-0.033	-0.257	-0.363	-0.307	-0.354
10 pm3	0.007	-0.105	-0.033	-0.257	-0.363	-0.307	-0.354
11 pm3	-0.005	-0.109	-0.041	-0.245	-0.390	-0.278	-0.348
12 pm3	0.010	-0.106	-0.045	-0.242	-0.388	-0.279	-0.347
13 pm3	-0.005	-0.109	-0.028	-0.258	-0.362	-0.308	-0.351
14 pm3	0.011	-0.105	-0.042	-0.244	-0.360	-0.309	-0.345
15 pm3	0.007	-0.105	-0.043	-0.258	-0.363	-0.307	-0.351
16 pm3	0.019	-0.102	-0.037	-0.252	-0.360	-0.309	-0.354
17 pm3	0.005	-0.106	-0.033	-0.254	-0.387	-0.282	-0.353
18 pm3	0.005	-0.106	-0.033	-0.254	-0.387	-0.282	-0.353
19 pm3	-0.006	-0.109	-0.028	-0.256	-0.389	-0.281	-0.351
20 pm3	0.009	-0.106	-0.042	-0.242	-0.388	-0.270	-0.345
21 pm3	0.020	-0.102	-0.042	-0.253	-0.360	-0.309	-0.351
22 pm3	0.011	-0.106	-0.033	-0.255	-0.358	-0.310	-0.350
23 pm3	0.017	-0.103	-0.037	-0.249	-0.388	-0.279	-0.353
24 pm3	0.006	-0.106	-0.043	-0.255	-0.386	-0.282	-0.351
25 pm3	0.009	-0.106	-0.033	-0.253	-0.390	-0.278	-0.350
26 pm3	0.018	-0.103	-0.042	-0.250	-0.388	-0.279	-0.350
27 pm3	-0.028	-0.110	-0.020	-0.237	-0.368	-0.308	-0.360
28 pm3	-0.025	-0.107	-0.012	-0.240	-0.369	-0.308	-0.365

29 pm3	-0.002	-0.107	-0.029	-0.260	-0.362	-0.308	-0.348
30 pm3	-0.029	-0.111	-0.020	-0.235	-0.391	-0.283	-0.359
31 pm3	-0.027	-0.108	-0.013	-0.238	-0.391	-0.284	-0.364
32 pm3	0.007	-0.110	-0.080	-0.241	-0.359	-0.309	-0.302
33 pm3	-0.003	-0.108	-0.030	-0.257	-0.387	-0.282	-0.348
34 pm3	0.005	-0.111	-0.080	-0.238	-0.388	-0.279	-0.302
35 pm3	-0.008	-0.104	-0.010	-0.253	-0.369	-0.309	-0.362
36 pm3	-0.007	-0.102	-0.003	-0.253	-0.369	-0.308	-0.369
37 pm3	0.003	-0.104	-0.021	-0.263	-0.362	-0.309	-0.358
38 pm3	-0.009	-0.105	-0.011	-0.251	-0.393	-0.283	-0.361
39 pm3	-0.009	-0.106	-0.011	-0.251	-0.399	-0.283	-0.361
40 pm3	-0.009	-0.105	-0.011	-0.251	-0.393	-0.283	-0.361
41 pm3	-0.008	-0.103	-0.004	-0.251	-0.392	-0.283	-0.367
42 pm3	0.002	-0.105	-0.022	-0.261	-0.388	-0.281	-0.357
43 pm3	0.007	-0.100	-0.022	-0.245	-0.391	-0.272	-0.350
44 pm3	0.011	-0.104	-0.107	-0.261	-0.359	-0.311	-0.321
45 pm3	0.008	-0.104	-0.107	-0.258	-0.393	-0.276	-0.322
46 pm3	-0.004	-0.115	-0.077	-0.241	-0.364	-0.308	-0.314
47 pm3	-0.006	-0.115	-0.077	-0.238	-0.390	-0.281	-0.314

In the previous paper we have shown that the best fitting with an experimental geometry is that of conformer 04pm3.<sup>1</sup>

### Analysis of some energetic properties

After an analysis of the HOMO and LUMO energies (Table 1) we cannot find a differentiation of the conformers with respect to the following geometric properties: pseudochirality, *syn-anti* arrangement or the puckering class of the thiazolidinic ring (a, b, c).<sup>1</sup> For comparison of electronic properties their average values were calculated.

The average values of the HOMO and LUMO energy with respect to the (R, S) pseudochirality are:

$$\begin{aligned} \text{HOMO(R)} &= -9.615 \pm 0.206 \text{eV}, \\ \text{LUMO(R)} &= -0.353 \pm 0.180 \text{eV}, \\ \text{HOMO(S)} &= -9.660 \pm 0.085 \text{eV}, \\ \text{LUMO(S)} &= -0.402 \pm 0.109 \text{eV}. \end{aligned}$$

The average values and SD show that HOMO and LUMO energies are not statistically distinct and they do not depend on pseudochirality.

The HOMO and LUMO energy average values calculated with respect to the *syn-anti* criterion are:

$$\begin{aligned} \text{HOMO}(\textit{syn}) &= -9.679 \pm 0.072 \text{eV}, \\ \text{LUMO}(\textit{syn}) &= -0.436 \pm 0.075 \text{eV}, \\ \text{HOMO}(\textit{anti}) &= -9.594 \pm 0.204 \text{eV}, \\ \text{LUMO}(\textit{anti}) &= -0.318 \pm 0.181 \text{eV}. \end{aligned}$$

Compared to *anti*, the *syn* conformers have the HOMO energy average values lower and the LUMO energy values lower. Even if they are influenced by the *syn-anti* arrangement, the differences and SD of the HOMO and LUMO energies, respectively, are statistically distinct.

The HOMO and LUMO energy average values calculated with respect to the puckering criteria of the thiazolidinic cycle (a, b, c) are:

$$\begin{aligned} \text{HOMO(a)} &= -9.644 \pm 0.153 \text{eV}, \\ \text{LUMO(a)} &= -0.384 \pm 0.143 \text{eV}, \\ \text{HOMO(b)} &= -9.703 \pm 0.986 \text{eV}, \\ \text{LUMO(b)} &= -0.414 \pm 0.104 \text{eV}, \\ \text{HOMO(c)} &= -9.531 \pm 0.182 \text{eV}, \\ \text{LUMO(c)} &= -0.316 \pm 0.199 \text{eV}, \end{aligned}$$

There is a small difference between the average values of the three puckering classes.

In Fig. 2 is given the orbital distribution in HOMO and LUMO levels for the conformer 04pm3 with the best concordance with experimental geometry.<sup>1</sup> For all the conformers the contribution of the sulfur atom, S1 to the HOMO or LUMO level is the most important. For the HOMO level are of importance also the contributions of the  $\sigma$  bonds between C2-C9, C2-C10, C5-C6 and C5-H19 and of lower importance the local contributions of the N4, O8, N14 and O17 atoms. For the LUMO level are of importance the contributions of the C2 and C5 atoms and in lower measure those of the N3, C6, C7 and O8 atoms. Relatively narrow SD and the independence of HOMO and LUMO levels of all conformers on the three geometrical criteria could be explained by the dominant contribution of the sulfur S1 atom to these levels.

Analyzing the data in Table 1 one can observe that there is no influence of the pyramidalization on the dipole moment, as it results from the average values calculated with the R-S criterion:  $\mu(\text{R}) = 3.270 \pm 1.214$  Debye,  $\mu(\text{S}) = 3.028 \pm 1.209$ . Even if these average values seem different, due to their high standard deviation they are not statistically distinct.

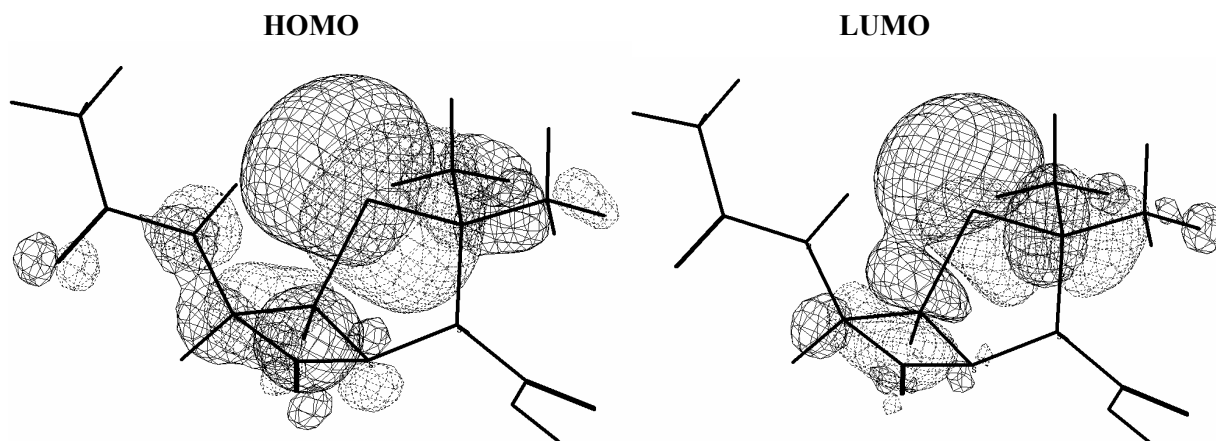


Fig. 2 – The weight of the sulphur (S1) atom in HOMO and LUMO of conformer 04pm3 (with the best concordance with experimental geometry) computed with PM3 (continuous lines – positive surface; dashed lines – negative surface).

Analyzing the data in Table 1 one can observe that there is no influence of the pyramidalization on the dipole moment, as it results from the average values calculated with the R-S criterion:  $\mu(\text{R}) = 3.270 \pm 1.214$  Debye,  $\mu(\text{S}) = 3.028 \pm 1.209$ . Even if these average values seem different, due to their high standard deviation they are not statistically distinct.

The influence of the *syn-anti* arrangement on the dipole moment is more obvious. The average values are  $\mu(\text{syn}) = 2.123 \pm 0.618$  Debye, and  $\mu(\text{anti}) = 4.209 \pm 0.556$  Debye. Statistically these average values are distinct.

The influence of the puckering on the dipole moment is proven by the average values for the three puckering classes (a,b,c):

$$\begin{aligned}\mu(\text{a}) &= 3.185 \pm 1.153 \text{ Debye} \\ \mu(\text{b}) &= 2.854 \pm 1.399 \text{ Debye} \\ \mu(\text{c}) &= 3.489 \pm 0.971 \text{ Debye}\end{aligned}$$

Even if these average values are different, due to the high dispersion they are not statistically distinct.

The influence of the *syn-anti* arrangement on the average values of the dipole moment calculated with respect to the (a,b,c) puckering classes are:

$$\begin{aligned}\mu(\text{syn},\text{a}) &= 2.175 \pm 0.464 \text{ Debye} \\ \mu(\text{anti},\text{a}) &= 4.195 \pm 0.752 \text{ Debye} \\ \mu(\text{syn},\text{b}) &= 1.770 \pm 0.675 \text{ Debye} \\ \mu(\text{anti},\text{b}) &= 4.249 \pm 0.502 \text{ Debye} \\ \mu(\text{syn},\text{c}) &= 2.572 \pm 0.421 \text{ Debye} \\ \mu(\text{anti},\text{c}) &= 4.188 \pm 0.648 \text{ Debye}\end{aligned}$$

From these data, for each puckering class there is an evident differentiation of the dipole moments function of the *syn-anti* arrangement.

As expected, these average values prove that for all conformers the dipole moment is strongly influenced by the *syn-anti* spatial arrangement of the O17 and H28 atoms of the exocyclic amidic group.

The minimum vibration frequency,  $\nu_{\text{min}}$ , is a backbone vibration and all atoms are implicated in it through their out-of-the-plane vibration (Fig. 3). The  $\nu_{\text{min}}$  average value is of  $30.126 \pm 7.084 \text{ cm}^{-1}$ , with a large dispersion, which proves the dependence of this vibration on the conformation. The maximum vibration frequency,  $\nu_{\text{MAX}}$ , is an elongation vibration implicated only in the bond between the O13 atom and the H27 atom. Its average value is  $3849.83 \pm 2.09 \text{ cm}^{-1}$ . Its dispersion is low, which proves its non-dependence on the conformation. If the dependence of these vibrations on R-S pseudochirality, on the *syn-anti* arrangement and on the puckering classes are taken into account, the following average values are obtained:

$$\begin{aligned}\nu_{\text{min}}(\text{R}) &= 28.20 \pm 7.38 \text{ cm}^{-1} \\ \nu_{\text{MAX}}(\text{R}) &= 3849.71 \pm 2.10 \text{ cm}^{-1} \\ \nu_{\text{min}}(\text{S}) &= 31.98 \pm 6.41 \text{ cm}^{-1} \\ \nu_{\text{MAX}}(\text{S}) &= 3849.95 \pm 2.11 \text{ cm}^{-1} \\ \nu_{\text{min}}(\text{syn}) &= 30.06 \pm 7.22 \text{ cm}^{-1} \\ \nu_{\text{MAX}}(\text{syn}) &= 3849.54 \pm 2.04 \text{ cm}^{-1} \\ \nu_{\text{min}}(\text{anti}) &= 30.20 \pm 7.10 \text{ cm}^{-1} \\ \nu_{\text{MAX}}(\text{anti}) &= 3850.14 \pm 2.14 \text{ cm}^{-1} \\ \nu_{\text{min}}(\text{a}) &= 30.64 \pm 4.44 \text{ cm}^{-1} \\ \nu_{\text{MAX}}(\text{a}) &= 3849.97 \pm 2.01 \text{ cm}^{-1} \\ \nu_{\text{min}}(\text{b}) &= 33.91 \pm 6.11 \text{ cm}^{-1} \\ \nu_{\text{MAX}}(\text{b}) &= 3849.94 \pm 2.21 \text{ cm}^{-1} \\ \nu_{\text{min}}(\text{c}) &= 23.69 \pm 8.27 \text{ cm}^{-1} \\ \nu_{\text{MAX}}(\text{c}) &= 3849.42 \pm 2.19 \text{ cm}^{-1}\end{aligned}$$

Comparing these average values one observes that the  $\nu_{\text{min}}$  backbone vibration depends on the puckering classes, but is not influenced by

pseudochirality and the *syn-anti* arrangement. At the same time the  $\nu_{\text{MAX}}$  vibration is not influenced neither by the pseudochirality, the *syn-anti* arrangement, nor by the puckering classes. We can generalize that independent of the conformer the vibrations of the functional groups are not

influenced by the three factors: pseudochirality, *syn-anti* arrangement and puckering classes, while the vibrations implicating the backbone depend on the puckering classes and do not depend on pseudochirality and *syn-anti* arrangement.

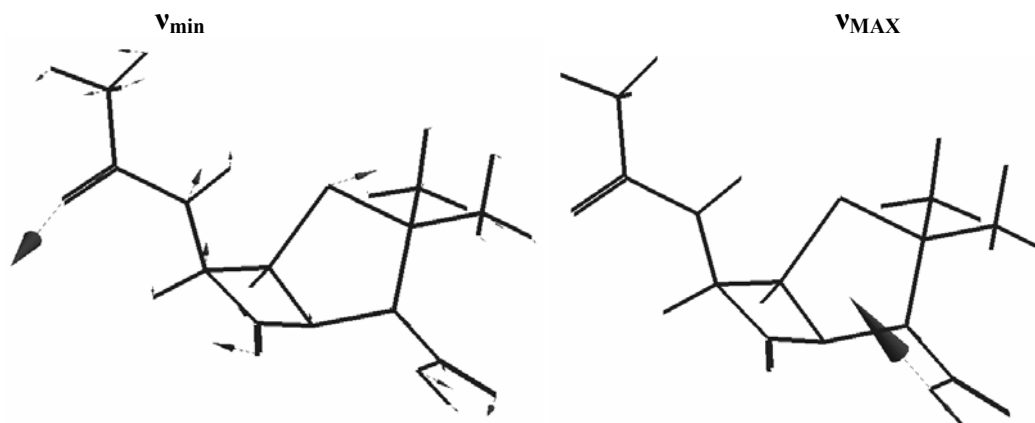


Fig. 3 – Movement vectors proportional with force constants for vibration  $\nu_{\text{min}}$  and  $\nu_{\text{MAX}}$  of conformer 04pm3 computed with PM3.

The average values of the puckering classes calculated for the backbone vibration frequencies  $\nu_{\text{min}}$ , and  $\nu_{\text{MAX}}$ , with respect to the R-S pseudochirality or the *syn-anti* arrangement are:

$$\begin{aligned} \nu_{\text{min}}(\text{a,R}) &= 28.55 \pm 5.35 \text{ cm}^{-1} \\ \nu_{\text{MAX}}(\text{a,R}) &= 3850.05 \pm 2.05 \text{ cm}^{-1} \\ \nu_{\text{min}}(\text{a,S}) &= 32.73 \pm 1.81 \text{ cm}^{-1} \\ \nu_{\text{MAX}}(\text{a,S}) &= 3849.90 \pm 2.07 \text{ cm}^{-1} \\ \nu_{\text{min}}(\text{b,R}) &= 32.38 \pm 3.23 \text{ cm}^{-1} \\ \nu_{\text{MAX}}(\text{b,R}) &= 3849.84 \pm 2.29 \text{ cm}^{-1} \\ \nu_{\text{min}}(\text{b,S}) &= 34.83 \pm 7.34 \text{ cm}^{-1} \\ \nu_{\text{MAX}}(\text{b,S}) &= 3850.00 \pm 2.28 \text{ cm}^{-1} \\ \nu_{\text{min}}(\text{c,R}) &= 24.11 \pm 10.52 \text{ cm}^{-1} \\ \nu_{\text{MAX}}(\text{c,R}) &= 3849.12 \pm 2.20 \text{ cm}^{-1} \\ \nu_{\text{min}}(\text{c,S}) &= 22.97 \pm 2.40 \text{ cm}^{-1} \\ \nu_{\text{MAX}}(\text{c,S}) &= 3849.93 \pm 2.39 \text{ cm}^{-1} \\ \nu_{\text{min}}(\text{a,syn}) &= 29.09 \pm 5.19 \text{ cm}^{-1} \\ \nu_{\text{MAX}}(\text{a,syn}) &= 3849.76 \pm 2.01 \text{ cm}^{-1} \\ \nu_{\text{min}}(\text{a,anti}) &= 32.19 \pm 3.05 \text{ cm}^{-1} \\ \nu_{\text{MAX}}(\text{a,anti}) &= 3850.18 \pm 2.10 \text{ cm}^{-1} \\ \nu_{\text{min}}(\text{b,syn}) &= 30.69 \pm 5.81 \text{ cm}^{-1} \\ \nu_{\text{MAX}}(\text{b,syn}) &= 3849.66 \pm 2.14 \text{ cm}^{-1} \\ \nu_{\text{min}}(\text{b,anti}) &= 38.05 \pm 3.57 \text{ cm}^{-1} \\ \nu_{\text{MAX}}(\text{b,anti}) &= 3850.30 \pm 2.41 \text{ cm}^{-1} \\ \nu_{\text{min}}(\text{c,syn}) &= 27.61 \pm 11.65 \text{ cm}^{-1} \\ \nu_{\text{MAX}}(\text{c,syn}) &= 3848.85 \pm 2.23 \text{ cm}^{-1} \\ \nu_{\text{min}}(\text{c,anti}) &= 20.43 \pm 0.46 \text{ cm}^{-1} \\ \nu_{\text{MAX}}(\text{c,anti}) &= 3849.89 \pm 2.23 \text{ cm}^{-1} \end{aligned}$$

Comparing these data it results that both pseudochirality and *syn-anti* arrangement do not influence the  $\nu_{\text{min}}$  and  $\nu_{\text{MAX}}$  vibration frequencies.

Based on the above results we can conclude that both the backbone vibrations as well as the vibrations of the functional groups do not depend on the *syn-anti* arrangement or the pseudochirality. But they depend significantly on the puckering class. For all the 47 conformers optimized with the PM3 method, the nature of these two types of vibrations do not change. The equilibrium energy (Zero Point Vibrational Energy ZPVE) is relatively constant around the average value of  $149.876 \pm 0.262$  kcal/mol. The calculated average values for ZPVE using the *syn-anti* arrangement, R-S pseudochirality and puckering classes criteria lead to the conclusion that this energy does not change. ZPVE is a measure who does not depend significantly on the conformer's nature.

### Analysis of the electronic properties

The net charge on the S1 atom varies in relatively small limits, between 0.020 and -0.029. The average value for all conformers is  $q_{\text{S1}} = -0.001 \pm 0.013$ . The standard deviation is higher than the average value. To evidence the influence of geometrical properties on the S1 charge a statistical analysis was performed applying the three criteria: the influence of the R-S pseudochirality, the influence of the *syn-anti* arrangement and the influence of the three puckering classes. The obtained average values are

all almost zero and do not allow evidencing any dependence of the S1 atom charge on the three criteria:

$$\begin{aligned} q_{S1}(S) &= 0.001 \pm 0.015 R \\ q_{S1}(R) &= -0.003 \pm 0.009 \\ q_{S1}(anti) &= -0.009 \pm 0.012 \\ q_{S1}(syn) &= 0.007 \pm 0.007 \\ q_{S1}(a) &= -0.006 \pm 0.011 \\ q_{S1}(b) &= 0.010 \pm 0.007 \\ q_{S1}(c) &= -0.008 \pm 0.011 \end{aligned}$$

The net charges on the atoms:  $q_{N4}$ ,  $q_{N14}$ ,  $q_{O8}$ ,  $q_{O12}$ ,  $q_{O13}$ ,  $q_{O17}$  are all negative for all conformers (Table 2). Even if the two nitrogen atoms (N4, N14) are of the same type (amidic nitrogen), the N4 atom is strongly pyramidalized due to its steric vicinities in the two cycles, as can be seen in the experimental determined structure.<sup>8</sup> The N4 atom has a real pyramidalization, while N14 has a pyramidalization generated by the quantum chemical computation method. The N4 atom has a negative net charge significantly higher than the exocyclic N14 nitrogen atom. The average value of the charge is  $q_{N4} = -0.106 \pm 0.004$ , while  $q_{N14} = -0.037 \pm 0.023$ . The net charge on N4 varies in quite small limits, between -0.102 and -0.115, i.e., it can be considered almost constant and independent of the conformation, while the net charge on N14 varies in large limits between -0.004 and -0.107 and depends on the conformer geometry. For these two atoms (N4, N14) the average charges for the conformers with R and S pseudo-chirality are:

$$\begin{aligned} q_{N4}(R) &= -0.107 \pm 0.004; \\ q_{N14}(R) &= -0.033 \pm 0.021; \\ q_{N4}(S) &= -0.106 \pm 0.003; \\ q_{N14}(S) &= -0.042 \pm 0.026; \end{aligned}$$

For the conformers with *syn-anti* arrangement they are:

$$\begin{aligned} q_{N4}(syn) &= -0.106 \pm 0.004; \\ q_{N14}(syn) &= -0.045 \pm 0.021; \\ q_{N4}(anti) &= -0.108 \pm 0.003; \\ q_{N14}(anti) &= -0.030 \pm 0.024; \end{aligned}$$

and for the three puckering classes the average charges for N4 and N14 are:

$$\begin{aligned} q_{N4}(a) &= -0.108 \pm 0.004; \\ q_{N14}(a) &= -0.035 \pm 0.019; \\ q_{N4}(b) &= -0.106 \pm 0.003 \\ q_{N14}(b) &= -0.053 \pm 0.025; \\ q_{N4}(c) &= -0.106 \pm 0.003; \\ q_{N14}(c) &= -0.019 \pm 0.009; \end{aligned}$$

These average charges show that the net charges for the two amidic nitrogen atoms do not

differ significantly from the average values of all conformers. Even if for the N14 atom the values are different between R and S and *syn* and *anti*, because of the high standard deviations their charges are not statistically distinct. For N14 there is a certain dependence on the puckering classes. The lowest charge on N14 is for class c, followed by class a, and the most negative charge is obtained for the puckering class b. Taking the puckering classes into account together with *syn-anti* or R-S criterion one does not observe any supplementary regularity. From this data one can draw the conclusion that the net charges on the N4 atom are relatively constant and independent on the conformer geometry. For the N14 atom the net charge depends obviously on the conformer geometry. This fact is demonstrated by the high dispersion of the values. There is a certain regularity, but only function of the puckering class.

The average value of the net charge on the O8  $\beta$ -lactamic atom:  $q_{O8} = -0.259 \pm 0.007$  is lower than the average values of the net charges for the other oxygen atoms:  $q_{O12} = -0.377 \pm 0.014$ ,  $q_{O13} = -0.293 \pm 0.015$ ,  $q_{O17} = -0.348 \pm 0.015$ . The  $q_{O8}$  net charge is relatively constant, independent on the conformer geometry and varies between -0.235 and -0.263. On the atoms O12, O13 and O17 the net charge varies between -0.359 and -0.399, -0.270 and -0.310 and respectively -0.302 and -0.369. For these oxygen atoms the average values of the net charge taking the R and S pseudo-chirality into account, are:

$$\begin{aligned} q_{O8}(R) &= -0.249 \pm 0.006 \\ q_{O8}(S) &= -0.251 \pm 0.008 \\ q_{O12}(R) &= -0.379 \pm 0.014 \\ q_{O12}(S) &= -0.375 \pm 0.014 \\ q_{O13}(R) &= -0.292 \pm 0.014 \\ q_{O13}(S) &= -0.295 \pm 0.014 \\ q_{O17}(R) &= -0.350 \pm 0.013 \\ q_{O17}(S) &= -0.347 \pm 0.017 \end{aligned}$$

Taking into account the *syn-anti* arrangement, the average charges are:

$$\begin{aligned} q_{O8}(syn) &= -0.251 \pm 0.007 \\ q_{O8}(anti) &= -0.249 \pm 0.007 \\ q_{O12}(syn) &= -0.376 \pm 0.014 \\ q_{O12}(anti) &= -0.378 \pm 0.015 \\ q_{O13}(syn) &= -0.292 \pm 0.015 \\ q_{O13}(anti) &= -0.295 \pm 0.014 \\ q_{O17}(syn) &= -0.344 \pm 0.015 \\ q_{O17}(anti) &= -0.352 \pm 0.015, \end{aligned}$$

and taking the three puckering classes into account the following average values are obtained:

$$\begin{aligned}
 q_{O8}(a) &= -0.250 \pm 0.007 \\
 q_{O8}(b) &= -0.248 \pm 0.005 \\
 q_{O8}(c) &= -0.251 \pm 0.009 \\
 q_{O12}(a) &= -0.376 \pm 0.013 \\
 q_{O12}(b) &= -0.374 \pm 0.015 \\
 q_{O12}(c) &= -0.382 \pm 0.014 \\
 q_{O13}(a) &= -0.295 \pm 0.013 \\
 q_{O13}(b) &= -0.293 \pm 0.017 \\
 q_{O13}(c) &= -0.291 \pm 0.014 \\
 q_{O17}(a) &= -0.350 \pm 0.014 \\
 q_{O17}(b) &= -0.340 \pm 0.018 \\
 q_{O17}(c) &= -0.357 \pm 0.005,
 \end{aligned}$$

Statistically, it results that the oxygen net charges do not differ significantly from the average values for all conformers. If the puckering classes and the *syn-anti* or R-S criteria are taken into account, there is also no regular pattern. The only exception is the N14 atom whose net charge depends on the conformer geometry. This means

that the conformational changes generated by the free rotation of the C3-C11, C6-N14 and N14-C15 in the conformers with different puckering classes do not influence significantly the charges, except for the N14 atom.

The lack of the influence of conformer geometry on this relatively constant distribution of the net atomic charges determines an isosurface of the electrostatic potential relatively similar for all conformers. This is illustrated by the isosurfaces of the electrostatic potential presented in Fig. 4, for the conformer 04pm3 with the best concordance with the experimental geometry and conformer 43pm3 with the worst concordance with the experimental geometry.<sup>1,8</sup> The isosurfaces of the electrostatic potential depend in a small degree on the spatial arrangement mode of the exocyclic groups attached to the N14 amidic atom.

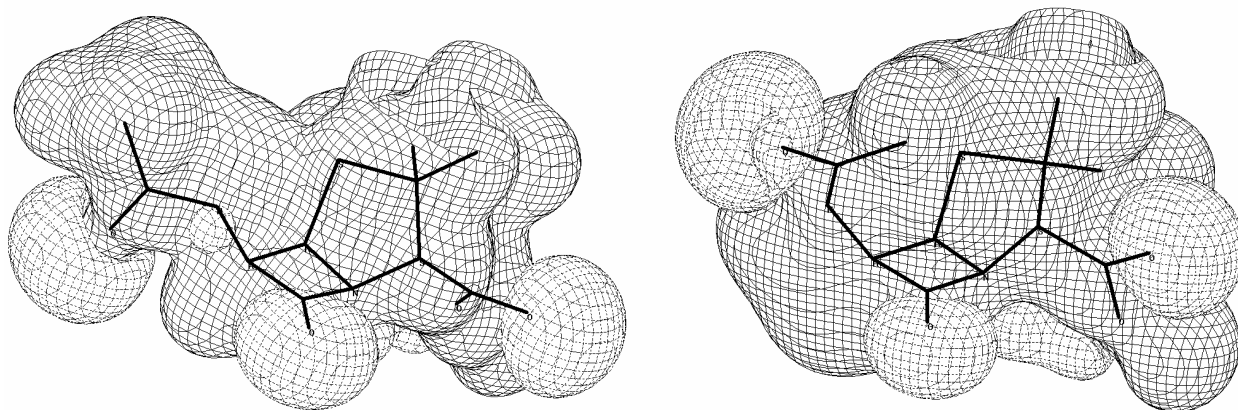


Fig. 4 – Isosurface of the electrostatic potential computed with PM3 for the contour value of 0.04, for conformers 04pm3 and 43pm3 (continuous lines – positive surface; dashed lines – negative surface).

## CONCLUSIONS

From the conformational analysis with the PM3 method it results that for all conformers the pseudo-chirality of the N14 atom does not influence significantly the HOMO and LUMO energies. On the contrary, the energy of these levels is influenced by the *syn-anti* arrangement of the exocyclic amidic group as well as by the puckering of the thiazolidinic ring.

From the analysis of the orbital contribution to the HOMO and LUMO levels, one can conclude that in these levels the S1 atom has the dominant contribution.

The dipole moment is influenced strongly by the *syn-anti* arrangement. The puckering slightly

influences the dipole moment, while the pseudo-chirality does not influence it at all.

From the vibration analysis one can draw the conclusion that both the backbone vibrations as well as the vibrations of the functional groups do not depend by the *syn-anti* arrangement or by the R-S pseudo-chirality, but depend significantly on the puckering. ZPVE does not depend significantly on the geometry of the conformer.

Analysing the average values of the net atomic charges one can conclude that the charge densities on the N4 atom are not dependent on conformation, they being almost constant. For the N14 atom the net density is dependent on the conformation and this is proven by the high dispersion of the values. There is a certain pattern



with the respect of the puckering, but such pattern was not observed for pseudochirality or the *syn-anti* arrangement. The constant charge distribution leads to an isosurface of the electrostatic potential relatively similar for all conformers.

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## REFERENCES

1. M. Mracec, M. Schulz, E. Şişu and M. Mracec, *Rev. Roum. Chim.*, **2008**, *53*, 847–858.
2. C. D. Nenişescu, "Chimie organică" Vol. 2, Editura Didactică and Pedagogică, Bucureşti, 1968, Ediția a 6-a, p. 629-630.
3. M. Schulz, M. Mracec, E. Şişu, and M. Mracec, *Rev. Roum. Chim.*, **2007**, *52*, 859-867.
4. a) M. J. S. Dewar, E. G. Zoebisch, E. F. Healy and J. J. P. Stewart, *J. Am. Chem. Soc.*, **1985**, *107*, 3902-3909; b) J. J. P. Stewart, *J. Comput. Chem.*, **1989**, *10*, 209-220, 221-264; c) J. J. P. Stewart, *J. Comput. Chem.*, **1991**, *12*, 320-334; d) J. J. P. Stewart, *J. Mol. Model.*, **2004**, *10*, 155-164.
5. \*\*\* HyperChem™, Release 7.52 for Windows, Copyright 2003, Hypercube, Inc, 1115 NW 4<sup>th</sup> Street, Gainesville, FL 32601, US.
6. I. N. Levine, "Quantum Chemistry", 5<sup>th</sup> Edition, Prentice Hall, Inc., Upper Saddle River, New Jersey 07458, 2000, Chap. 15, Chap. 17.
7. \*\*\* ChemPlus 1.6™, "Extension for HyperChem®, Molecular Modeling for Windows™", Hypercube, Inc., Gainesville, Florida, US, 1994, Chap. 8.
8. I. Csoregh and T.-B. Palm, *Acta Crystallogr., Sect. B: Struct. Crystallogr. Cryst. Chem.*, **1977**, *33*, 2169-2171; <http://www.middlebury.edu/~ch0337/atteridge/penicillin/fig1.gif>.

