



# Molecular Complexes of Amino Acid ( $\alpha$ -Alanine) and Their Manifestation in the Raman Spectra, *Ab initio* Calculations

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**Abstract:** The purpose of this work is to determine the role of hydrogen bonds in the crystallization of alanine, which includes carboxyl COOH and amino NH<sub>2</sub> groups, and the manifestation of these intermolecular interactions in the Raman spectra. FT-Raman spectra were analyzed and *ab initio* calculations by the RHF and B3LYP methods were performed in order to consider in more detail the possibility of the formation of aggregated intermolecular complexes in the crystalline state of  $\alpha$ -alanine. The intra-molecular and inter-molecular interactions, the dynamics of molecular groups and structural changes of  $\alpha$ -alanine have been studied by the method of Raman light scattering and *ab initio* calculations. *Ab initio* calculations have shown that the change in the Raman spectra is explained by the formation of several types of hydrogen bonds. These hydrogen bonds play a special role in the formation of the crystal structure of  $\alpha$ -alanine. In Raman spectra, the presence of a hydrogen bond between molecules is manifested in the form of asymmetry and splitting of vibrational bands. An analysis of the Raman spectra of alanine with water shows that with an increase in the strength of the hydrogen bond leads to increase in the bond energy in the OH group. The hydrogen atom in the N-H group of the alanine molecule is actively involved in the formation of a hydrogen bond. It was shown by *ab initio* calculations that dimeric, trimeric and other chain molecular complexes exist in alanine. The complexes formed due to the hydrogen NH<sub>2</sub> bond are weaker than those formed due to the OH bond. Upon the formation of a complex of alanine with water, the O-H vibration band shifts to the low-frequency side by 320 cm<sup>-1</sup> and 415 cm<sup>-1</sup>, respectively. It was shown that an intramolecular hydrogen bond is formed between the NH and CO groups. It was found that in the Raman spectra of O-H and N-H, the alanine bands in the region of 2900-3050 cm<sup>-1</sup> are complex and consist of several bands. The structure and connectivity of the bands is explained by the fact that the spectrum of the O-H bond consists of a symmetric valence band of vibrations and overtones of bending vibrations.

**Keywords:** Amino Acids, Alanine, Hydrogen Bonding, Raman Spectra, Molecular Aggregation, Aggregate Structure, Depolarization Ratio, *Ab Initio* Calculations

## 1. Introduction

The hydrogen bond among specific interactions is of particular interest due to the fact that it underlies the most subtle phenomena of life and plays an essential role in the functioning of molecules [1]. Studies of bonds of this type have been going on for many decades, and the development of the theory of hydrogen bonds was largely facilitated by the study of the vibrational spectra of compounds with hydrogen bonds. However, the nature and mechanisms of its formation have not been finally clarified [2].

Currently, more and more attention is attracted by aqueous solutions of amino acids, which are constituent components of proteins and play an important role in various biological and biochemical processes [1]. In addition, they are convenient model systems for studying the processes of intermolecular interaction. In this case, Raman spectra provide more complete information about the molecular structure for understanding more complex amino acid molecules [2, 3].

Amino acids are carboxylic acids in which the hydrogen atom in the radical is replaced by an amino group - (NH<sub>2</sub>)-(CH-R)-COOH. In this case, the amino acid residue R

determines the structure of a particular amino acid, if R-H we obtaine alanine, which is  $\text{NH}_2\text{-CH}(\text{CH}_3)\text{-COOH}$  (2aminopropanoic acid). Depending on the amino acid residue, alanine molecules can exist in the form of two mirror-symmetric (left and right) isomers [4].

In addition, alanine exists in two forms:  $\alpha$ -alanine is a structural element of proteins,  $\beta$ -alanine is a part of many biological compounds. Alanine has two centers that can form a hydrogen bond through the hydroxyl group as a proton donor and through the  $\text{NH}_2$  group as a proton acceptor. Alanin is of interest not only for fundamental research, but has extremely high practical significance.

Studies of the vibrational spectra of amino acids using Raman spectroscopy show that the formation of molecular complexes through intermolecular hydrogen bonds leads to a change in the spectral parameters of interacting molecules [4, 5]. In these spectral studies much attention is paid to the region of intramolecular interactions, in which the vibrational degrees of freedom of amino acid residues are manifested, as well as the vibrations of the C-H and N-H groups [3].

Literature data show that it is not reliably determined whether the regularities in frequency shifts, broadening, change in the integral intensity during the formation of hydrogen bonds are observed in vibrational spectra [1]. In this regard, the determination of the role of hydrogen bonds in the formation of molecular complexes, using the example of  $\alpha$ -alanine and the manifestation of these intermolecular interactions in the Raman spectra is very important.

An analysis of a number of experimental data shows that the Raman spectrum of the amino acid exhibits a line with a frequency of  $1650\text{ cm}^{-1}$  characteristic of the carboxyl group and intense lines with a frequency of  $3380\text{ cm}^{-1}$ , and for the amino group  $\text{NH}_2$  vibrations with a frequency of  $3340\text{ cm}^{-1}$ .

In this work, *ab initio* calculations were carried out in order to consider in more detail the possibility of the formation of aggregated intermolecular complexes in the liquid crystalline state of  $\alpha$ -alanine. To confirm the earlier assumptions [6-8] regarding the spectral manifestations of the existence of monomers and intermolecular complexes (dimer, trimer, tetramer, pentamer, etc.), *ab initio* quantum-chemical calculations of normal vibration spectra, band depolarization ratios and Raman activity were performed. Such calculations should complement the spectroscopic data and help to create a more complete picture of the structure of  $\alpha$ -alanine aggregates in the liquid and crystalline state.

### 3. Experimental Technique

The Raman spectra of alanine crystals were recorded on a Thermo Nicolet 6700 FTIR/FT-Raman spectrometer with a Raman attachment and OMNIC software. The spectra were taken at two excitation wavelengths  $\lambda=514\text{ nm}$  and  $\lambda=488\text{ nm}$  and are identical. Ranges were  $525\div4500\text{ cm}^{-1}$  with MCT-A detector (LN2 cooled), and  $375\div4500\text{ cm}^{-1}$  with DTGS TEC (RT) detector. The initial luminous flux in this case is characterized by high power and narrow directivity, which makes it possible to obtain a spectrum using 1–10 mg of the

substance. The sample can be injected either as a clear liquid or solution, or as a solid powder.

*Ab initio* calculations of the optimized structure of the  $\alpha$ -alanine molecule and its complex were carried out using the RHF, B3LYP, MP methods using the 6-31G ++ (d, p) Gaussian function basis for the monomer and other high molecular weight isolated clusters of alanine molecules [9].

Energy optimization was carried out for all internal coordinates of the objects under study. The calculation of the frequencies of normal vibrations is carried out by the software by analytical calculation of the second derivatives of the total energy by coordinates.

## 2. Results and Discussion

Figure 1 shows the Raman spectrum of  $\alpha$ -alanine in the region  $00\div4000\text{ cm}^{-1}$  is. It follows from the figure that the Raman spectrum of  $\alpha$ -alanine is a complex band. Analysis of the band complexity in the region  $2800\div3200\text{ cm}^{-1}$  allows understanding the mechanism of molecular complex formation. As can be seen from the figure, this complex band refers to symmetric and antisymmetric O-H and N-H vibrations of alanine molecules. There are various assumptions regarding the composition of this complex band [4, 10-12].

Analysis of the band shape in the  $2800\div3200\text{ cm}^{-1}$  region shows that the features of the observed O-H and N-H band of alanine vibrations can be easily explained if we assume that the observed band consists of several bands with different values of the depolarization ratio, which is in satisfactory agreement with the data [4, 5]. It is known that the process of vibration and translational motion of individual parts of alanine molecules is accompanied by the appearance of dipole moments of molecules [5].

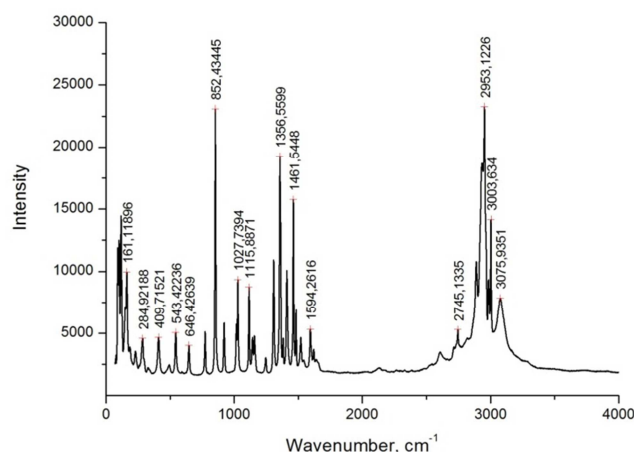


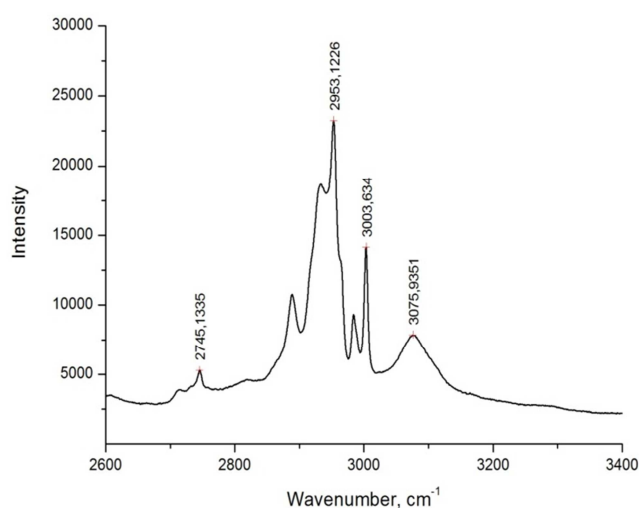
Figure 1. Raman spectra of  $\alpha$ -alanine crystals.

Polarized Raman spectra of single crystals of L-alanine and N-deuterated L-alanine were studied [12]. The results of experimental studies are given that a band is observed at  $2926\text{ cm}^{-1}$  in IR and at  $2923\text{ cm}^{-1}$  in Raman spectra, and this phenomenon is explained by the stretching of the CH group towards the  $\text{CH}_3$  group [13]. The corresponding theoretical

values are  $2955\text{ cm}^{-1}$  and  $2830\text{ cm}^{-1}$ . The other two bands related to CH vibrations with a frequency of  $2882\text{ cm}^{-1}$  and  $2768\text{ cm}^{-1}$  appear only in the Raman spectra. The band with a frequency of  $2604\text{ cm}^{-1}$  is observed in the IR spectrum and refers to the  $\text{NH}_3$  vibrations and this band is observed in the Raman spectra of alanine molecules.

For the case of alanine, the difference between the maxima of the O-H and N-H vibrations is about  $50\text{ cm}^{-1}$ . If we isolate it from the general band, then the bands of O-H and N-H vibrations of alanine will consist of symmetric and asymmetric vibrations with different depolarization ratios (Figure 2).

In Figure 2 it can be seen that the bands of O-H and N-H vibrations are complex, they consist of several bands in the region of  $2800\text{--}3200\text{ cm}^{-1}$ . In Raman spectra of alanine, as a rule, the scattering cross section of symmetric bands is larger than that of antisymmetric ones. The degrees of depolarization ratios of these bands in the Raman spectra are different: the high-frequency band (antisymmetric vibration) is depolarized, the low-frequency band (symmetric vibration) is polarized. The difference in band frequencies is about  $100\text{ cm}^{-1}$ . It should be noted that this value is significantly (almost two times) less than the difference in the frequencies of the components of the complex band.



**Figure 2.** Raman spectrum of the band related to O-H and N-H vibrations of  $\alpha$ -alanine crystals.

In our studies [14, 15] it was shown that the free O-H vibration refers to the line with a frequency of  $3455\text{ cm}^{-1}$ . During transitions in a condensed medium with respect to the hydrogen bond, this band shifts to the low-frequency sides and broadening ( $600\text{ cm}^{-1}$ ) of the bands is observed. The Raman spectra of these stretching vibrations are not symmetric and their shape strongly depends on temperature. Based on this, we can conclude that the structure and connectivity of this band is explained by the fact that the Raman spectrum of O-H vibrations consists of a symmetric stretching vibration  $\nu_1$  and an overtone of bending vibrations  $2\nu_2$ .

If we assume (Figure 2) that the broad band with a

frequency of  $3075.9\text{ cm}^{-1}$  belongs, respectively, to O-H vibrations (stretching and bending vibrations, then a wide band with a frequency of  $2953.1\text{ cm}^{-1}$ ) and antisymmetric (bending) vibrations of the  $\text{NH}_2$  group. It is known from the literature [16] that O-H vibrations correspond to a band with a frequency of  $3600\text{ cm}^{-1}$  and N-H vibrations correspond to a band with a frequency of  $3300\text{ cm}^{-1}$ .

In our case, with the appearance of a strong hydrogen bond, the O-H vibration band shifts by  $524.1\text{ cm}^{-1}$  and the N-H vibration band shifts by  $346.9\text{ cm}^{-1}$  to the low-frequency side and, accordingly, the half-width of these bands changes. As a result in the spectra of Raman scattering in this region there is a complex band consisting of four closely spaced bands.

It follows from this that alanine contains complex, long-lived molecular complexes. The presence of such a change is evidenced by spectroscopic data. The entire complex band in the range of  $2800\text{--}3300\text{ cm}^{-1}$  in the spectrum is due to the superposition of several bands corresponding to “symmetric” (weakly depolarized component) and “antisymmetric” (depolarized component) vibrations. The concepts of “symmetric” and “antisymmetric” vibrations refer to a single molecule of alanine, with the aggregation of molecules these concepts lose their meaning, but vibrations of this type should remain, the definition of the concept in the future should be understood with this remark in mind.

In this work, we carried out *ab initio* quantum-chemical calculations, optimization of the density functional theory method for  $\alpha$ -alanine molecules in order to further simulate complex aggregate states, provided that sufficiently accurate results are obtained in the shortest time.

In [13], a normal coordinate analysis was carried out for optically active intra- and inter-molecular vibrations of the L-alanine crystal. Large splits of factor groups observed for asymmetric deformation of  $\text{CH}_3$  and torsional modes of  $\text{CH}_3$  were well defined using a potential. Good agreement was obtained between the observed and calculated frequencies of lattice vibrations when adding terms due to stretching of the hydrogen bond and long-range Coulomb interaction with an intermolecular potential.

Raman spectra of all standard amino acids, analysis of various possible oscillations and their comparison under different conditions with changes in temperature or pressure are given in [17].

In [18, 19], the Raman study of the  $\text{NH}_3^+$  torsion regime in the amino acid L-alanine was carried out. The temperature dependence of this mode was observed in the temperature range  $110\text{--}350\text{ K}$ . One of the hydrogen bonds, oriented along the axis of the crystal cell, is insensitive to a decrease in temperature, while the other two are enhanced [19].

The Raman spectra of 13 amino acids, including L-alanine, were obtained in the open air using portable Raman spectrometers with laser excitation at  $785\text{ nm}$  [20].

The observed changes in the intensity are interpreted as due to the additional contribution of the charge transfer of the electronic excited state arising from the formation of hydrogen bonds. With a change in polarizations and temperature, the spectral bandwidth changes, respectively,

the relaxation time fits with the change in the movement of molecules with hydrogen bonds.

We have analyzed the literature data in order to be convinced that the above assumption is indeed true. For example, vibrational spectra were calculated using several calculation methods. Shown are infrared and Raman spectra with corresponding depolarization coefficients [21].

Intramolecular transfer of a proton from oxygen to nitrogen atoms in the amino acid alanine was studied by the methods HF/6-31G\*, B3LYP/6-31++G\*\* and MP2//6-31++G\*\*. Calculations show that the barrier height associated with the conformational change in alanine is greater than the proton transfer process [22]. For the alanine chain of the formed three and six molecular complexes, the potential energy of surfaces was investigated, and stable molecular clusters were determined [23]. An experimental measurement of the IR and Raman spectra of BSA was carried out and the vibrational spectra of the zwitterionic forms of 20 amino acids and their dipeptides were calculated [24]. The influence of anharmonicity and intermolecular interactions (IMI) on the vibrational spectra of amino acids is considered. The energy and frequencies of bond vibrations were calculated, a decrease in the frequency of the stretching vibration of the bond and an increase in the intensity of the Raman line were observed, for the stretching and deformation vibrations of the polar groups COO<sup>-</sup> and NH<sub>3</sub><sup>+</sup> in the case of ion-ion and ion-dipole IMW, a frequency shift is observed, which is  $\sim 5$ -80 cm<sup>-1</sup>, and the intensity changes by a factor of  $\sim 3$ -10 [24].

Various molecular forms of  $\alpha$ -alanine have been studied at the B3LYP/6-31++G\*\* level calculation using SCRF. In this case, the intramolecular proton transfer from oxygen to nitrogen atoms of  $\alpha$ -alanine and the vibrational spectrum were analyzed in various solvents [25].

It was shown that the hydrogen bond in amino acids is very important, but it is difficult to experimentally determine the energy of the intramolecular hydrogen bond. Because in amino acids there are 13 types of conformers for neutral alanine. Alanine exists in zwitterionic form in solid form. However, experimental studies have shown that there is only a neutral form of alanine in the gas phase [26].

The structure and intramolecular proton transfer of alanine radicals were studied; it was shown that the energy barrier in the process of proton transfer is about 6 kcal / mol [27]. Using *ab initio* calculations for  $\alpha$ -alanine, exact geometric structures and relative energies were obtained for different structures of molecular aggregates [28].

In the calculations it was observed that L-alanine is the stable form among all other forms of alanines in solid form. The obtained vibrational frequencies in the calculations correspond to the experimental values, in addition, the energy of formation calculated by various methods is given [29]. For the concept of the structure and function of proteins and other biomolecules, the property of amino acid complexes in the gas phase and aqueous solution is considered [30]. A comparative analysis of calculation methods for describing the structure, energy and vibrational properties is carried out. The Raman spectrum and the Raman scattering intensity

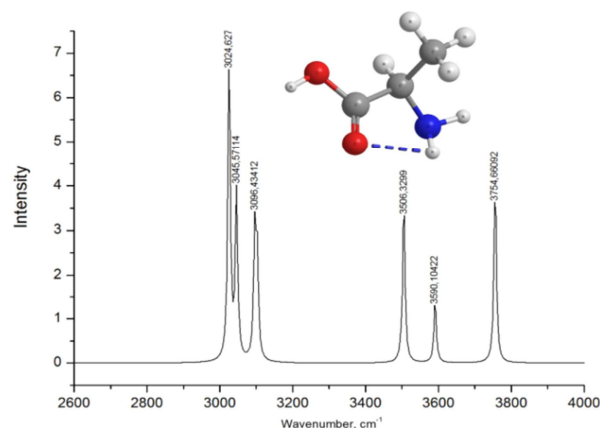
were modeled [30]. A complete understanding of the conformational equilibria of proteogenic amino acids is the first step towards understanding the three-dimensional structures of peptides and proteins. Therefore, using *ab initio* calculations, the electronic structure and energy dependence of the conformational equilibrium in the gas phase of alanine were studied. 12 structures with different torsion angles were analyzed. A correlation was found between the CO bond length of alanine conformers and their relative energies, and two groups of conformers (low and high energy) were identified. The importance of an accurate and systematic quantum-chemical description of the intramolecular hydrogen bond in gaseous (free) alanine is discussed. Benchmarks are provided for other (simpler) modern methods of quantum chemistry (e.g., dispersion-corrected density functional theory or DFT-D) [31].

In order to estimate the relative changes in the Raman spectra corresponding to "symmetric" and "antisymmetric" vibrations, to calculate the degrees of depolarization of the bands that appear as a result of the aggregation of alanine molecules into clusters, we performed *ab initio* calculations.

Based on the above published data, alanine was investigated using the RHF, B3LYP, MP methods using the basis of Gaussian functions 6-31G++ (d, p) for the monomer and other molecular isolated clusters of alanine molecules.

Alanine clusters were studied using wave function, geometric optimization and molecular dynamics to find the optimal value and the most suitable potential. In addition, the energies of intermolecular hydrogen bonds were calculated and to determine their geometric parameters, we carried out *ab initio* calculations of up to 5 molecules of alanine. After optimization of the geometry of the clusters, their geometric parameters were determined.

In the case of isolated molecules (monomer) of alanine in the Raman spectra, we observe (Figure 3) a band of OH vibrations (3754.7 cm<sup>-1</sup>), a band related to symmetric (3506.3 cm<sup>-1</sup>) and antisymmetric (3590.1 cm<sup>-1</sup>) vibrations of the NH<sub>2</sub> group. The calculation results show that in the monomers there exist within a molecular hydrogen bond, the length of which is 2.4 Å, formed by a group of COHN molecules.



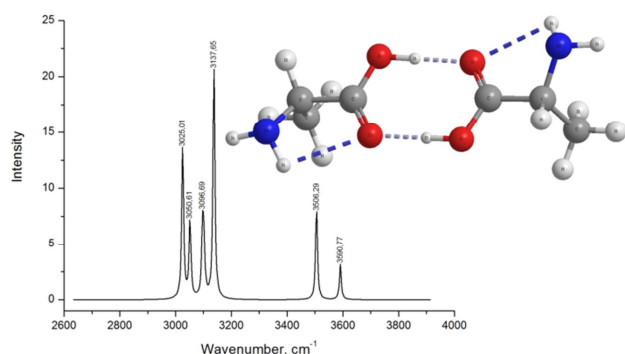
**Figure 3.** Raman spectra of OH and NH vibrations and the structure of an isolated alanine molecules.



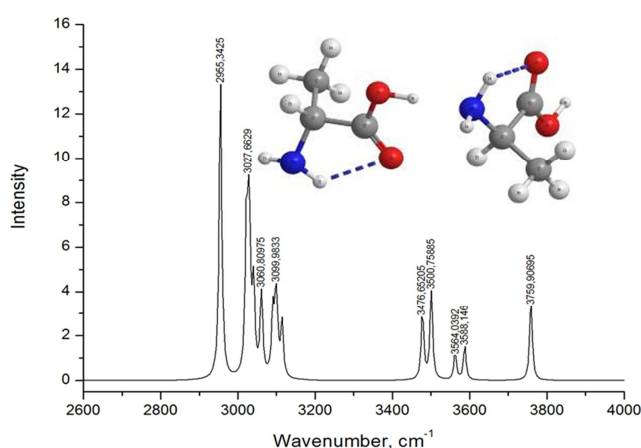
The optimized structure of isolated dimeric alanine aggregates is observed in two forms, which can be conventionally called closed and open forms. In the first form of the dimeric aggregate, two intermolecular (OHO) and two intramolecular (NHO) hydrogen bonds are observed (Figure 4). Calculations show that the length of the intermolecular hydrogen bond is 1.68 Å, for the intramolecular hydrogen bond this distance is 2.37 Å, respectively, the energy of formation is 35 kcal / mol.

In the Raman spectra of dimers of alanine molecules we observe a shift of the OH vibration bands, which is 617  $\text{cm}^{-1}$  due to the strong hydrogen bond. No significant changes are observed in the NH band. This can be explained by the existence within the molecular hydrogen bond forming with the help of the  $\text{NH}_2$  group.

Alanine molecules can form a dimeric aggregate through a non-classical (weak) hydrogen bond (Figure 5). In this case, the intermolecular bond is formed with the help of the COH group and the N molecules of the second molecule. The length of this bond is 1.74 Å; the energy of formation of this cluster is 12 kcal / mol. These two types of aggregates are the main material for the crystalline structure of alanine.



**Figure 4.** Raman spectra of OH and NH vibrations and the structure of an isolated dimeric (closed) aggregate of alanine molecules.



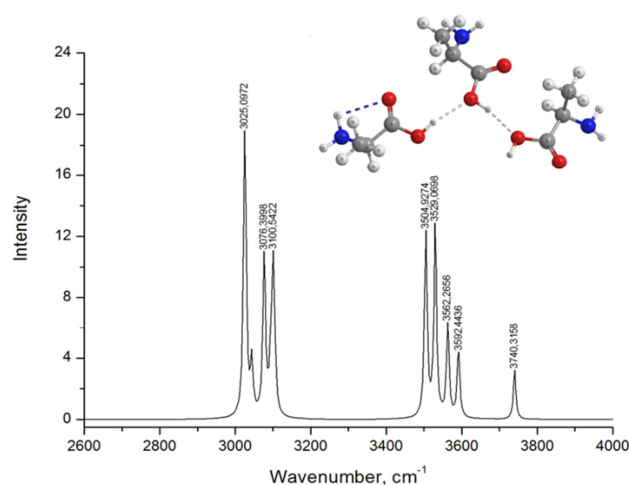
**Figure 5.** Raman spectra of OH and NH vibrations and the structure of an isolated dimeric (open) aggregate of alanine molecules.

The band participating in the intermolecular OH bond shifts towards low frequencies by 804.6  $\text{cm}^{-1}$ , which confirms the above assumption.

Three alanine molecules form a chain aggregate using the intermolecular hydrogen bond of the COH group (Figure 6). The length of the intermolecular hydrogen bond is 1.87 and 1.88 Å, respectively, the energy of formation of such an aggregate is 2.51 kcal / mol.

For this aggregate, the band of interacting OH vibrations is shifted towards low frequencies by 191.2  $\text{cm}^{-1}$  and 225.6  $\text{cm}^{-1}$  (3562.3 and 3529.1), respectively. For the band corresponding to NH vibrations, no significant changes are observed.

For tetrameric aggregation of alanine molecules, we observe 3 intermolecular and 3 intramolecular hydrogen bonds (Figure 7). The length of the intermolecular hydrogen bond in all 3 cases is 1.73 Å, the energy of each intermolecular hydrogen bond is 2.51 kcal / mol.



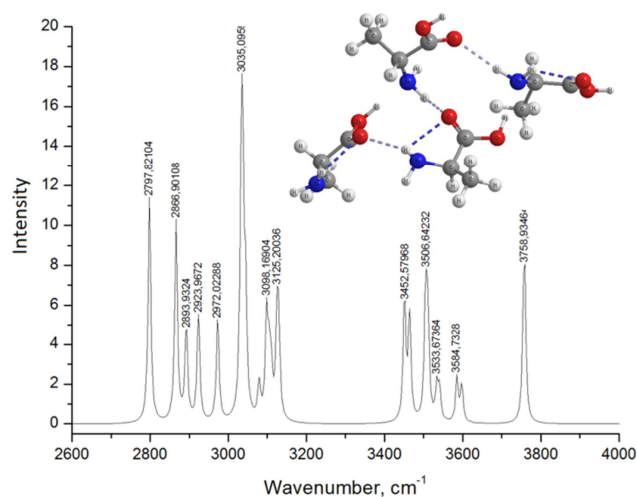
**Figure 6.** Raman spectra of OH and NH vibrations and the structure of an isolated trimeric (open) aggregate of alanine molecules.

In the case of a tetrameric molecular complex, the formation of chain aggregates is also observed. Analysis of the calculated Raman spectra of alanine up to four molecular clusters (Figure 7) shows that the Raman spectra are shifted towards low frequencies.

According to our assumptions, in the first case, due to the strong hydrogen bond, the O-H bond band is shifted by 956  $\text{cm}^{-1}$  (from 3754.7  $\text{cm}^{-1}$  to 2798.2  $\text{cm}^{-1}$ ) towards the low-frequency side. When the number of molecules in a cluster is dragged away, the band shifts toward lower frequencies than the previous cluster.

From our calculations, it can be seen that alanine molecules form aggregates of different types, while due to the O-H hydrogen bond, the band is shifted from ~3550  $\text{cm}^{-1}$  to ~2950  $\text{cm}^{-1}$ . In this case, the half-width and intensity increase.

In addition, the C=O vibration band also shifts to the low-frequency side by 40÷50  $\text{cm}^{-1}$ . With an increase in the number of molecules in a cluster, the energy of intermolecular interaction per hydrogen bond is saturated.



**Figure 7.** Raman spectra of OH and NH vibrations and the structure of an isolated tetrameric aggregate of alanine molecules.

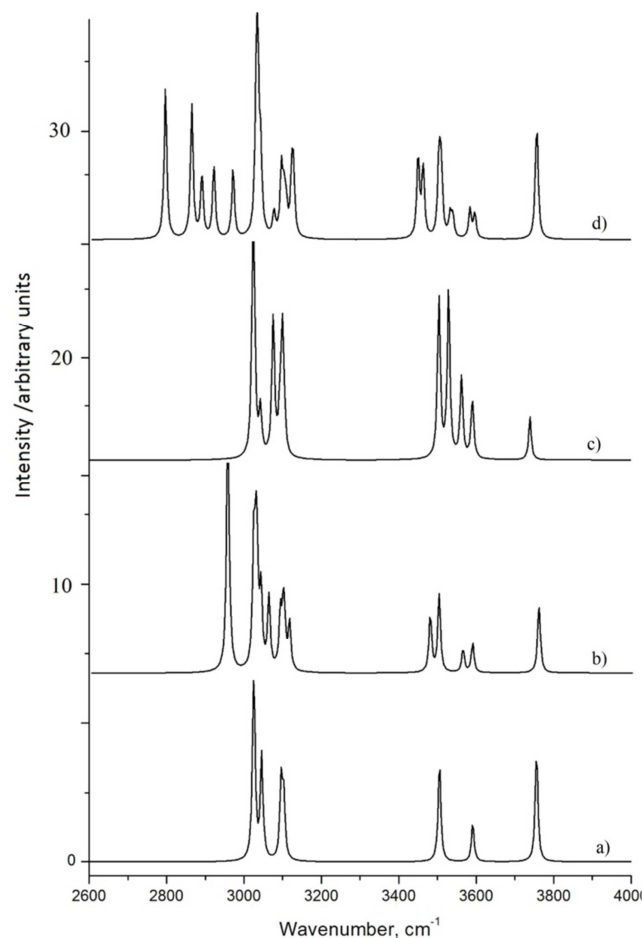
Raman spectra allow us to consider the change in the vibration frequency with an increase in interconnected molecules (Figure 8).

The bands of NH and OH vibrations lie in the high-frequency side, since the molecules turned out to be bound, then each vibration split into several vibrations, namely in the region of bending vibrations and in the region of stretching vibrations with depolarization coefficients, respectively.

Above, we showed that with an increase in the number of molecules in clusters, the bands shift to the low-frequency side, which means that there are stable molecular clusters in alanine.

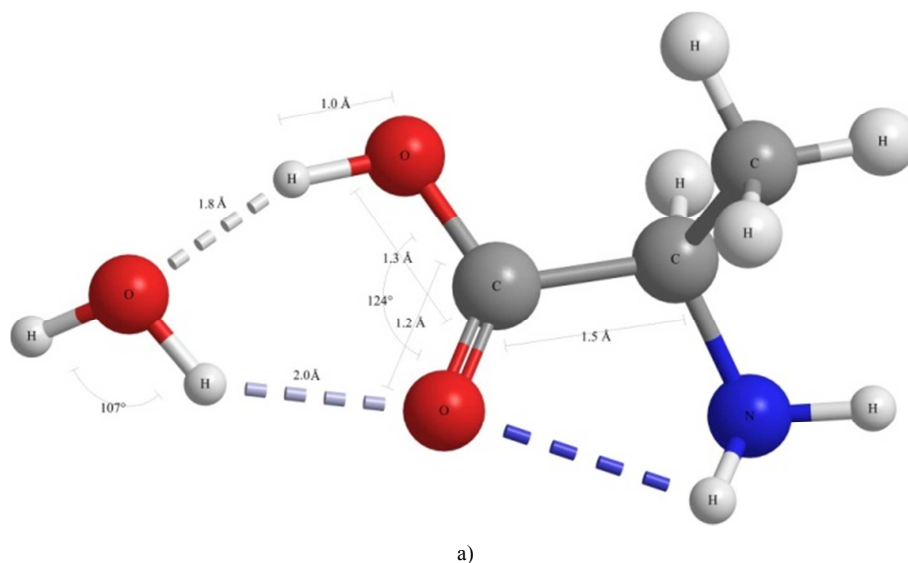
The hydrogen atom in the N-H group of the alanine molecule is actively involved in the formation of a hydrogen bond. The results of experimental studies and *ab initio* calculations show that dimeric, trimeric and other chain molecular complexes exist in alanine.

The complexes formed due to the hydrogen NH<sub>2</sub> bond are weaker than those formed due to the OH bond, which is in satisfactory agreement with the works [32, 33].

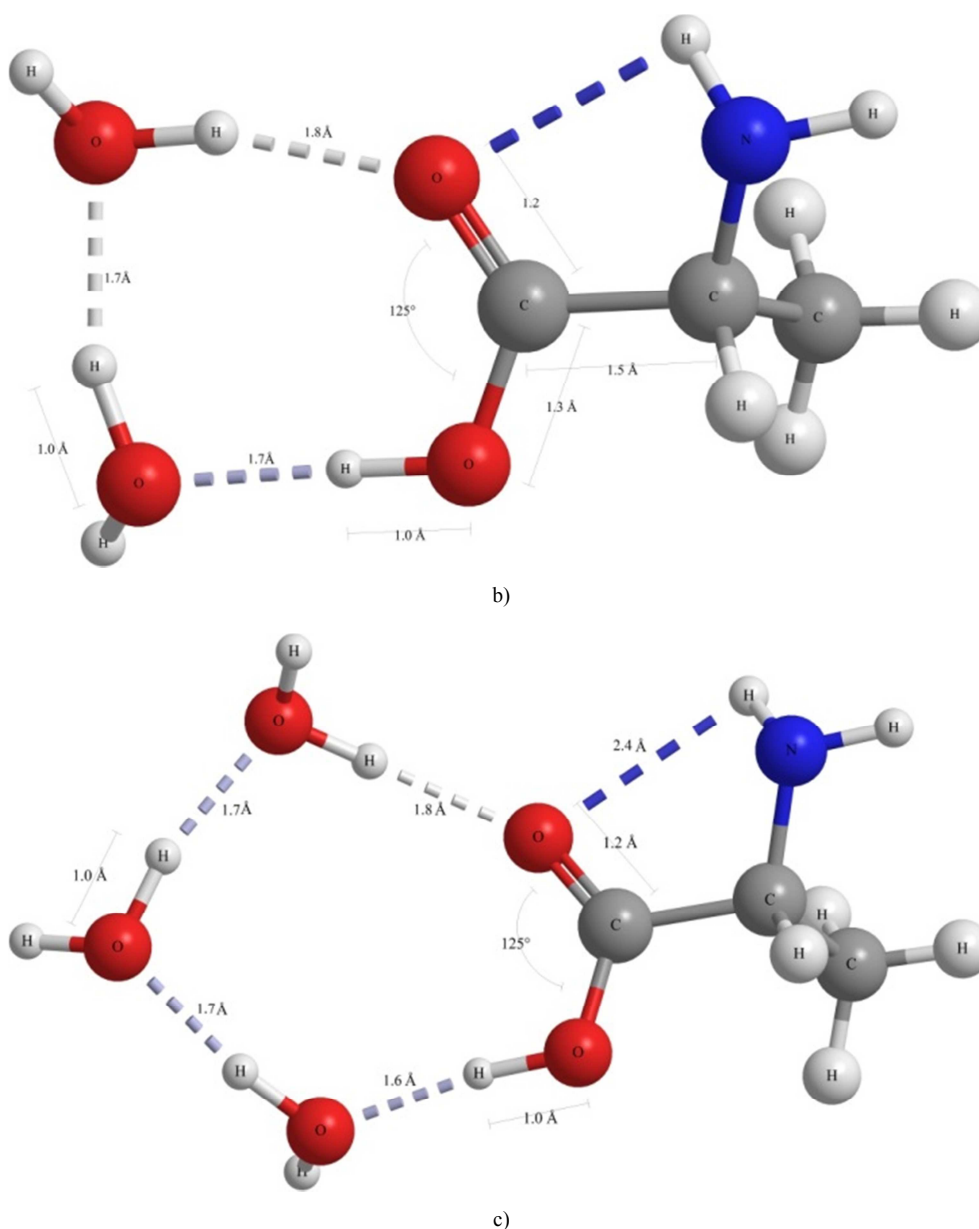


**Figure 8.** Calculated spectra of C-H, O-H and N-H vibrations of  $\alpha$ -alanine Raman scattering for various aggregated formations: a) monomer; b) dimer; c) trimer; d) tetramer.

Currently, more and more attention is attracted by aqueous solutions of amino acids, which are constituent components of proteins and play an important role in the life of organisms. For this purpose, we performed calculations of the complexes of  $\alpha$ -alanine with water (Figure 9).



a)



**Figure 9.** The structure of aggregates of  $\alpha$ -alanine with water a) alanine with one water molecule, b) alanine with two water molecules, c) alanine with three water molecules.

The calculation results show that the energy of formation of the first complex is 12.4 kcal / mol, and the second complex is 24.9 kcal / mol. In this work, the role of hydrogen bonds in the crystallization of alanine, which includes the carboxy COOH and amino NH<sub>2</sub> groups, and the manifestation of these intermolecular interactions in the Raman spectra are determined. The structures and binding energies of molecular aggregates of  $\alpha$ -alanine were studied using modern density functional methods, the processes of vibrational and translational movements of individual parts of amino acid molecules. Intramolecular hydrogen bond is formed in proteins between the NH and CO groups of adjacent helix turns, thereby ensuring the stability of the

secondary structure of the protein. The calculation results for alanine and its complexes are shown in Table 1.

In the crystal structure of alanine, a common structure-forming element can be distinguished - head-to-tail chains. These endless chains in alanine molecules form spiral layered crystal structures.

The spectral composition of alanine molecules is formed due to fluctuations of anisotropy, which change with the collective and individual movement of molecules.

The change in the Raman spectra is explained by the formation of several types of hydrogen bonds. These hydrogen bonds play a special role in the formation of the crystal structure of  $\alpha$ -alanine.

**Table 1.** Results of *ab initio* calculations for  $\alpha$ -alanine and its complexes.

№	Aggregate of $\alpha$ -alanine molecule	NH...O=C (Å)	CHO...OCH, *COH...NCH <sub>2</sub> and **COH...OH <sub>2</sub> (Å)	$\nu_{O-H}$ (cm <sup>-1</sup> )	$\rho$	D, Debay	$\Delta E$ ккал/моль	$\Delta E_{HB}$ ккал/моль
1.	Monomer	2,4	-	3755,9	0,23	2,23	-	-
2.	Dimer (close)	2,3	1,6	3137,5	0,28	2,92	14,6	7,2
3.	Dimer (open)	2,4	*1,7	2954,8	0,33	2,995	10,4	5,2
				3758,7	0,25			
4.	Trimer	2,3	1,9	3529,8	0,14	4,361	9,3	4,6
				3740,1	0,21			
				2298,3	0,33			
5.	Tetramer	2,3	*2,0	2798,2	0,33	4,15	27,5	6,9
				3756,5	0,26			
				3758,6	0,25			
				3396,6	0,23			
6.	Alanine+ water (one molecule in a water)	2,4	**1,8 2,0	3658,4	0,16	0,30	8,1	4,05
				3887,1	0,29			
				3151,9	0,21			
				3444,2	0,15			
7.	Alanine+water (two molecules in a water)	2,4	**1,7 1,7 1,8	3561,5	0,33	1,44	17,8	5,9
				3884,6	0,28			
				3887,6	0,21			
				3121,0	0,20			
				3363,6	0,12			
			1,6	3446,8	0,43			
8.	Alanine+water (three molecules in a water)	2,4	1,7 1,7 1,8	3582,6	0,34	0,49	25,6	6,4
				3882,5	0,74			
				3883,6	0,09			
				3888,8	0,24			

## 4. Conclusions

When a complex of alanine with water is formed, the O-H vibration band shifts to the low-frequency side by 320 cm<sup>-1</sup> and 415 cm<sup>-1</sup>, respectively. It is shown that with an increase in the strength of the hydrogen bond, the bond energy of the OH group increases.

It was found that in the Raman spectra of O-H and N-H the bands of  $\alpha$ -alanine in the region of 2900-3050 cm<sup>-1</sup> are complex and consist of several bands. The structure and connectivity of the bands is explained by the fact that the spectrum of the O-H bond consists of a symmetric valence band of vibrations  $\nu_1$  and overtones of bending vibrations  $2\nu_2$ .

It was shown that  $\alpha$ -alanine contains complex long-lived molecular complexes. The half-width and intensity of the O-H band increases and shifts from ~3550 cm<sup>-1</sup> to ~2950 cm<sup>-1</sup>, and the C=O band by 40÷50 cm<sup>-1</sup>. *Ab initio* calculations show that  $\alpha$ -alanine molecules form closed C=O-H...O=C and open N-H...O=C dimeric complexes.

The change in the Raman spectra is explained by the formation of several types of hydrogen bonds.

These results can be used to further study the intermolecular interactions and the structure of the formation of molecular clusters of various amino acids.

## References

- [1] Jeffrey G. A. Hydrogen bonding in biological structures / G. A. Jeffrey, W. Saenger. - Berlin, Heidelberg: Springer, 1994. - X1., 569 p. doi.org/10.1007/978-3-642-85135-3.
- [2] Desiraju G. R., Steiner T. The Weak Hydrogen Bond in Structural Chemistry and Biology.- New York: Oxford University Press, 1999.- 480 p.
- [3] Breen M. S., Kemena C., Vlasov P. K., Notredame C., Kondrashov F. A. Epistasis as the primary factor in molecular evolution // Nature. 2012. Vol. 490. Iss. 7421. P. 535-538. Doi: 10.1038/nature11510.
- [4] M. A. Belyanchikov, V. S. Gorelik, B. P. Gorshunov, A. YU. Pyatyshev. Infrared and Raman Spectroscopy of Glycine and Tyrosine Polycrystals // Herald of the Bauman Moscow State Technical University Series Natural Sciences. 2016 №4. S. 4-13. [In Russian]. DOI: 10.18698/1812-3368-2016-4-4-13.
- [5] Jenkins A. L., Larsen R. A., Williams T. B. Characterization of amino acids using Raman spectroscopy // Spectrochimica Acta Part A: Molecular and Biomolecular Spectroscopy. 2005. Vol. 61. Iss. 7. P. 1585-1594. https://doi.org/10.1016/j.saa.2004.11.055.
- [6] F. H. Tukhvatullin, V. E. Pogorelov, A. Jumabaev, H. A. Hushvaktov, A. A. Absanov, A. Shaymanov. Aggregation of molecules in liquid methyl alcohol and its solutions. Raman spectra and *ab initio* calculations. // Journal of Molecular Structure, Volume 881, Issues 1-3, 2008, P. 52-56. https://doi.org/10.1016/j.molstruc.2007.08.036.
- [7] F. H. Tukhvatullin, A. Jumabaev, H. Hushvaktov, A. Absanov, A. Shaymanov. Intermolecular h-bond in propan-2-ol and its solutions with acetonitrile // Journal of Raman Spectroscopy. 2007. Volume 38, Issue 12, Pages 1633 – 1638. https://doi.org/10.1002/jrs.1882.
- [8] F. H. Tukhvatullin, A. Jumabaev, V. E. Pogorelov, U. N. Tashkenbaev, H. Hushvaktov, et al. Intermolecular interaction in liquid dimethylformamide and its manifestation in Raman spectra // J. Raman spectroscopy. 2003. Issue 10 (34). P. 813-818. https://doi.org/10.1002/jrs.1057.



- [9] M. J. Frisch, G. W. Trucks, H. B. Schlegel, G. E. Scuseria, M. A. Robb, J. R. Cheeseman, G. Scalmani, V. Barone, B. Mennucci, G. A. Petersson, H. Nakatsuji, M. Caricato, X. Li, H. P. Hratchian, A. F. Izmaylov and *et al.*, Gaussian 03, Revision A.02 (Gaussian, Inc., 2003).
- [10] Zhu G., Zhu X., Fan Q., Wan X. Raman spectra of amino acids and their aqueous solutions // *Spectrochimica Acta Part A: Molecular and Biomolecular Spectroscopy*. 2011. Vol. 78. Iss. 3. P. 1187-1195. DOI: 10.1016/j.saa.2010.12.079.
- [11] C. H. Wang, R. D. Storms. Raman Study of Hydrogen Bonding and Long-Wavelength Lattice Modes in an L-Alanine Single Crystal//*J. Chem. Phys.* 55, 5110 (1971). <https://doi.org/10.1063/1.1675629>.
- [12] Katsunosuke Machida, Akira Kagayama, Yutaka Saito, Toyozo Uno. Polarized Raman spectra and intermolecular potential of L-alanine crystal// *Spectrochimica Acta Part A: Molecular Spectroscopy*. Volume 34, Issue 9, 1978, Pages 909-914. [https://doi.org/10.1016/0584-8539\(78\)80011-7](https://doi.org/10.1016/0584-8539(78)80011-7).
- [13] Santosh Kumar, Amareshwar Kumar Rai, S. B. Rai, D. K. Rai, A. N. Singh, V. B. Singh. Infrared, Raman and electronic spectra of alanine: A comparison with ab initio calculation// *Journal of Molecular Structure*, Volume 791, Issues 1-3, 19 June 2006, Pages 23-29. <https://doi.org/10.1016/j.molstruc.2006.01.004>.
- [14] F. H. Tikhvatullin, V. Ye. Pogorelov, A. Jumabaev, H. A. Hushvaktov, A. A. Absanov, A. Usarov. Polarized components of Raman spectra of O-H vibrations in liquid water // *Journal of Molecular liquids*, 160, (2011), P. 88-93. <https://doi.org/10.1016/j.molliq.2011.02.015>.
- [15] H. Hushvaktov, F. H. Tikhvatullin, U. N. Tashkenbaev, A. Jumabaev, A. Absanov, B. Hudoyberdiev. Raman spectra and ab initio calculation of a structure of aqueous solutions of methanol // *Journal of Molecular structure*, 1131 (2017) P. 25-29. <https://doi.org/10.1016/j.molstruc.2016.10.061>.
- [16] K. Kohlrausch. Raman spectra. Ed. M.: "Foreign Literature". 1952.-466 [In Russian].
- [17] Paulo T. C. Freire, Felipe M. Barboza, José A. Lima Jr, Francisco E. A. Melo and Josue Mendes Filho. Raman Spectroscopy of Amino Acid Crystals. Open access peer-reviewed chapter Raman Spectroscopy and Applications. February 15th 2017. DOI: 10.5772/65480.
- [18] S. Forss. A Raman spectroscopic temperature study of NH<sub>3</sub><sup>+</sup> torsional motion as related to hydrogen bonding in the L-alanine crystal // *Journal of Raman Spectroscopy*, 1982. V. 12 Issue 3. P. 266-273. <https://doi.org/10.1002/jrs.1250120313>.
- [19] Ivanov A. A., Korolik E. V., Insarova N. I., Zhibankov R. G., and Golubovich V. P., Low-temperature vibrational spectra and molecular structure of L-alanine, *Zh. app. range*. 1990. T. 53, No. 2. S. 265-270. [In Russian] DOI: <https://doi.org/10.1007/BF00659399>.
- [20] A. Culka, J. Jehlička, H. G. M. Edwards. Acquisition of Raman spectra of amino acids using portable instruments: outdoor measurements and comparison. *Spectrochimica acta. Part A, Molecular and Biomolecular Spectroscopy*, Volume 77, Issue 5, 2010, 77 (5): 978-983. DOI: 10.1016/j.saa.2010.08.034.
- [21] M. Thomas, M. Brehm, R. Fligg, P. Vohringer, B. Kirchner. Computing vibrational spectra from *ab initio* molecular dynamics. *J. Phys. Chem. Chem. Phys.* 15 (2013) 6608-6622.
- [22] J. R. Sambrano, A. R. de Sousa, J. J. Queralt, J. Andres, E. Longo. A theoretical analysis on the intramolecular proton transfer of  $\alpha$ -alanine in an aqueous medium. *Chemical Physics Letters* 294 1998, P. 1-8. [https://doi.org/10.1016/S0009-2614\(98\)00820-3](https://doi.org/10.1016/S0009-2614(98)00820-3).
- [23] Ilia A. Solov'yov, Alexander V. Yakubovitch, Andrey V. Solov'yov and Walter Greiner. Ab initio study of alanine polypeptide chains twisting//*Article in Physical Review E* 73 (2 Pt 1): 021916. March 2006. DOI: 10.1103/PhysRevE.73.021916.
- [24] Ten G. N., Gerasimenko A. YU., Shcherbakova N. Ye., Baranov V. I. Interpretatsiya IK i KR spektrov al'bumina // *Izvestiya Saratovskogo universiteta. Novaya seriya. Seriya Fizika*. 2019. T. 19, vyp. 1. S. 43-57. [In Russian] DOI: <https://doi.org/10.18500/1817-3020-2019-19-1-43-57>.
- [25] Nobrega G. F., Sambrano J. R., de Souza A. R., Queralt J. J., Longo E. DFT study of  $\alpha$ -alanine as a function of the medium polarity // *J. Mol. Struct.* 2001. Vol. 544. P. 151-157. DOI: [https://doi.org/10.1016/S0166-1280\(01\)00374-8](https://doi.org/10.1016/S0166-1280(01)00374-8).
- [26] Gab-Yong Lee. A DFT Study of the Intramolecular Hydrogen Bonding of Alanine and Its Effects on Ionization Energies. *Journal of the Korean Chemical Society* 2015, Vol. 59, No. 6 Printed in the Republic of Korea <http://dx.doi.org/10.5012/jkcs.2015.59.6.541>.
- [27] Lee, Gab-Yong. Structure and Intramolecular Proton Transfer of Alanine Radical Cations. *Bulletin of the Korean Chemical Society*. Volume 33 Issue 5/ Pages. 1561-1565. /2012/0253-2964 (pISSN)/1229-5949 (eISSN). <https://doi.org/10.5012/bkcs.2012.33.5.1561>.
- [28] Attila G. Császár. On the structures of free glycine and  $\alpha$ -alanine *Journal of Molecular Structure*. Volume 346, 15 February 1995, Pages 141-152. [https://doi.org/10.1016/0022-2860\(94\)09017-J](https://doi.org/10.1016/0022-2860(94)09017-J).
- [29] R. Kumar, A. K. Sharma, S. D. S. Chauhan, D. Kulshreshtha, R. Gupta, P. K. S. Chauhan and O. P. Singh Ab Initio Study of Vibrational Spectra of Alanine in Gas Phase//*Material Science Research India*. Vol. 6 (2), 551-558 (2009).
- [30] K. J. Jalkanena, M. Elstnerb, S. Suhaic. Amino acids and small peptides as building blocks for proteins: comparative theoretical and spectroscopic studies. *Journal of Molecular Structure (Theochem)* 675 (2004) 61-77. DOI: 10.1016/j.theochem.2003.12.045.
- [31] Roman M. Balabin. Conformational equilibrium in alanine: Focal-point analysis and ab initio limit//*Computational and Theoretical Chemistry* 965 (2011) 15-21. DOI: 10.1016/j.comptc.2011.01.008.
- [32] Moovendaran K., Martin Britto Dhas S. A., Natarajan S. Spectral characterization of a non-centrosymmetric organic compound: D-alanine // *Spectrochimica Acta Part A: Molecular and Biomolecular Spectroscopy*. 2013. Vol. 112. P. 326-330. DOI: 10.1016/j.saa.2013.04.025.
- [33] Daniel A., Prakassaro A., Dornadula K., Ganesan S. Polarized Raman spectroscopy unravels the biomolecular structural changes in cervical cancer // *Spectrochimica Acta Part A: Molecular and Biomolecular Spectroscopy*. 2016. Vol. 152. P. 58-63. DOI: 10.1016/j.saa.2015.06.053.