Research Inventy: International Journal of Engineering And Science
Vol.7, Issue 1 (January 2017), PP -22-27
Issn (e): 2278-4721, Issn (p):2319-6483, www.researchinventy.com

Crystallization of L-Glutamic Acid: Mechanism of Heterogeneous β -Form Nucleation

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Abstract: *The mechanism of heterogeneous nucleation of β-form L-glutamic acid was deeply investigated in cooling crystallization. The present study found that the β-form crystals were epitaxially grown on the α-form crystals and they were preferably crystallized on the (011) and (001) surfaces instead of the (111) surfaces of α-form crystals. This result was explained via the molecular simulation. The molecular simulation indicated that the different surfaces of α-form crystals provided different functional groups, resulting in different sites for the heterogeneous nucleation of β-form crystals. Here, the functional group were COO^-, C=O and O-H on the (011) and (001) surfaces of α-form crystals, respectively, while it was the NH_3^+ on the (111) surfaces of α-form crystals. As such, the degree of lattice matching (E) between the β-form crystals and the various surfaces of α-form crystal was distinguished, where the degree of lattice matching (E) between the β-form crystals and the (011), (001) and (111) surfaces of α-form crystal were estimated as 5.30, 5.25 and 2.39, respectively, implying that the (011) and (001) surfaces of α-form crystal were more favorable to generate the heterogeneous nucleation of β-form crystals than the (111) surfaces of α-form crystal.*

Keywords: *crystallization, crystallography, crystal growth, nucleation, polymorphism.*

I. Introduction

Crystallization is significant separation, purification and particle technology, which is widely used in the life-science industries including pharmaceutical, food and agrochemicals [1-2]. In crystallization, polymorphism is a very important and interested phenomenon, where the solid product can adopt more than one crystal structure due to the different packing arrangement and conformation of molecules in the crystal lattice. The different polymorphic solid forms exhibit a marked difference in the physical-chemical properties including bio-availability, solubility, hardness, stability, etc. In the pharmaceutical industry, there are more than 90% products commercialized in the solid forms, and at least 50% of these products has the polymorphism, meaning that control of polymorphism becomes a vital issue in any pharmaceutical crystallization process [3-6].

Once the polymorphism can affect the quality, safety and efficacy of the pharmaceutical solid products, the Food and Drug Administration (FDA) in US requires all the pharmaceutical company to submit the approval of a new drug application as if this drug has the polymorphic forms. Also, the FDA has provided a detail of the tree recommendation in order to monitor and control polymorphic pharmaceutical products. In present study, the amino acid L-glutamic acid was chosen as a model crystal product to demonstrate the control of polymorphism in crystallization. Here, the amino acid L-glutamic acid has widely applied in all the food and pharmaceutical industry, where it has two kinds of polymorphism including metastable α-form and stable β-form crystal. In the L-glutamic acid crystallization, the polymorphic nucleation phenomenon is very complicated and elusive because it depends on many crystallization conditions. For example, Lai et al [7] indicated that the α-form nucleation was facilitated at low temperature of 25^0C, while the β-form nucleation was promoted at high temperature of 45^0C in the continuous MSMPR crystallizer. In order to increase the recovery of β-form crystal product at low temperature of 25^0C, Florence et al [8] reported that the seeding technique was really effective as using the continuous Oscillatory baffles crystallizer (OBC). Recently, Tahri et al [9] revealed that the nucleation of α-form and β-form crystal were both generated in the stirred condition, while the only β-form nucleation was performed under the stagnant condition, meaning that the agitation condition was also important since it impacted on the polymorphic nucleation of L-glutamic acid, etc.

In contrast to previous study, our current work investigated the mechanism of heterogeneous nucleation of β-form L-glutamic acid in the stirred cooling crystallization, which was not fully described in the previous studies. As such, the mechanism of polymorphic nucleation L-glutamic acid in stirred cooling crystallization would be more understood. In addition, although the crystallization research is very common in the developed countries such as USA, EU, Japan, Korea, China, India, Singapore, etc, it is still limited in the South and East Asian countries including Vietnam, Indonesia, Philippine, Malaysia, etc [10-11], meaning that the crystallization research should be developed in these countries.

II. Experimental

L-glutamic acid material (>98% purity) was bought from Sigma Aldrich company and the feed solution was prepared by dissolving material into the distilled water at 70^0C. The standard stirred tank crystallizer with 400 ml working volume was designed by Nguyen et al [4-6]. Initially, the stirred tank crystallizer was fully filled with feed solution at concentration of 40 (g/L) and then operated as the batch mode cooling crystallization at cooling rate of 6.0 (^{0}C/min) and agitation speed of 300rpm. The temperature of crystallizer was controlled via the circulating coolant from the chiller. The suspension products were periodically taken from the crystallizers and quickly filtered by using a vacuum pump. The crystal samples were then dried in a desiccator and analyzed to define properties of products including the shape, structure and crystal fraction of α-form and β-form. Here, the shape and structure of crystal product were monitored and confirmed by Video microscope and XRD patterns (M18XHF-SRA, Japan), respectively, while the crystal fraction was estimated by the FTIR spectroscopy (Perkin Elmer, System 2000 FT-IR, U.S.A.). The molecular simulation was carried out via the crystallographic software including Encifer, Mercury, Diamon and GRACE.

III. Results And Discussions

1. Polymorphism of α-form and β-form crystal product: The morphology of α-form and β-form crystals was clearly different via the prism shape of α-form and needle shape of β-form crystal, respectively, as shown in Figure 1. Moreover, the crystal structure of two polymorphs was distinguished via the characteristic peaks at 10^0, 15^0, 16^0, 18^0, 21^0, 23^0, 26.5^0, 27.5^0 degrees of XRD pattern, as depicted in Figure 2.

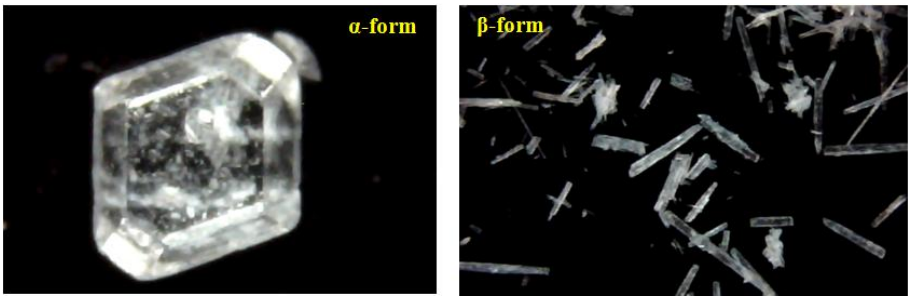

Figure 1. Morphology of α-form and β-form crystal product

Therefore, L-glutamic acid has two kinds of polymorphism including α-form and β-form crystal. For a deeper insight into differences of crystal structure, the crystallography of two polymorphs was studied. Here, the parameters and position of each atom of unit cell were obtained from the previous studies [12-13], and then coded via the Encifer software. The conformation and packing of L-glutamic acid molecules in unit cell were simulated via the Mercury and Diamon software. As shown in Figure 3, the conformation of molecule L-glutamic acid in α-form crystal was different from that of β-form crystal. For instance, the O(1)-C(1)-C(2)-N torsional angles were -50.41 in the α-form, while they were -42.26 in the β-form. Furthermore, although the crystal system and number of molecules in a unit cell of two polymorphs are orthorhombic with space group $P2_12_12_1$ and Z=4, respectively, the unit cell parameters and packing of molecules of two polymorphs were definitely distinguished. Here, the unit cell parameters of α-form crystal were a = 7.068, b = 10.277, c = 8.755 Å, while those of β-form crystal were a = 5.159, b = 17.300, c = 6.948 Å, as depicted in Figure 4. Plus, the different packing of molecules of two polymorphs was clearly recognized at the "c" and "a" direction, as shown in Figure 4.

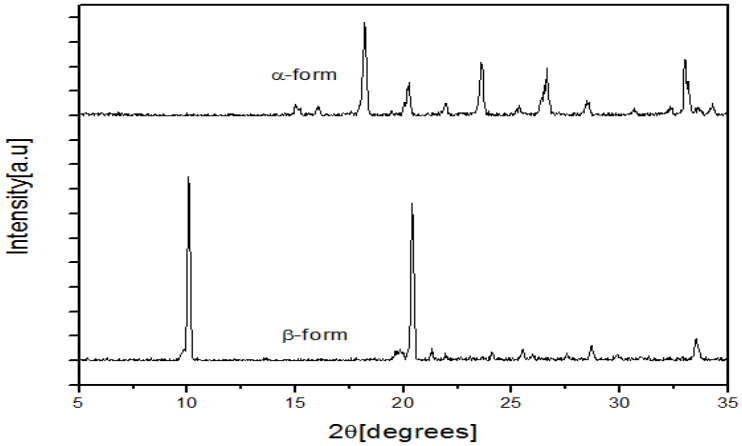

Figure 2. Crystal structure of α-form and β-form crystal product

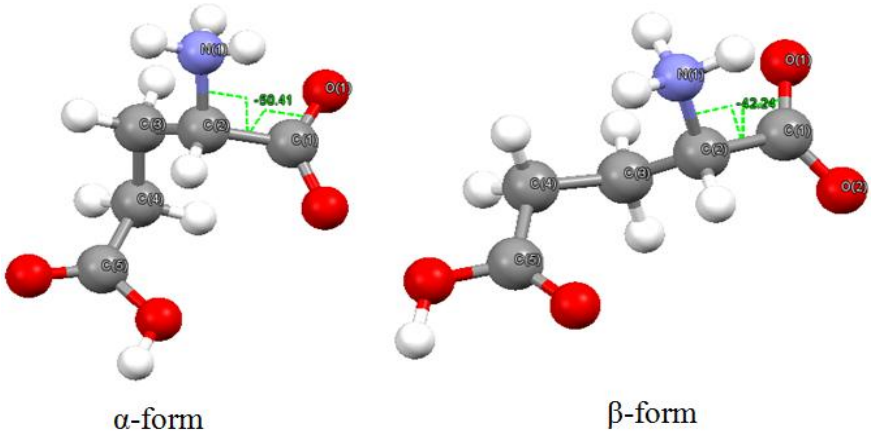

Figure 3. Conformation of molecules in α-form and β-form crystal product

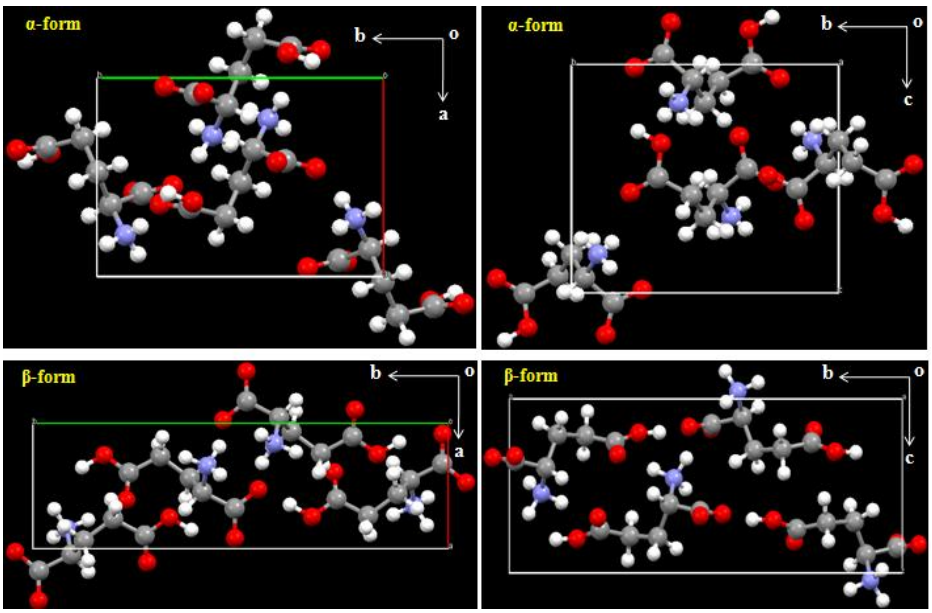

Figure 4. Packing of molecules in "c" and "a" direction in unit cell of α-form and β-form crystal product

2. Heterogeneous nucleation of β-form crystal product: In L-glutamic acid cooling crystallization, the metastable phase α-form crystal was initially crystallized and then completely transformed into the stable phase β-form crystal after a crystallization time of 40 hours [14]. As such, it took a long time to complete the phase transformation from α-form to β-form crystal. Based on our previous study [14], the phase transformation of α-form to β-form crystal should be significantly promoted if the nucleation of β-form crystal was accelerated. In addition, it is well known that the heterogeneous nucleation is dominant in the stirred industrial crystallization due to the sweep of fluid hydrodynamic on the surface of mother crystal and collision between crystal and crystal, crystal and crystallizer's wall, crystal and impeller, etc. Thus, the mechanism of heterogeneous nucleation of β-form was certainly important and should be deeply investigated in the stirred cooling crystallization of L-glutamic acid. As shown in Figure 5, the heterogeneous nucleation of β-form crystal was obviously occurred and epitaxially grown on the surfaces of α-form crystal, meaning that the surfaces of α-form crystal were considered as the substrate to generate the heterogeneous nucleation of β-form crystal. This result was expected in terms of a low free energy barrier of the substrate in order to generate the heterogeneous nucleation [1-2]. However, the favorable site for the heterogeneous nucleation of β-form on a specific surface of α-form crystal was not fully understood in the stirred cooling crystallization of L-glutamic acid. Thus, in present study, the formation of heterogeneous nucleation of β-form on a specific surface of α-form crystal was investigated. Since the epitaxial growth of β-form molecules on a specific surface of α-form crystal directly depended on the functional group of molecules oriented on the surfaces of α-form, the distinguished functional group of molecules on each α-form surface should be clearly clarified. As shown in Figure 6, the various surfaces of α-form crystal including (011), (001) and (111) and the heterogeneous nucleation of β-form possibly occurred on these surfaces were estimated via the (hkl) Miller planes method [1-2].

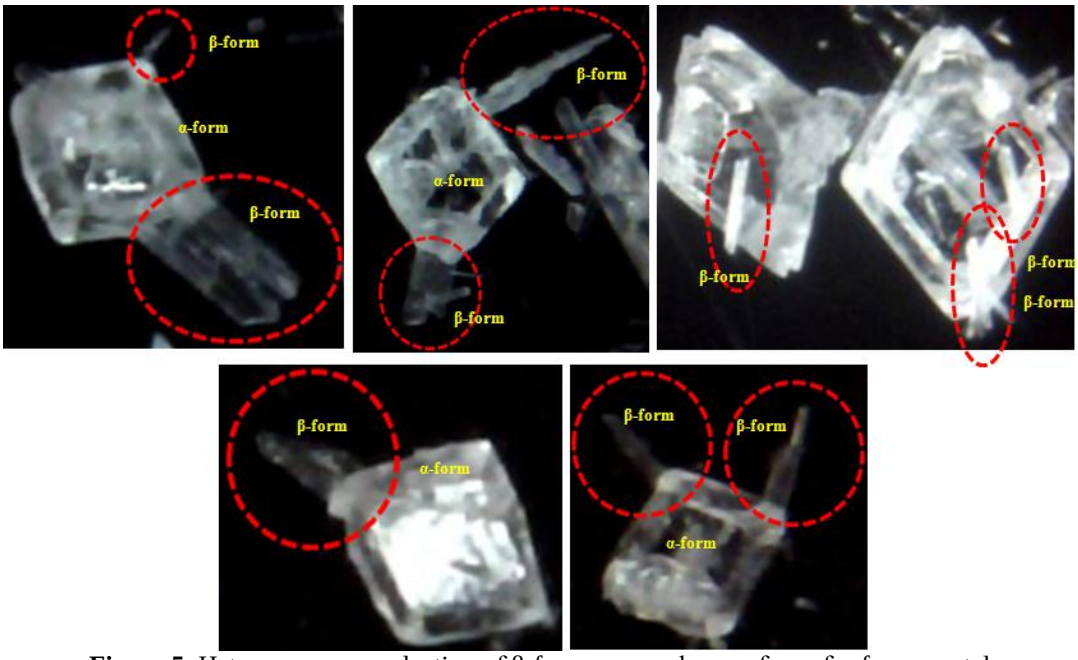

Figure 5. Heterogeneous nucleation of β-form occurred on surface of α-form crystal

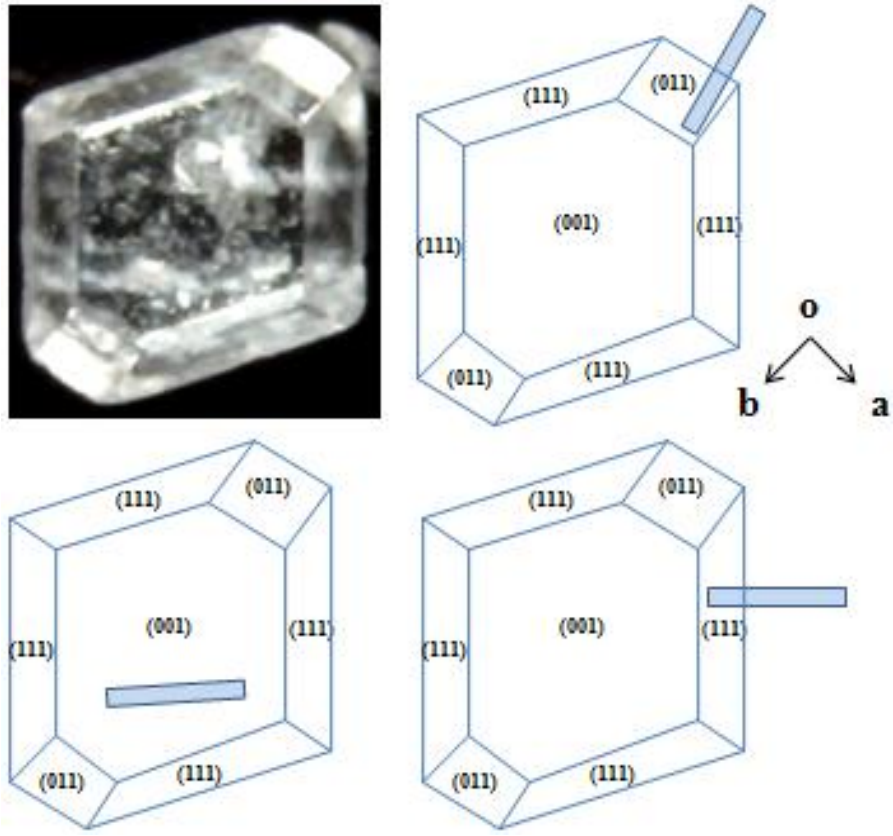

Figure 6. Schematic heterogeneous nucleation of β-form possibly occurred on the different surfaces of α-form crystal

The functional group of L-glutamic acid molecules oriented on the surface of α-form crystal was estimated via the Diamon software, as displayed in Figure 7. This result showed that there was a significant difference of functional group on three surfaces, in which the functional group including C=O, O-H and COO^- appeared on the (001) and (011) surfaces, respectively, while it was NH_3^+ on the (111) surface. As such, the different functional groups on the surfaces of α-form crystal might provide distinguished sites for the heterogeneous nucleation of β-form crystal. This hypothesis was concreted by the different degree of lattice

matching (E) between the β-form molecules aggregate and a specific surface of α-form crystal, where the degree of lattice matching (E) was estimated via the GRACE software with varied θ azimuth angles from -90^0 to 90^0 degree and a search area of $100A^0$ x $100A^0$ [15]. As shown in Figure 8, the result showed that the prominent peaks of (001) and (011) surfaces were both appeared at $\theta = 90^0$, while the characteristic peak of (111) surfaces was observed at $\theta = 15^0$. Plus, the degrees of lattice matching E of (011), (001) and (111) surfaces were calculated as 5.30, 5.25 and 2.39, respectively, meaning that the (011) and (001) surface were more favorable site for the heterogeneous nucleation of β-form crystal compared to the (111) surfaces of α-form crystal. This result was consistent with the experimental result as illustrated in Figure 5, where the needle shape of β-form crystal was predominant on the (011) and (001) surface instead of the (111) surfaces of α-form crystal.

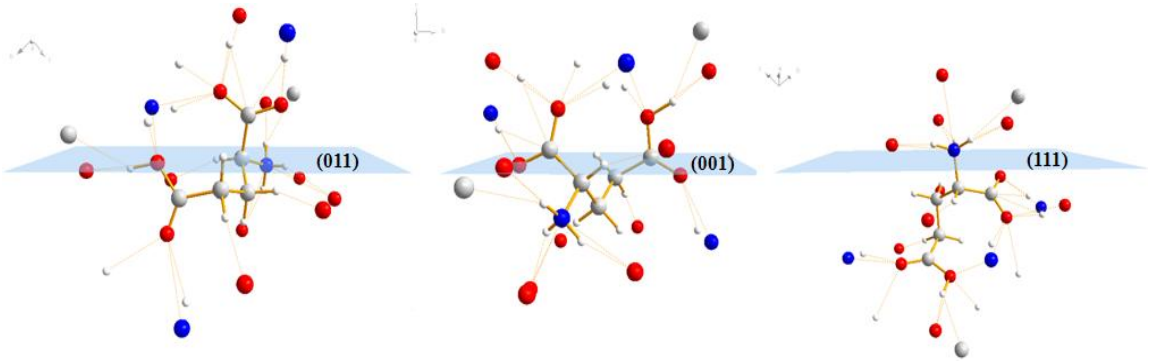

Figure 7. Different functional group oriented on the various surfaces of α-form crystal

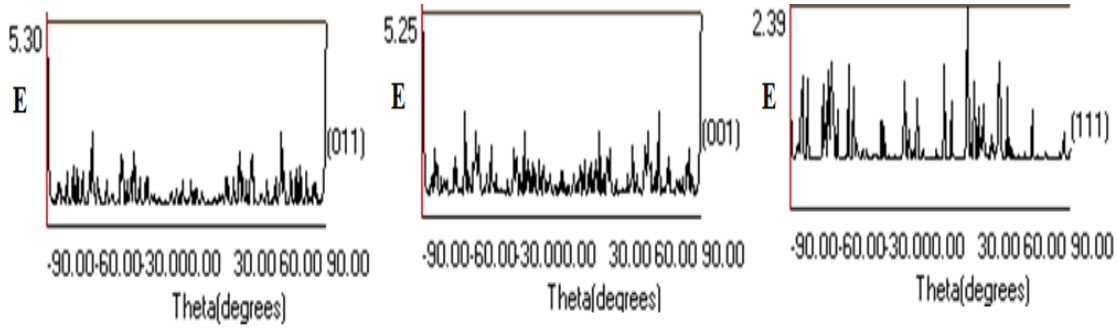

Figure 8. Lattice matching (E) between β-form molecules and the surface of α-form crystal

IV. Conclusions

The present study provided a proposed mechanism of heterogeneous nucleation of β-form L-glutamic acid in the stirred cooling crystallization. Here, the heterogeneous nucleation of β-form was preferably performed on the (011) and (001) surfaces of α-form crystal instead of the (111) surfaces at feed concentration of 40 (g/L) and agitation speed of 300 rpm. This result was successfully explained via the molecular simulation, where the molecular simulation result showed that the functional group of molecules on the (011) and (001) surfaces were COO^-, C=O and O-H, respectively, while it was the NH_3^+ on the (111) surface of α-form crystals. Thus, the degree of lattice matching (E) between the β-form crystal and surfaces of α-form crystal was certainly distinguished. Here, the degrees of lattice matching (E) between the β-form crystal and the (011), (001) and (111) surfaces of α-form crystal were estimated as 5.30, 5.25 and 2.39, respectively, meaning that the (011) and (001) surfaces of α-form crystal were the more favorable place for generating the heterogeneous nucleation of β-form crystal in the stirred cooling crystallization of L-glutamic acid.

V. References

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