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Self-assembly of novel cholesterol derivative based on hydrogen bond

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ABSTRACT

In this paper, a novel cholesterol derivative was found that it can form regular arrangement with millimeter size when we investigated the synthesis of new cholesterol monomer. The quite interesting thing is that the arrangement can form on common glass sheet after the sample is heating above its melting point and then cooling to room temperature. This is different from most reported regular arrangements formed in solutions. According to the single crystal results of the sample, we believe that the arrangement formed in solid state can be attributed to hydrogen bonding formed between molecular and van der Waals force among molecular.

1. Introduction

Self-assembly behavior is one common phenomenon in nature. The small or macro-molecules could aggregate into new or special structures with various functions under a certain conditions [1]. In recent years, study on self-assembly has been attracted broad interests because of the possibility of forming millimeter- [2] or nanometer-scale [3] materials with certain well-organized structure [3], morphology [4] and functions [5]. Self-assembly behavior is mainly driven by non-covalent interactions, such as hydrogen bond, van der Waals force, π–π interaction [6]. In addition, the hydrogen bond, one kind of moderate intensity and directionality force, has more favorable molecular orientation than charge interactions and van der Waals force.

Usually these kinds of molecules suitable for assembling well-ordered arrangements and structures have functional groups such as hydroxyl, carboxyl, and amine among these groups non-covalent interactions can be formed in solution or in solid state. In most cases, the preparation methods are firstly dissolving the sample in suitable solvent and then casting them on ordered substrates, especially on metals (e.g. Si, Cu, Ag and Au) [7–10] or graphite [11–13]. Obviously, the ordered surface is helpful for the assembly process, seldom reports [14] have been mentioned forming ordered arrays on the surface of sheet glass.

Cholesterol can be obtained from organism, and it belongs to the family of steroid compounds. There are two functional groups in cholesterol structure, hydroxyl on C3 position (C3—OH) and double bond between C5 and C6 (C5=C6). However, many research work have been done based on C3—OH, seldom work [15,16] based on C5=C6. There are many researchers who are interested in the cholesterol derivatives in the world, such as Mallia, Ikeda, Kasi, Zhu groups [17–24]. But in Zhu' group, they have done majority work relating to cholic acid and investigated the self-assembly of cholic acid derivatives because the derivatives always still have amphiphilic properties.

In our group, we have ever synthesized new type of cholesterol derivatives on C6 position [15,16], the hydroxyl groups on C3 position was etherification protected. As a continuation of the research work, this time we still designed to synthesize derivatives on C6 position but the hydroxyl group was esterification protected. We found that one of the derivatives has an interesting well-organized structure on common glass substrates when we investigated cholesterol derivatives on POM. And another interesting thing is the self-organized process is different from the common methods reported, putting a little amount solid sample between two glass substrates and then heating the solid sample above its melting temperature and then cooling the sample to room temperature, well-organized structures appeared.

2. Experimental section

2.1. Materials

Cholesterol, acetic anhydride, dicyclohexylcarbodiimide (DCC), pyridine, 3-chloroperoxybenzoic acid (m-CPBA), dichloromethane, α-methacrylic acid, hydroquinone were all purchased from GuangHua Chemical Co. of China and used as received. Tetrahydrofuran (THF, analytical reagent; GuangHua chemical reagents) was purified through standard methods.

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2.2. Instruments

FTIR spectra were measured on a FT-IR (Nicolet) spectrophotometer. Proton nuclear magnetic resonance (^{1}H NMR) spectra were recorded on a BrukerARMx-400 operating at 400 MHz in deuterated chloroform. Differential scanning calorimetry (DSC) was carried out on a thermal analysis (TA) DSC-Q30 with a liquid nitrogen cooling system. The samples were heated from room temperature to 175 °C, at a heating rate of 10 °C/min, followed by cooling at the same rate. The crystal structure was obtained on CAD4SDP-44M/H four-circle single crystal diffractometer made by Enraf–Nonius Company of Holland. A polarizing optical microscope (POM) equipped with a hot stage was used to observe phase transition temperatures and optical textures for the monomer. Elemental analysis was carried out using an Italian CARLO ERBA 1106 Elemental analyzer, which simultaneously detects C, H, N, percentage.

2.3. Synthesis process

The final cholesteryl derivatives were synthesized starting from cholesterol. The hydroxyl group of cholesterol was initially protected by esterifying with acetic anhydride, and then the double bond in cholesterol skeleton was epoxided. Finally the epoxy group was opened using methacrylic acid. The synthesis route is shown in Scheme 1.

2.3.1. Synthesis of cholesteryl acetate (*1*)

In a typical experiment, cholesterol (30 g, 0.08 mol), acetic anhydride (15 ml), DCC (8 g, 0.04 mol), and a few drops of pyridine were added to fleshly distilled THF (100 ml) and the reaction mixture was refluxed for 72 h under N_2 atmosphere. Then the white precipitate was removed by filtration and the filtrate was evaporated, the crude product was recrystallized twice from ethanol, obtaining product **1** with yield of 90.2%.

^{1}H NMR (400 MHz, $CDCl_3$): δ (ppm) = 5.37 (s, 1H, 6-H), 4.62 (s, 1H, 3-H), 2.01 (m, 3H, —CH_3);

IR (KBr pellet, cm^{-1}): 2942.9–2867.2 (—CH_2—), 1735.7 (—C=O), 1667.6 (—C=C), 1244.7, 1032.0 (C—O—C).

Elem. Anal. Calcd. for $C_{29}H_{48}O_2$: C, 81.25%; H, 11.29%. Found: C, 81.12%; H, 11.20%.

2.3.2. Synthesis of 5,6-oxy-cholesteryl acetate (*2*)

Compound **2** was prepared according to the literature procedure [25]. Cholesteryl acetate (8.57 g, 0.02 mol), m-CPBA (4.14 g, 0.024 mol), CH_2Cl_2 (60 ml) were added to a 150 ml round-bottom flask with flap loosely capped, and then stirred for 24 h at room temperature. After removing the white precipitate, the filtrate was washed by saturated sodium bicarbonate and sodium chloride solution several times, sequentially, and purified from methanol at last. The yield was 79%.

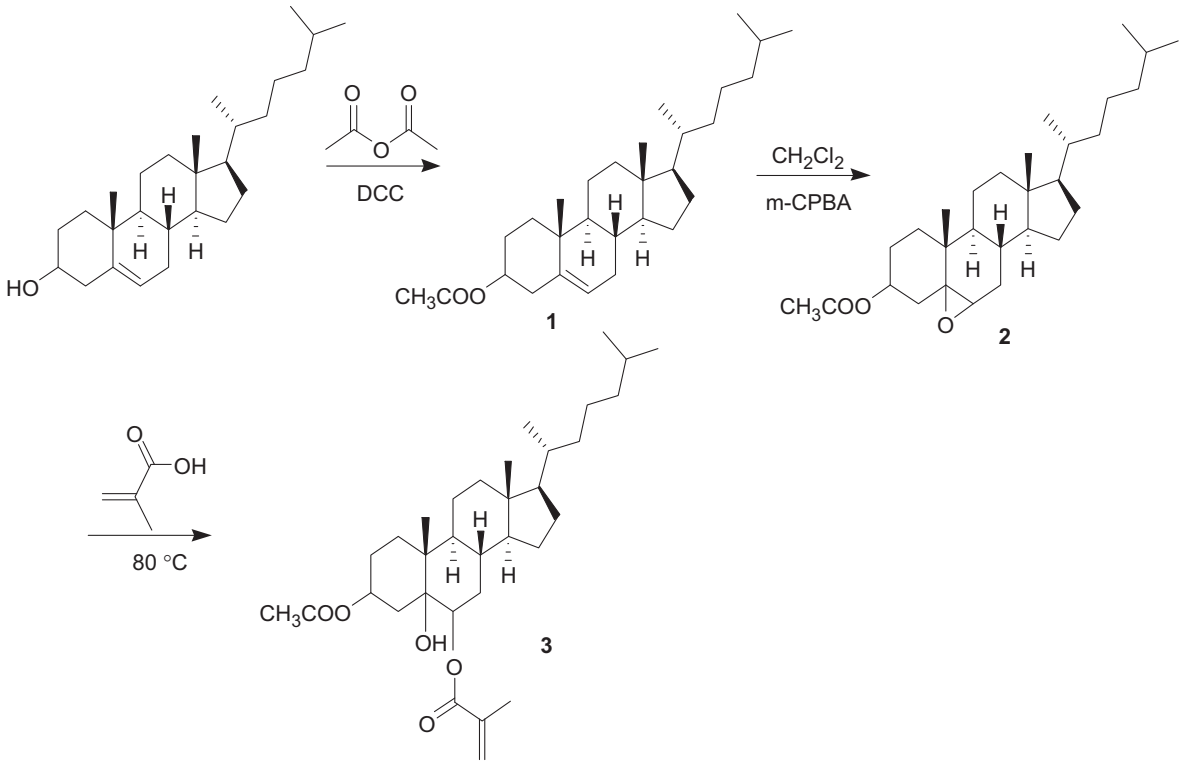
^{1}H NMR (400 MHz, $CDCl_3$): δ (ppm) = 5.02 (s, 1H, 6-H), 4.85 (s, 1H, 3-H), 2.2 (m, 3H, —CH_3);

IR (KBr pellet, cm^{-1}): 2952.4–2868.8 (—CH_2—), 1732.7 (—C=O), 1240.9, 1039.4 (C—O—C).

Elem. Anal. Calcd. for $C_{29}H_{48}O_3$: C, 78.33%; H, 10.88%. Found: C, 78.35%; H, 10.97%.

2.3.3. Synthesis of 5-hydroxyl, 6-methacrylate-cholesteryl acetate (*3*)

Compound **2** (3 g, 6.7 mmol) and 1 mg hydroquinone was dissolved in 15 ml α-methacrylic acid without any catalysts. The reaction was carried out at 80 °C for 48 h under nitrogen atmosphere. The mixture was dissolved in acetic ether followed by washing using saturated sodium bicarbonate and sodium chloride solution several times, sequentially. The organic layer was dried by magnesium sulfate anhydrous for 12 h, obtaining the crude product after removing the solvent under reduced pressure. And then the product was purified by silica gel column chromatography with the mixture of mineral ether and acetic ether (8:1). Yield: 58%.



Scheme 1. The synthesis route of novel cholesterol derivate monomer.

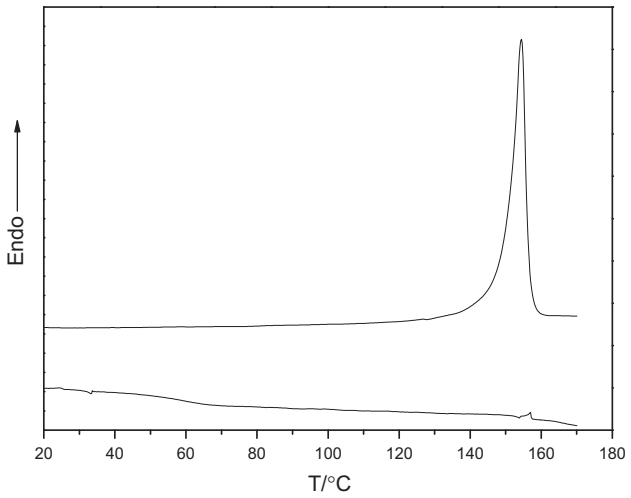


Fig. 1. The DSC heating and cooling curves of compound **3.**

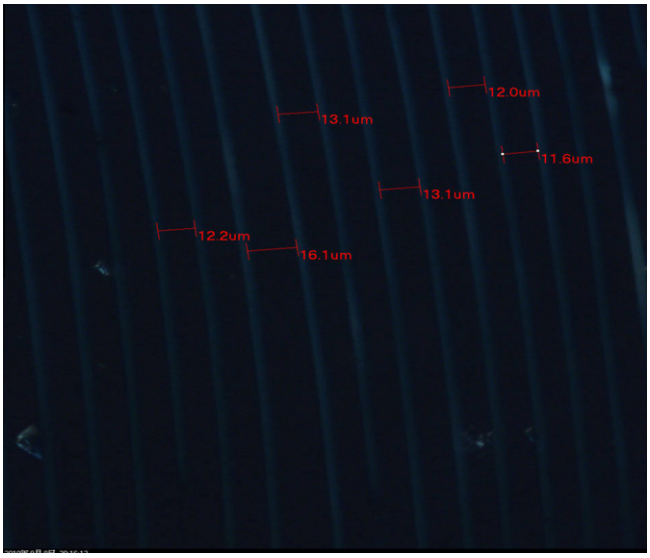


Fig. 2. The POM image after the compound **3** cooling from melting state to room temperature.

^{1}H NMR (400 MHz, $CDCl_3$): δ (ppm) = 5.62, 6.13 (s, 2H,=CH_2), 5.31 (s, 1H, 6-H), 4.78 (s, 1H, 3-H), 2.2 (m, 3H, —CH_3);

IR (KBr pellet, cm^{-1}): 3474.9 (—OH), 3101.7 (—C=CH_2), 2941.8–2869.5 (—CH_2—), 1737.0 (—C=O), 1636.1 (—C=C), 1241.7, 1025.2 (C—O—C).

Elem. Anal. Calcd. for $C_{33}H_{54}O_5$: C, 74.67%; H, 10.25%. Found: C, 74.55%; H, 10.28%.

3. Results and discussion

3.1. Synthesis of monomers

In cholesterol structure there is a hydroxyl group on C3 position and a double bond between C5 and C6 position. The hydroxyl group of C3 position initially was esterified by acetic anhydride [26]. Then the double bond between C5 and C6 position was oxidized to epoxy and the epoxy group was opened by using methacrylic acid. ^{1}H NMR results show that there exists obvious double bond displacement in compound **3**, suggesting the ring-opening reaction was successful. Moreover, in the FTIR spectra, the characteristic absorption peak of CH_2=CH— was found at about 1636 cm^{-1}. All the results above indicated that the monomer synthesized was the one we expected. Structurally, it is difficult to introduce large groups into the C5 position due to the steric hindrance and the methacrylate group should be connected to C6 position. The corresponding characterization was shown in the following content.

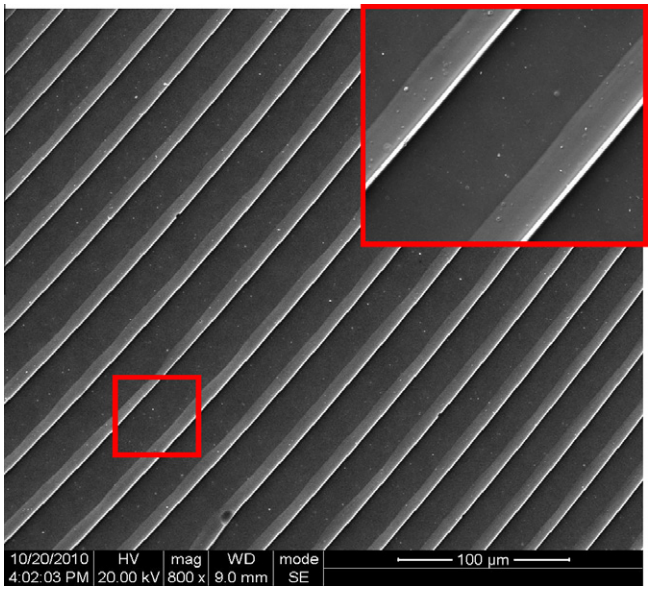


Fig. 3. SEM images of compound **3** obtained by cooling melting sample to room temperature.

3.2. DSC curve analysis

It is well known that cholesterol esters and hydrocholesterol [27] usually have liquid crystalline properties. In our experiment a question occurred in our mind that whether the final monomer starting from cholesterol ester has liquid crystalline property or not. Thermal behavior of compound **3** was characterized by DSC at a rate of 10 °C/min. The heating curve shown in Fig. 1 reveals that there is only a melting peak, and no other thermal transition peaks are detected. Moreover, no corresponding peak presents during cooling process. During the second heating and the second cooling, there are not any peaks observed. However, if the compound **3** was given enough time under room temperature, it began to crystallize again, and the melting could be observed again in the process of heating. In addition, we found that if the temperature was heated above 200 °C, polymerization of the monomer occurred. That is because the powder after being heated above 200 °C cannot be completely dissolved in ethyl acetate.

3.3. POM and SEM results

The melting and cooling process of compound **3** was also observed by POM. The monomer melting takes place after being heated to about 153 °C, and then it is isotropic liquid. What interesting is some ordered stripes were observed when the melted sample was cooled to room temperature, as shown in Fig. 2. The most prominent features are the ordered domains, which consist of parallel stripes, the distance between the adjacent two stripes ranges from 11 to 16 µm. Besides, we found this area was colorful on the surface of glass sheets. It is known that cholesteric phase exhibit iridescent colors if the helical pitch or reflected wavelength coincides with the wavelength of visible light. Even though the two glass sheets are separated from each other, the well-defined stripes still obviously present on glass sheets.

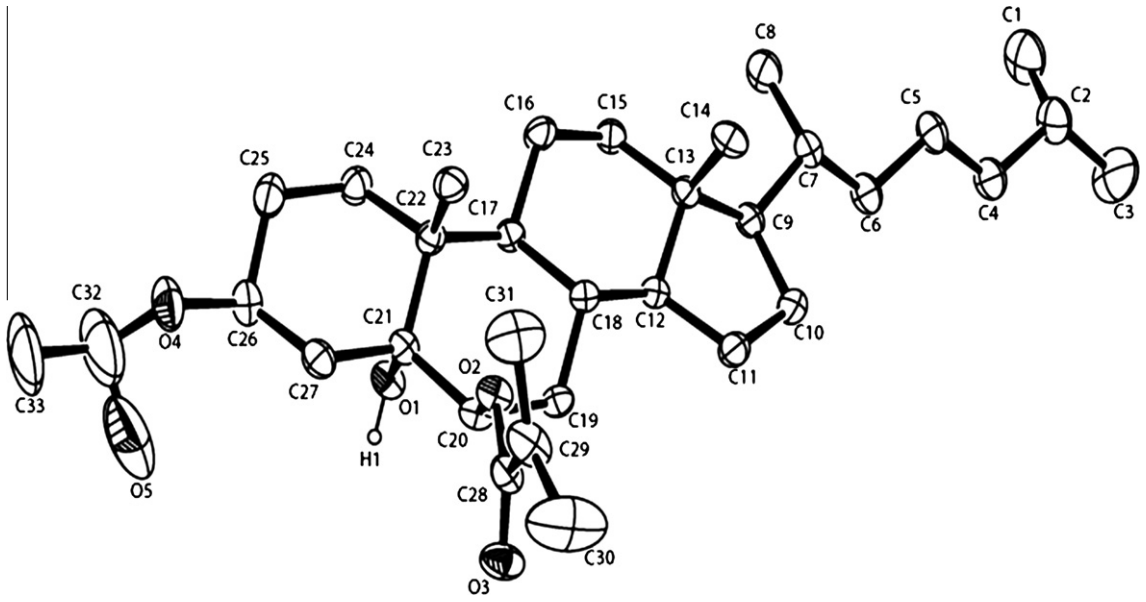


Fig. 4. The crystal structure of compound **3.**

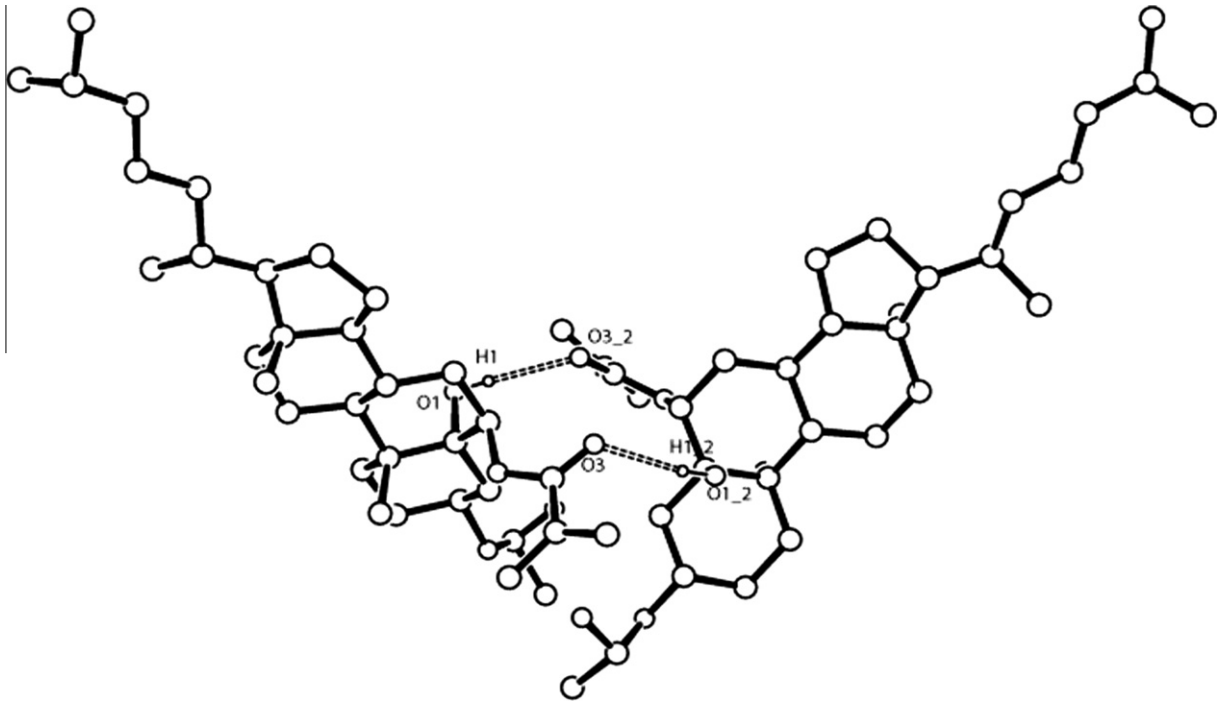


Fig. 5. The hydrogen bond between H (1) and O (3) of two molecules.

In order to identify and repeat the organization of the parallel stripe, samples were sprayed on glass sheet and covered another empty glass sheet on the sample. Then the sample was heated to the temperature above its melting point and then cooled down to room temperature. SEM was employed to characterize the morphology and microstructure. Fig. 3 shows the similar arrangement as shown in POM results. Pictures of much greater magnification could be recorded in a certain scope, one can discern the white and black regions from it. It should be noticed that the same stripes still could be observed no matter how fast the rate of quenching, and the observation is repeatable through pressing the glass plate slightly when melting, and it is repeatable.

3.4. Characterization of X-ray monocrystal diffraction

In order to study how the stripes form, we focus on the research of interaction and molecular packing of single crystal. The interactions between molecules revealed in the single crystal may offer valuable insight into the ordered structures of superamolecule. Therefore, the single crystal of compound **3** was cultivated from its acetic ether solution, and clear pictures were obtained through four-circle single crystal diffractometer. As Fig. 4 shows C3 position connects with —CH_3COO (up), C5 with —OH (down) and C6 with —CH_3C (CH_2) COO (up). The data of single crystal of **3** is shown as follows: formula: $C_{33}H_{54}O_5$, formula weight: 530.76, crystal size:

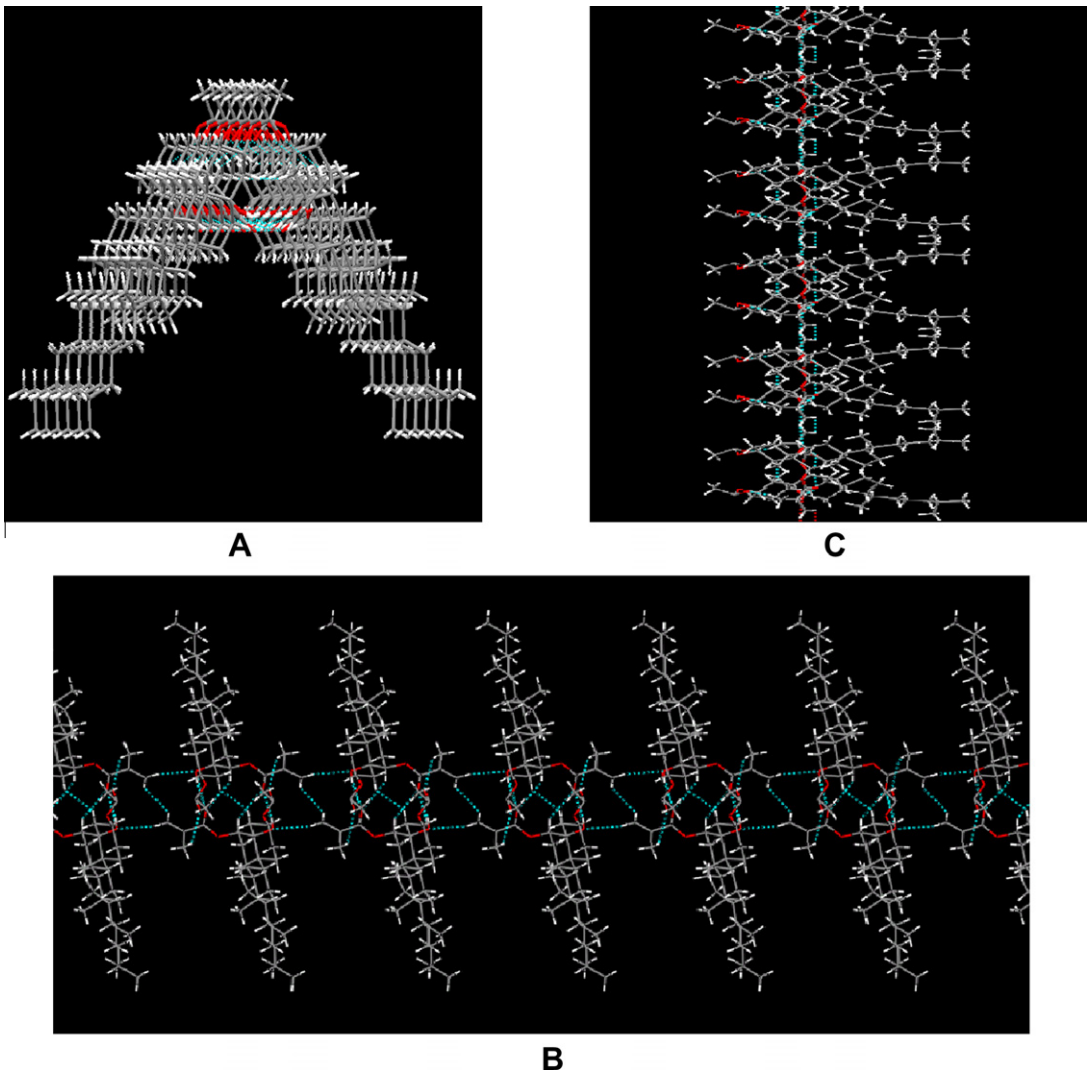


Fig. 6. Molecular packing and hydrogen bonds (green lines) formed in the single crystal of **3**. (A); (B) and (C) are views observed from a, b and c axes respectively. (For interpretation of the references to color in this figure legend, the reader is referred to the web version of this article.)

$0.36 \times 0.28 \times 0.19\ mm^3$, temperature: 296 K, crystal system: monoclinic, space group: C_2, a: 31.9922 Å, b: 9.9421 Å, c: 10.3358 Å, α: 90°, β: 93.758°, γ: 90°, volume: 3280.4 $Å^3$, Z: 4, ρ_{calc}: 1.075 mg/mm^3, $F(000)$: 1168, reflections collected: 7423, final R indices (all data): $R_1 = 0.0756$, $wR_2 = 0.1692$.

As Fig. 5 shows, there is intermolecular hydrogen bond between every two molecules. Since the group connected on C5 and C6 faces to the opposite direction, it is feasible to form strong interaction between two molecules in space. The C═O at C6 position of compound **3** forms hydrogen bond with the OH at C5 position of another molecule of compound **3**, at the same time, another hydrogen bond is formed at the opposite position. The single crystal data show the angle of hydrogen bond is 168.5° which is nearly 180°. In the basis of the above results, we assert the hydrogen bond plays an important role in the formation of ordered structures of superamolecular.

Except hydrogen bonding formed between every two molecules, van der Waals force also plays an important part in forming regular structure. After purification by silica gel column chromatography, we got the molecular solids through rotary evaporators. The solids are crystalline, and all the crystals accumulated together through van der Waals interactions because of fast dissolvent volatilizing. While, the single crystal was obtained from its acetic ether solution after slowly solvent volatilizing. There are H—bonding interactions between every two molecules which could form small aggregate as shown in Fig. 5. Meanwhile, the van der Waals force integrates these small aggregates into a whole supermolecule which presents regular arrangement of stripes in a macroscopic view. Maybe that only happens in ideal environment, however, when the solids were heated up to the melting point, the molecules desultorily arranged in the form of isotropic liquid. That is the ideal environment when the isotropic liquid was cooled at a slow rate. Every two molecules could be combined together through H—bonding. With further cooling, van der Waals force plays an important role in accumulating the small aggregates into the superamolecules. Views from a, b, c axes of the forming structure are shown in Fig. 6. Thereinto, view from b explains the regular arrays of rod stripes observed from POM. The intermolecular packing and assembling forms the superamolecule ordered structures and they are stable at room temperature.

The present observation is not identical with the general self-assembly, because it takes place during the melting cooling process rather than the traditional self-assembly in solution, which is novel and interesting.

The experiment of forming well-defined arrays is repeatable; however, the forming ordered domain is not wide enough. The

next step we will try to increase the effective domain sizes and explore the mechanism of the formation of parallel stripes. Furthermore, it is necessary to study the effects of various substrates in the self-assembly process.

4. Conclusions

New types of lateral cholesteryl derivates were synthesized. Through various methods of characterization, the parallel arrays of rod stripes were observed for hydrogen bond and van der Waals force. This novel self-assembly behavior occurred in cooling process of melted sample on common glass sheet, the process is quite simple and repeatable.

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References

[1] T.B. Yu, J.Z. Bai, Z.B. Guan, Angew. Chem. Int. Ed. 48 (2009) 1097.
[2] T.L. Breen, J.S. Tien, R.J. Oliver, T. Hadzic, G.M. Whitesides, Science 284 (1999) 948.
[3] Y. Yin, Y. Xia, Adv. Mater. 13 (2001) 267.
[4] E. Rogalska, M. Rogalski, T. Gulik-Krzywicki, A. Gulik, C. Chipot, PNAS 96 (1999) 6577.
[5] O. Ikkala, G. Brinke, Science 295 (2002) 2407.
[6] D.Z. Kan, L. Wan, J.L. Fang, X.H. Yu, Polym. Bull. 3 (2002) 5.
[7] A. Hierlemann, J.K. Campbell, L.A. Baker, R.M. Crooks, A.J. Ricco, J. Am. Chem. Soc. 120 (1998) 5323.
[8] L. Wan, K. Itaya, Langmuir 13 (1997) 7173.
[9] T. Shimooka, S. Yoshimoto, M. Wakisaka, J. Inukai, K. Itaya, Langmuir 17 (2001) 6380.
[10] H. Tokuhisa, M.Q. Zhao, L.A. Baker, V.T. Phan, D.L. Dermody, M.E. Garcia, R.F. Peez, R.M. Crooks, T.M. Mayer, J. Am. Chem. Soc. 120 (1998) 4492.
[11] H. Schönherr, V. Paraschiv, S. Zapotoczny, M. Crego-Calama, P. Timmerman, C.W. Frank, G.J. Vancso, D.N. Reinhoudt, PNAS 99 (2002) 5024.
[12] L. Simona, B. Hans-Jü rgen, Langmuir 18 (2002) 2398.
[13] N. Miyashita, H. Möhwald, D.G. Kurth, Chem. Mater. 19 (2007) 4259.
[14] Y.M. Huang, Key Eng. Mater. 12 (2010) 428.
[15] B. Wang, J.H. Zhang, Acta Chim Sin. 68 (2010) 1247.
[16] B. Wang, H.Y. Du, J.H. Zhang, Steroids 76 (2011) 204.
[17] V.A. Mallia, P.K.S. Antharjanam, S. Das, Chem. Lett. 30 (2001) 752.
[18] A. Takahashi, V.A. Mallia, N. Tamaoki, J. Mater. Chem. 13 (2003) 1582.
[19] Y.X. Zhou, V.A. Briand, N. Sharma, S. Ahn, R.M. Kasi, Materials 2 (2009) 636.
[20] Y. Nomura, M. Ikeda, N. Yamaguchi, Y. Aoyama, K. Akiyoshi, FEBS Lett. 553 (2003) 271.
[21] A. Benrebouh, D. Avoce, X.X. Zhu, Polymer 42 (2001) 4031.
[22] D. Avoce, H.Y. Liu, X.X. Zhu, Polymer 44 (2003) 1081.
[23] M. Nichifor, M.C. Stanciu, X.X. Zhu, React. Funct. Polym. 59 (2004) 141.
[24] Y.L. Chen, J.T. Luo, X.X. Zhu, J. Phys. Chem. B 112 (2008) 3402.
[25] K.L. Williamson, Macroscale and Microscale Organic Experiments, second ed., Houghton Mifflin, Boston, 1994 (pp. 300).
[26] V.A.E. Shaikh, N.N. Maldar, S.V. Lonikar, C.R. Rajan, S.J. Ponrathnam, Appl. Polym. Sci. 70 (1998) 195.
[27] J.H. Zhang, C.G. Bazuin, S. Feiberg, F. Brisse, X.X. Zhu, Polymer 46 (2005) 7266.