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Purification of Erythromycin by Antisolvent Crystallization or Azeotropic Evaporative Crystallization

Kui Chen, Li-Jun Ji and Yan-Yang Wu

1. Introduction

Crystallization plays an important role in separation and purification of the antibiotics. And it is also an indispensable step in preparation of pharmaceuticals with biological activities and specific crystal form. As the last step in purification, crystallization determines the purity, crystal habit, granularity and its distribution as well as pharmacologic effect, biologic activity and product stability [1], which are actually dependent on specific mechanism for its processes and operational conditions. So it’s necessary to study thermodynamics, kinetics and conditions of crystallization process, which helps increase the yield and reduce cost.

As a representative of macrolide antibiotics, erythromycin has been widely used since its introduction in 1952 [2]. As erythromycin derivatives, clarithromycin and azithromycin have exhibited remarkable improvement on stability in acid solutions and metabolism dynamics [3, 4]. A lot of researches have been done on new derivatives with features of combating drug resistance [5, 6]. In the meanwhile, high-purity erythromycin, as the raw material, is fundamental to produce its pharmaceutical derivatives.

Erythromycin is obtained from microbial fermentation in industry. Subsequent separation and purification involve multiple unit operations, such as extraction, absorption, chromatography and crystallization. Different process involves different combinations of unit operations [7].

Among them, solvent extraction accompanied with intermediate precipitation is widely used, in which butyl acetate is commonly adopted to extract erythromycin from the fermentation filtration. It is followed by reactive crystallization to form an intermediate prior to conversion into erythromycin alkaline and dissolving in acetone. Lastly, erythromycin is pu-
rified by antisolvent crystallization [8]. That is to say, both reactive crystallization and antisolvent crystallization have to be employed in this separation process.

In contrast, the technological process with membrane separation and resin absorption [9] is drawing more attention compared with the traditional solvent extraction in the above [10]. This process usually goes as follows: firstly, microfiltration is used to remove solid impurities from the fermentation broth, and the filtrate is purified by macroporous absorption resin, and then the adsorbed erythromycin is eluted with butyl acetate. Finally, either evaporative crystallization or reactive crystallization can be used to obtain the final product [11].

![Figure 1. Schematic diagram for the purification erythromycin](image)

The flowsheet of these two technological processes is demonstrated in Figure 1. It can be seen that crystallization is the final step to prepare erythromycin no matter which one is adopted. Different crystallization method has been used for different pretreatment.

Crystallization is a complex process involving mass transfer, heat transfer and surface reactions, which includes the formation of a supersaturated solution, nucleation and crystal growth. The operating parameters of crystallization process, such as temperature, agitation intensity and seed crystals, can affect the generation rate and scale of the supersaturation. The structure of the crystallizers and stirrer will influence the fluid mechanics properties of the crystallization system. Thus it can be seen that all these factors profoundly influence crystal nucleation and growth [12]. Over a long period of time, the crystallization processes have been carried out on according to experiences rather than theoretical researches due to the little study on thermodynamics and kinetics. Not surprisingly, it’s hardly to obtain erythromycin with high purity, complete crystal form, narrow distribution of crystal size, less crystal bonding, which are very important for the stability and bioavailability of the drug.
In this paper, two crystallization processes of erythromycin in different systems, which include the antisolvent crystallization for mixed solvents of acetone and water and the azeotropic evaporative crystallization for butyl acetate-water system, are described in details. The thermodynamics and kinetics of the crystallization of erythromycin, which help to thoroughly understand the effect of a variety of factors on the nucleation, crystal growth and crystal habit, are summarized systematically. On the basis of these fundamental studies, effective control techniques are proposed to improve the quality of erythromycin product.

2. Purification of erythromycin by antisolvent crystallization

In the solvent extraction process for purification of erythromycin, erythromycin alkaline was converted from erythromycin thiocyanate by adding ammonia or NaOH solution and dissolving in acetone. Then erythromycin product was prepared by antisolvent crystallization, in which water was served as antisolvent.

The traditional crystallization process, which was too simple, only involved modulating two indicators including antisolvent quality and crystallization temperature.

Water was poured into erythromycin acetone solution at room temperature, and then the product was obtained by filtration after standing for a period of time. Such operation made obvious differences of supersaturation, nucleation rate and crystal growth rate and then caused the discrepancy in product quality for different batch.

2.1. Solubility and metastable limit of erythromycin

As we know, the phase equilibrium between solid and its solution is fundamental to choose crystallization method and also determines the maximum yield of solution crystallization [12]. Erythromycin is soluble in acetone, but insoluble in water [13]. Thus, erythromycin can be precipitated by adding water into erythromycin acetone solution.

2.1.1. Solubility

The solubility of erythromycin in acetone increases with the increasing temperature, whereas it becomes less soluble with the higher temperature in water. So, the solubility of erythromycin in acetone-water binary solvent system is influenced by the solvent composition and temperature.

Some data on solubility of erythromycin in acetone-water solution was reported in literatures [14,15]. In this paper, the solubility above 303.15K has been measured. As can be seen in Figure 2, the solubility of erythromycin in the medley acetone-water solution increased with increasing acetone concentration and increasing temperature, respectively. In the same range of acetone content, the slope of the solubility curve increased with increasing temperature, which meant the rate of increase of erythromycin solubility increased.
The impact of acetone on the solubility of erythromycin increased as the mass fraction of acetone increasing. It was not hard to infer that the difference of the solubility at different temperatures tended to decrease with the mass fraction of water increase.

An empirical model was proposed to relate the experimental data of the solubility of erythromycin and the parameters was obtained by fitting. The empirical equation for the solubility of erythromycin in mixed solvents of acetone and water was expressed as below:

$$C^* = \frac{1.02 \times e^{0.1767x}}{0.395 - 0.00383T + x^{0.038T + 1.295}}$$

where $C^*$ was solubility (g Erythromycin/100g Acetone-Water Solution) and $x$ was the mass proportion of water to acetone ($x=m_w/m_a$). Equation (1) was practicable in the range of 293.15K≤$T$≤323.15K, 1.0≤$x$≤2.0.

Equation (1) could be used to calculate erythromycin solubility $C_{Cal}$ under various experimental conditions. The calculated solubility $C_{Cal}$ and the experimental solubility $C_{Exp}$ were shown in Figure 3. It was indicated that Equation (1) was appropriate to predict the solubility of erythromycin within the range of temperature and acetone concentration for the equation.
2.1.2. Metastable zone

Metastable zone width is fundamental to choose suitable supersaturation of crystallization. It is also used as a restrictive operating condition to avoid crystallization system going to unstable zone [16] that results in the worse product.

Figure 3. Comparison of simulated value and experimental data of solubility

Supersolubility of erythromycin was measured by the method of laser scattering [15]. As shown in Figure 4, the experimental device consisted of crystallizer, mixing system, feeding system, temperature control system and detection system. Wherein, the crystallizer was a double-wall kettle with internal diameter 75mm and height 130mm. Stirrer with four inclined propellers was driven by variable speed motor, the propeller diameter was 12mm, and the stirring shaft diameter was 5mm. The peristaltic pump continuously pumped antisolvent water at a fixed temperature into crystallizer. The detection system consisted of He-Ne laser generator and laser power detector. He-Ne laser generator outputted 632.8nm red line, scattering and diffraction occurred when monochrome laser beam encountered with body of similar length of wavelength, the laser intensity received by detector was drastically reduced, thus the nucleation could be detected.

The relationship between metastable zone width ΔC of erythromycin and solvent composition at 323.15K was shown in Figure 5. It could be seen from the figure that metastable zone width decreased gradually with the increase of the quality of water in solution. In mw:ma range of 1.0 to 1.8, the supersolubility presented apparent downward trend. After mw:ma reached 1.8, the change of the metastable zone width weakened, but the metastable zone width of this region was already narrow and was not suitable for crystallization operation.

The equation was obtained by correlating the metastable zone width and solvent composition, which was listed as follows:

$$\Delta C = 3.09 \times x^{-2.40}$$

where $x$ was the mass ratio of water to acetone, $x=m_w:m_a$.

![Figure 5. Effect of solvent composition on metastable zone width of erythromycin at 323.15K](image-url)
As shown in Figure 6, the metastable zone width of erythromycin decreased with the increase of temperature. The metastable zone width was similar at 308.15K and 313.15K, while it was quite different at 313.15K and 323.15K, which indicated metastable zone width was temperature sensitive in the range of 313.15K to 323.15K. The variation of metastable zone width with agitation power presented a consistency at different temperatures. The metastable zone width was wider under the same agitation power at lower temperature.

2.2. Antisolvent crystallization kinetics of erythromycin

In this paper, the intermittent dynamic method [17] was used to study the kinetics of erythromycin antisolvent crystallization under different conditions. With the empirical models deduced from the Larson-Randolph population balance equation [18,19], the model parameters were obtained from the experimental data through the matrix conversion and the method of linear squares regression. Thus, the equations of nucleation and crystal growth of antisolvent crystallization of erythromycin were established to help find the suitable operation parameters.

The experimental apparatus were shown in Figure 4. Firstly, at the start of recording the time, antisolvent water at set temperature was poured into the erythromycin-acetone solution in the crystallizer. Once the nucleation appeared in the solution, water was stopped im-
porting and the time was recorded. Then the agitation rate and temperature were maintained constant. It was sampled at different interval of time. The indexes of each sample, such as magma density, degree of supersaturation and crystal size distribution (CSD) of production, were measured respectively.

2.2.1. Crystal size correlation of crystal growth

The crystal nucleation and growth kinetics were solved according to the size-independent model [16], using a set of the experimental data of magma density and CSD at 323.15K. The calculated value was in good agreement with the experimental data, as shown in Figure 7. In the crystal size \(L_i\) range, erythromycin crystal population density logarithm \(\ln n_i\) was basically a straight line. At the same time, the results of matrix conversion also showed that the use of size-dependent model to describe the crystal growth was of large error. Therefore, erythromycin crystal growth was size-independent.

![Figure 7. Typical population density distribution of erythromycin](image)
2.2.2. Kinetics model

On the basis of the above, the size-independent model was adopted to describe the crystal growth rate of erythromycin. According to the study on the effects of temperature, agitation and dosing rate of antisolvent on nucleation rate and crystal growth rate, the corresponding equations for nucleation rate and crystal growth rate were shown as follows:

The nucleation equation

\[ \beta = 3.23 \times 10^{14} \exp\left(\frac{-2343}{T}\right)P_V^{0.378}M_T^{0.317} \]  

(3)

The crystal growth equation

\[ \Delta c = 1.18 \times 10^{-3} \exp\left(\frac{4539}{RT}\right)P_V^{0.102} \]  

(4)

where \( M_T \) was magma density (kg/m\(^3\)), \( P_V \) was unit volume of stirring power (W/m\(^3\)).

In the antisolvent crystallization of erythromycin, slurry density had less effect on the nucleation rate than supersaturation did. The influence of stirring intensity and supersaturation on nucleation rate was greater than those on crystal growth rate. The supersaturation series 3.303 in the nucleation equation was much smaller than the primary nucleation kinetics series [12]. So the mechanism of nucleation of antisolvent crystallization of erythromycin was secondary nucleation.

2.2.3. Online study of crystallization process

In order to further reveal the intrinsic principles of the antisolvent crystallization process of erythromycin, the Focused Beam Reflectance Measurement (FBRM) technique was adopted to monitor in situ the variation of crystal quantity and crystal size distribution in this paper.

The total number and the chord length distribution (CLD) of crystal particles were measured by using the equipment and method shown in literature [20]. A mathematical procedure based on Monte Carlo simulation was established to transform the CLD into CSD.

The change of the number of crystals and CSD of erythromycin antisolvent crystallization were studied under different temperature and feeding rate of antisolvent. The results indicated that the faster water was fed, the earlier new crystals came into being, the faster the crystal grew at the initial stage. The total number of crystals at the stable stage tended to decrease as temperature increased [20].
Figure 8. CSD after peak value of overall crystal number count at 308.15K; ■: 0min; ●: 30min; ▲: 90min; ◆: 180min

Figure 9. CSD after peak value of overall crystal number count at 314.15K; ■: 0min; ●: 30min; ▲: 90min; ◆: 180min
The proportion of particles of different size was the grain size frequency distribution. Figure 8 and Figure 9 showed the size frequency distribution curve after nucleation at 308.15K and 314.15K respectively. As could be seen from those, the curves were similar at different temperatures, which were sharp and steep. Particle size which was less than 20μm accounted for the vast majority and the peak of the curves was close to 20%.

It could be found from Figure 8 and Figure 9 that the number of both small size crystal and large size crystal hardly change with time. It meant that particles with small size were constantly dissolving, while saturated solute of erythromycin was precipitated to form new crystal, or the existing crystal grew larger in volume. The dissolution and precipitation of erythromycin reached equilibrium.

In order to properly characterize the crystal growth, volume mean diameter $D_V$ (also known as $D_{43}$) which was the equivalent diameter of the particles with same volume (or mass), was used to investigate the changes of crystal size with time at different temperatures. As shown from Figure 10, erythromycin $D_V$ monotonically decreased with the increasing temperature at the same crystallization time.

![Figure 10. The effect of temperature on $D_V$ of erythromycin crystal; ■- 0min; ○- 30min; ●- 60min; ◦- 90min; ▲- 120min; △- 150min; ◆- 180min](image-url)
2.3. The antisolvent crystallization technique of erythromycin

The thermodynamics and kinetics of the antisolvent crystallization of erythromycin were summarized systematically to understand thoroughly the effect of a variety of factors on the nucleation, crystal growth and crystal habit. On the basis of these fundamental studies, appropriate technological parameters were explored to develop the efficient industrialized crystallization process of erythromycin.

2.3.1. Technological parameters

Crystal quality, such as crystal purity, crystal habit, crystal size, and CSD, was related closely to the crystallization conditions. Accordingly, the effect of the dosing rate of antisolvent, crystallization time, stirring intensity and crystallization temperature on CSD of erythromycin was studied in details in this paper.

**Dosing rate of antisolvent** For antisolvent crystallization of erythromycin, the dosing rate of antisolvent determined the generation rate of supersaturation, and also affected the rate of nucleation and crystal growth.

The definition of dosing rate of antisolvent was the importing water volume of per unit time and per unit volume of erythromycin-acetone solution.

\[ \nu_d = \frac{V_w}{V} \]  

where \( \nu_d \) was dosing rate of antisolvent (min\(^{-1}\)), \( V_w \) was the volume rate of importing water (mL/min), \( V \) was the erythromycin-acetone volume (mL).

Figure 11 showed the relationship between the dosing rate of antisolvent \( \nu_d \) and erythromycin CSD, where \( d_p \) was the crystal diameter and \( R_v \) was the cumulative volume fraction. It could be seen from the figure that the proportion of crystals with large size increased with the increasing dosing rate of water, but the CSD tended to disperse. While the CSD of crystals obtained in lower water dosing rate was more concentrated.

Therefore, in process of the crystallization, an appropriate increase in generation rate of supersaturation could speed up the crystallization rate and improve the capability of the crystallizer. However, the rapid generation of crystals will increase the chance of crystal breakage and secondary nucleation and make the CSD disperse.

**Crystallization time** The cumulative volume distribution at different crystallization time was shown in Figure 12, where \( d_p \) was the crystal diameter and \( R_v \) was the cumulative volume fraction. As could be seen from the figure, the increase of the crystallization time was conducive to crystal growth, while the CSD did not tend to concentrate. The crystal growth needed some time, however, long time crystallization couldn’t promise CSD being more consistent.
Figure 11. CSD based on cumulative volume of erythromycin at different water-pumping velocities; ▲: 0.0138 min⁻¹; ●: 0.188 min⁻¹; ■: 0.024 min⁻¹; ◆: 0.0389 min⁻¹

Figure 12. CSD of erythromycin based on cumulative volume at different crystallization times; ▲: 40 min; ●: 50 min; ■: 70 min; ◆: 100 min
**Agitation power** The CSD was the result of the interaction of primary nucleation, secondary nucleation, and crystal growth. Meanwhile, agitation power had a significant impact on all the above. Figure 13 showed the particle volume distribution of erythromycin at different stirring intensity, where $x_v$ was the particle volume distribution.

It could be seen from Figure 13 that the erythromycin product had the widest CSD and the highest proportion of small size crystals when the stirring power was 13.99 W/m$^3$, and the distribution curve had smearing phenomenon in the range of large particle size. While the crystal had the narrowest CSD and the lowest proportion of small size crystals when the stirring power was 1.749 W/m$^3$, and the distribution curve had no smearing. The energy imported by stirring was conducive to nucleation and crystal growth. In the meanwhile, crystal breakage could easily occur with too strong stirring, while the obvious differences of supersaturation would occur with too weak stirring and then caused variation of rate of nucleation and crystal growth.

**Crystallization temperature** Figure 14 showed the variation of volume mean diameter ($D_v$) of erythromycin at different crystallization temperature. $D_v$ decreased with the increasing of temperature. The previous thermodynamic study showed that the metastable zone width of erythromycin reduced with the increase of temperature. The intensified thermal motion of molecule caused by the increasing temperature accelerated the frequency of contact and collision of crystals, and then promoted the formation of tiny crystals, and decreased the supersaturation required for nucleation. On the other hand, the driving force of crystallization
decreased with the narrowing metastable zone width, so did the rate of crystal growth. Therefore the volume mean diameter of the crystals decreased as the temperature increased.

Figure 14. Volume mean diameter of erythromycin at different temperatures

2.3.2. The novel technique of antisolvent crystallization of erythromycin

For the traditional antisolvent crystallization, water was poured into erythromycin acetone solution at room temperature. Then after standing for a period of time, the erythromycin alkaline product was obtained by filtration.

It was not difficult to find the shortages of this crystallization method. Firstly, the dosing rate of antisolvent was too fast. When the antisolvent water was fed rapidly, the supersaturation formed suddenly and leaded to the outbreak of the nucleation. Nucleation was active and occupied the dominant position of the crystallization process. Meanwhile, the impurities easily accompanied with crystals by precipitation in the fast crystallization process. Secondly, stirrer and stirring intensity were inappropriate. Poor mixing effect made uneven distribution of supersaturation, so it was hard to obtain erythromycin with complete crystal form and narrow distribution of crystal size [12,21]. Thirdly, crystallization temperature was uncontrolled. Then the differences of solubility between erythromycin and impurities in acetone-water solution could not be fully explored to improve the separation efficiency.

The operation of the crystallization mentioned above lacked of crystallization process control and could not play a good role in purification of erythromycin by crystallization. Then the erythromycin product would be highly influenced by fermentation broth and pre-purifi-
cation. That was to say, the quality of erythromycin was restricted by erythromycin thiocyanate. So it was hard to obtain the erythromycin product with stable and high quality and yield.

There were some other studies [8,22] on the improvement of erythromycin crystallization method by adding seed crystals.

On the basis of thorough research on the antisolvent crystallization process of erythromycin, a novel technique for antisolvent crystallization of erythromycin by dynamic control of temperature and stirring power was proposed in this paper, which was listed as follows.

1. Dosing the antisolvent. The polarity of mixed solvents was changed gradually when the antisolvent was imported into erythromycin acetone solution slowly. In the meantime, the solubility of erythromycin decreased gradually until crystal nucleus formed. The supersaturation could be controlled within the thermodynamic metastable zone by dosing antisolvent continuously, the crystal growth was moderated and in order, and the CSD of erythromycin tended to be narrow.

2. Appropriate stirring intensity. The suitable stirring power could be conducive to maintaining uniform supersaturation and crystallization rate. Meanwhile, stirring could promote dynamic balance of crystallization and dissolution, and reduce the crystal bonding, and then improve the purity of the crystal.

3. Increasing nucleation temperature. Substance usually had higher solubility at a higher temperature, so did the impurities. Increasing nucleation temperature could reduce the chance of impurities precipitation and improve the purity of erythromycin.

4. Cooling crystallization and aging with lower stirring intensity. After nucleation at high temperature, the stirring power should be reduced to avoid excessive shear force on the crystal collision and maintain a uniform concentration distribution in the slurry at the same time. Then, the supersaturation produced by cooling maintained crystal growth at a steady rate after dosing all antisolvent. Lastly, aging with lower stirring power at lower terminal temperature could improve the quality and yield of product.

On the basis of the above, the key operation parameters which affect the quality of crystal, such as temperature, dosing rate of antisolvent and stirring intensity, were determined by measuring the crystal shape, titer and yield [23]. Then the novel technique of erythromycin antisolvent crystallization was established in this paper, which was characteristic of dynamic control of temperature and stirring intensity [24].

Figure 15 and Figure 16 showed the crystal shape and CSD of industrial erythromycin products obtained by the traditional method (a) and novel technique (b), respectively. For the crystal shape, product (b) had a more regular and bigger size than product (a) did. For the CSD, product (b) was narrower. For titer, product (b) was 935.6 U/mg, while product (a) was 920 U/mg. Those meant that the quality of erythromycin had been improved by the novel technique of antisolvent crystallization [23].
In the commercial use of the antisolvent crystallization process, erythromycin with high specific activity was obtained at high yield. Over 90% of the products met the demands per year, which was much higher than the 53% with the traditional crystallization process.
3. Purification erythromycin by azeotropic evaporative crystallization

The development of the crystallization technique of erythromycin is limited to some extent by the extraction and purification prior to the crystallization. Taking the production of erythromycin as an example, the widely used process is frame filtration of fermentation broth - solvent extraction - salting-out crystallization – alkalization - antisolvent crystallization. Due to the limited interception capability for fine particles and macromolecules impurities such as proteins by frame filtration, and the low selectivity of the object over pigment and the small un-ionized organic molecules by solvent extraction, the impurity content is high in the organic phase. Therefore, the object should be further purified by coupling two crystallization methods in the subsequent refining process.

In recent years, a different technological process by membrane separation and resin absorption is gradually introduced into industrial application [25,11]. The process consists of several steps including membrane separation, resin absorption, elution and crystallization. Firstly, microfiltration is used to remove mycelium, a variety of fine suspension particles and some protein from fermentation broth, then pigment and small un-ionized organics are removed by resin absorption and the elution with butyl acetate. An improvement of the purity of erythromycin butyl acetate solution is obtained by using this pretreatment. And it makes crystallization preparation of erythromycin alkaline from butyl acetate elution become possible.

For the preparation of erythromycin alkaline from erythromycin butyl acetate solution, the product yield is low due to the high solubility of erythromycin. So the urgent task is to increase the yield. To remove butyl acetate is feasible, while high temperature for solvent evaporation may cause the destruction of erythromycin. Although erythromycin has better thermal stability than some other sorts of antibiotics, there is no precedent on the separation and purification of erythromycin with temperature being above 323.15K in industrial application till now. Thus, azeotropic evaporative crystallization of erythromycin is proposed in this paper. The method takes erythromycin, butyl acetate and water as crystallization system. Then butyl acetate-water azeotrope is removed by vacuum azeotropic evaporation to make erythromycin precipitate and disperse into water. Excessive water is added to the erythromycin butyl acetate solution for azeotropic evaporation, which can also play a role of washing crystals. The solubility of butyl acetate in water is quite small, so the azeotrope is easy to split into two phases at room temperature. The schematic diagram of azeotropic evaporative crystallization of erythromycin is demonstrated in Figure 17.

The solubility of erythromycin in butyl acetate-water saturated solution (solution A) and in water-butyl acetate saturated solution (solution B) was detected, respectively. The result indicated that the solubility of erythromycin in solution A was quite low and had little change with temperature. So for the azeotropic evaporative crystallization of erythromycin, the proportion of water was based on its effect on operation, such as the viscosity of the solution and crystal dispersion, as well as the utilization of equipment and the efficiency of production, rather than on the yield of crystallization.
3.1. Technological parameters

Once the azeotropic evaporative crystallization of erythromycin was established, the optimization of parameters was directed by the quality and yield of crystal. The process parameters related to the crystal shape, crystal size and CSD were shown as follow: firstly, the supersaturation, which was related to the quantity of butyl acetate removed by azeotropic evaporation; secondly, the generation rate of supersaturation, which was dependent on the azeotropic evaporation rate and the cooling rate; thirdly, the crystallization temperature, which was bound up with vacuum of system and the cooling rate; fourthly, the stirring intensity, and etc. The yield of erythromycin was determined by the evaporation quantity of butyl acetate and the terminal crystallization temperature.

**System vacuum and operation temperature** The butyl acetate-water azeotrope was removed from the crystallization system by vacuum evaporation, and the azeotropic temperature varied with the pressure. Then the high temperature leading to the damage of erythromycin could be avoided by adjusting the pressure.

According to the phase equilibrium data reported in the literatures [26,27], the azeotropic temperature and composition under different vacuum was calculated by using Pro II simulation software and NRTL thermodynamic model. When the system vacuum was controlled above 0.084MPa, the crystallization temperature was below 323.15K.

**Supersaturation** The supersaturation of erythromycin increased with the increasing volume of butyl acetate evaporated. The supersaturation varied with temperature and evaporation volume of butyl acetate, which affected CSD of the product.

Figure 18 showed the relationship between the cumulative volume distribution and supersaturation at 316.15K, where \( d_p \) was the crystal diameter and \( R_v \) was the cumulative volume fraction. As can be seen from the figure, the crystal size of erythromycin increased with the increase of supersaturation. However, it was necessary to choose the supersaturation range carefully due to the variation of solution viscosity and the difficulties of the crystal dispersion.

**Cooling rate** The cooling crystallization started after evaporating some amount of butyl acetate. The supersaturation caused by cooling made the crystallization process proceed continuously. It could improve the quality yield of the product by reducing the terminal crystallization temperature.
Figure 18. CSD based on cumulative volume of erythromycin at different supersaturation - ■: 39.91 g/100g; - ▲: 40.76 g/100g; - ●: 41.26 g/100g; - ◆: 41.60 g/100g

Figure 19 showed the relationship between the cumulative volume distribution of erythromycin and cooling rate, where $d_p$ was the crystal diameter and $R_v$ was the cumulative volume fraction of the crystal. It could be seen from the figure that speeding up the cooling rate was not conducive to the growth of crystal and made the crystal size decrease.

Figure 19. CSD based on cumulative volume of erythromycin at different cooling rate - ■: 273.17K/min; - ●: 273.20K/min; - ▲: 273.28K/min
3.2. The technique of erythromycin azeotropic evaporative crystallization

On the basis of the studies above, the crystallization technique combining the azeotropic evaporation with cooling crystallization was established to prepare the erythromycin from erythromycin butyl acetate solution directly. This process included mainly the following steps: firstly, introduction of entrainer. Adding entrainer (water) to erythromycin butyl acetate solution could form azeotropic crystallization system and decrease the evaporation temperature; secondly, vacuum evaporation. Adjusting the vacuum could promise the azeotropic evaporation temperature of butyl acetate and water was low enough to avoid the destruction of erythromycin; thirdly, appropriate evaporation quantity of butyl acetate. The supersaturation could be maintained within the thermodynamic metastable zone by adjusting the evaporation quantity of butyl acetate; fourthly, modulating cooling rate. The rate of crystallization could be regulated by adjusting cooling rate, so the supersaturation produced by cooling also could be maintained within the thermodynamic metastable zone to promise crystal growth; finally, the agitation power should be adjusted with the variation of crystallization stages.

There was an application for erythromycin purification by azeotropic evaporative crystallization. The technological conditions were listed as follows, the raw material of erythromycin was provided by a pharmaceutical company, the volume of water in the crystallization system was three times the volume of butyl acetate, the supersaturation was about 45g erythromycin/100g butyl acetate, cooling rate was 273.22K/min, the terminal crystallization temperature was 303.15K. With the conditions above and the technology in this paper, the purity of erythromycin A in the product was 95.87% and the yield in mass was 75.7%, which was higher than the yield 64.6% of erythromycin product by traditional antisolvent crystallization process using the same batch of raw materials.

4. Conclusion

In this paper, the thermodynamics, crystallization kinetics and operating conditions were studied systematically for the antisolvent crystallization of erythromycin. A brand-new technique with dynamic control of temperature and agitation intensity was henceforth presented. This process included nucleation at high temperature (313.15K–323.15K), regulation of temperature and agitation power according to the different stage of nucleation, crystal growth and crystal aging. It made the operation parameters of crystallization process more reasonable, and the erythromycin with high specific activity had high yield. The commercial use of the antisolvent crystallization technique had been successful.

Meanwhile, a novel purification method of erythromycin by azeotropic evaporative crystallization was also put forward. With this method, erythromycin could be produced from erythromycin butyl acetate solution directly. By the introduction of water, the evaporation temperature of azeotrope of butyl acetate and water was decreased and the supersaturation was induced. Then, crystallization nucleation and crystal growth were controlled by the regulation of cooling rate. With the azeotropic evaporative crystallization, qualified erythromycin...
cin product could be obtained without recrystallization, which led to less solvent consumption, simplified purification process and crystal product with narrow size distribution and perfect crystal shape.

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References


