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Application of Macroporous Resin to Recover and Purify Sucralose-6-ester from the Crystalline Residue

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A method for recovering and purifying the sucralose-6-ester from the crystallization residue was investigated by using macroporous resin. The residue containing the target compound was dissolved in water to prepare sample solution. Static adsorption/desorption characteristics of three macroporous resins for separating sucralose-6-ester from crude extracts have been studied, and D101 resin was chosen for further experiment. The dynamic adsorption/desorption experiments were carried out to optimize the separation process. Under these optimization conditions, the purity of sucralose-6-ester was increased from 21.3% to 64.7% in two separations. After separated twice, it reached the purity of sucralose-6-ester in the mixture of chlorination reaction, and it can be added to the conventional process for further separation and purification. This study provides a novel, effective, environmental and economical method for recovery and purification of sucralose-6-ester.

Key words: Sucralose-6-ester, Macroporous resin, Separation, Recovery.

Sucralose is a intense sweetener derived from sucrose by replacing the hydroxyls in the 4,1', and 6' position with chlorine.[1] The chemical structure of sucralose is shown in Figure 1. It is 600 times sweeter than sucrose,[2] does not participate in human metabolism[3], and is one of particular interest for use as low calorie sweetener to replace saccharin in various products, including foods, candy, beverages and orally received medicines such as cough drops[4]. Sucralose has the characteristics of high safety and particularly it exhibits the stability in acid aqueous solution[5]. Owing to these advantages, the sucralose is one of the most popular super sweeteners in the market.

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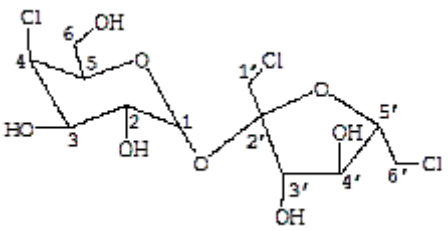

Fig. 1. Chemical structure of sucralose

According to the molecular structure of the sucralose, a major problem of synthesis sucralose is in the field of how to only direct the chlorine atoms to the desired positions[6]. Until now, several different routes for preparing sucralose have been developed, and the main synthesis methods are omni-group protection method[7-8], mono-group protection method[9-10] and chemistry-enzyme method[11-12] at presence. Among these methods, the second is widely used owing to its low cost, mild reaction conditions and high yield etc.

In the preparation of sucralose, the sucralose-6-ester is the key intermediate. Due to the activity of the 8 hydroxy of the sucrose, the chlorination reaction mixture contains many byproducts during the synthesis of sucralose-6-ester, such as monochlorosucrose-6-ester (believed to comprise primarily of 4- and 6'-monochloro isomers), 4,6'-dichlorosucrose-6-ester, 1',6'-dichlorosucrose-6-ester, tetrachlorosucrose-6-ester, polymers etc.[6] And their polarities are as follows: monochlorosucrose -6-ester > dichlorosucrose-6-ester > sucralose-6-ester > tetrachlorosucrose-6-ester.

In the industrial production of sucralose, after the chlorination process, high-purity trichloro-sucrose-6-ester is obtained by extraction and crystallization operations. As the extremely similar polarity among the desired product and impurities, it is inevitable to leave part of trichloro-sucrose-6-ester in the crystalline residue. However, until now, there is no study on how to recover the valuable substance from the residue.

Macroporous resin technology is already very developed and used in industrial production, which was found some advantages including simple procedure, high yield, low cost, easy regeneration and less poisonous organic residues.[13]

In this paper, we report a suitable for industrialization separation and purification method to recovery the sucralose-6-ester from the crystalline residue by using column chromatography of macroporous adsorption resins. The recovered sucralose-6-ester can be used as those newly synthesized to obtain sucralose.

MATERIALS AND METHODS

The crystalline residue of sucralose-6-ester was provided by JK Sucralose Inc. (Jiangsu, China). Methanol was of high-performance liquid chromatography (HPLC grade) from Merck KGaA, ethanol was of analytical grade (AR) from Sinopharm Chemical Reagent Co.,Ltd.

Adsorbents

Macroporous resins, D101 was purchased from Sinopharm Chemical Reagent Co.,Ltd (Shanghai,China), DM301 and S-8 were purchased from Anhui Sanxing Resin Technology Co.,Ltd (Anhui,China). The resins were pretreated before use according to the manufacturer. Each kind of resin was placed in a drying oven, dried at 70°C for 4 h, and then weighted precisely.

Preparation of sample solution

There is ethyl acetate contained in the sucralose-6-ester residue provided by JK Sucralose Inc, to avoid the organic solvents affect the adsorption and desorption of sucralose-6-ester in the resin, the ethyl acetate was removed by vacuum distillation at 50°C for 2 h. Then the residue was soaked in water and continuously stirred for more than 12 h, filtrated to obtain the sample solution, and detected by HPLC, the content of sucralose-6-ester is 21.3%.

HPLC Analysis of Sucralose-6-ester

HPLC analysis was performed on a Shimadzu LC-10AT series HPLC system equipped with a LC-20AD pump, and a RID-10A detector. HPLC separation was achieved by using a Venusil HILIC analytical C18 column (4.6×250 mm, 5 µm, Agilent Technologies) at 40°C. The mobile phase consisted of methanol-water (40:60, V/V), samples (20 µL) were eluted at a flow rate of 1 mL/min.

Static adsorption and desorption tests

Static adsorption experiments of sucralose-6-ester on macroporous resins were performed as follows: a certain amount of wet resin (equal to 1 g dry resin) together with 20 mL of sample solution contained 18.1 mg/mL sucralose-6-ester were added to a flask and shaken (150 rpm) for 12 h at room temperature. After reaching the adsorption equilibrium, the supernatant was separated from the resin and analyzed by HPLC. Then the saturated resin was washed with 2.0 mL distilled water. 10 mL 70% ethanol was added in the flask, followed by shaking at a frequency of 150 rpm at room temperature for 6 h to estimate desorption ratio of sucralose-6-ester from the resin. The concentration of sucralose-6-ester in the ethanol solution was measured. All experiments were repeated three times for accuracy.

The adsorption/desorption properties including the adsorption capacity, adsorption ratio and desorption ratio of each resin were calculated employing the following relationships:

$$q_e = \frac{(C_0 - C_e) \times V}{W} \quad ...(1)$$

$$E = \frac{(C_0 - C_e)}{C_0} \times 100\% \quad ...(2)$$

$$D = \frac{C_d \times V_d}{(C_0 - C_e) \times V} \times 100\% \quad ...(3)$$

Where q_e is the adsorption capacity at equilibrium (mg/g dry resin), E and D are the adsorption ratio and desorption ratio, respectively, C_0, C_e and C_d are the respectively initial, equilibrium and desorption concentrations (mg/ml) of sucralose-6-ester in the solutions. V and V_d are the solution volumes (ml) of the initial sample solution and desorption solution, respectively, and W is the weight of dry resin employed (1 g).

Resin selectivity is defined to compare the content of the object in product and feed solution in the desorption processes. The selectivity value was obtained from:

$$K = \frac{\alpha}{1 - \alpha} \times \frac{1 - \alpha_0}{\alpha_0} \quad ...(4)$$

Where α and α_0 are the contents of sucralose-6-ester in eluent and feed solution, respectively.

Dynamic adsorption and desorption

According to the static experiment results, the resin D101was selected for the dynamic adsorption and desorption experiments. The effect of the initial concentration, flow rate and the eluting solvent were also studied to ascertain the optimum dynamic adsorption and desorption conditions on D101 resin.

A certain amount of adsorbent (50 ml) was poured into the packed chromatography column (2 cm ×20 cm) at room temperature. The sample solution was then flowed into the bed at different concentrations(9.22, 12.3, 18.4 mg/ml). In addition to the initial concentration, the effect of flow rate (1.0 bed volume (BV)/h, 1.5 BV/h, and 2.0 BV/h) on the adsorption process was also investigated. For the breakthrough point, when the concentration in the leak solution is 5% of the initial concentration, adsorption is presumed to have reached saturation, the eluent was analysed at each 0.2 BV interval by HPLC.

After reaching adsorption saturation, the column was eluted with ethanol/water solutions. The test by HPLC also showed that the sucralose-6-ester and the impurities can be separated by gradient elution of the macroporous resin with different concentrations of ethanol solution. Usually, ethanol of higher concentration would increase the desorption of the adsorbate, but decrease the selectivity of the target compound in the eluent, leading to a high impurits in sucralose-6-ester product. So gradient elution with different concentrations of ethanol was adoped to separate sucralose-6-ester from the D101 resin. The gradient elution tests were taken as follows: the column was firstly washed by distilled water and 3% ethanol-water solution to remove the relatively polar impurities, and then eluted by 10% ethanol-water solution. The eluted fractions were collected and detected by HPLC every 0.1 BV.

RESULTS AND DISCUSSIONS

Adsorption and desorption properties

Three macroporous resins with different physical properties were employed for the separation of the sucralose-6-ester from the residue in the production of sucralose. The preliminary choice of resin was determined on the basis of the adsorption capacity, desorption ratio and selectivity.

Table 1. Adsorption/desorption properties of the resins for the sucralose- 6-ester

Resin	Adsorption capacity (mg/g resin)	Desorption ratio (%)	Selectivity
D101	45.5	87.0	1.22
DM301	54.5	72.9	1.08
S-8	50.2	79.6	1.05

As shown in Table 1, the adsorption capacity of sucralose-6-ester on DM301 resin was the highest, however, considering the desorption ratio and the selectivity of sucralose-6-ester, resin D101 was selected for further study.

Adsorption isotherms

The Langmuir and Freundlich theoretical equations were used to describe the interaction between sorbent and adsorbed material.[14] Adsorption isotherm experiments on selected resin (1 g of dry resin) were conducted by bringing into contact five aliquots of 20 mL sample solutions at different concentrations (17.8, 14.3, 11.9, 8.92, and 5.95 mg/mL) in a constant temperature shaker, and then shaking at 20, 30 and 40! for 12 h respectively. The concentrations were determined by HPLC. These equations may be expressed as:

Langmuir equation:

$$q_e = \frac{q_m \times C_e}{K_L + C_e} \quad ...(5)$$

Freundlich equation:

$$q_e = K_F C_e^{1/n} \quad ...(6)$$

where q_e and C_e denote the adsorption capacity (sucralose-6-ester/resin, mg/g) at adsorption equilibrium and the equilibrium concentration of sucralose-6-ester(mg/mL) respectively, q_m denotes the maximum adsorption capacity (sucralose-6-ester/resin, mg/g), K_L, K_F and 1/n are characteristic constant as relative indicators of adsorption capacity.

The experimental adsorption data are shown as Figure 2, and the estimated parameters are summarized in Table 2. The correlation coefficients of the Langmuir and Freundlich models were both high. The Langmuir isotherm model is based on the assumption that the sorbate form is only a single layer,[15] thus results in Table 2 showed that the adsorption process was mainly a monomolecular layer adsorption. At the same initial concentration, the maximum adsorption capacities changed slightly in the investigated temperature range. Therefore, room temperature was taken in the following experiments. In the Freundlich, the adsorption can easily take place when the 1/n is between 0.1 and 0.5, and not easy to happen if 1/n value is between 0.5 and 1.[16] And values in table 2 indicated that the adsorption process of sucralose-6-ester onto D101 could take place easily.

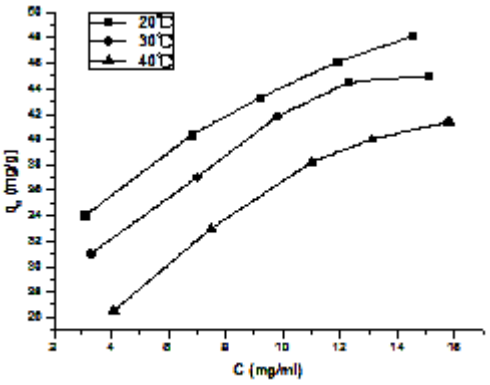

Fig. 2. Adsorption isotherm for sucralose-6-ester on D101 at different temperature

Dynamic adsorption

During the loading process of the sample solution, the effect of the initial concentration and the flow rate was studied to ascertain the optimum dynamic adsorption conditions on D101 resin. Figure 3 shows the effect of the initial concentration of sucralose-6-ester and the flow rate on the corresponding breakthrough curves for fixed-bed adsorption. As shown in Figure 3(a), the adsorption

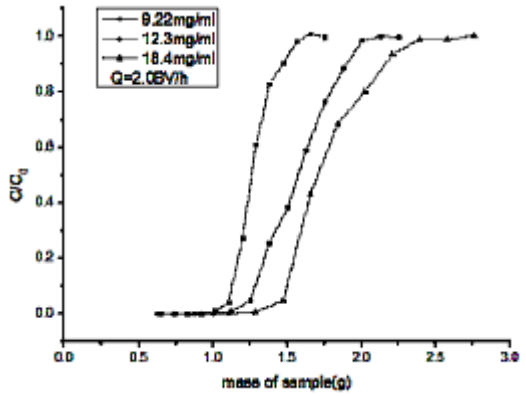

Fig. 3. (a) Effect of (a) concentration and (b) flow rate on the breakthrough curves in resin D101

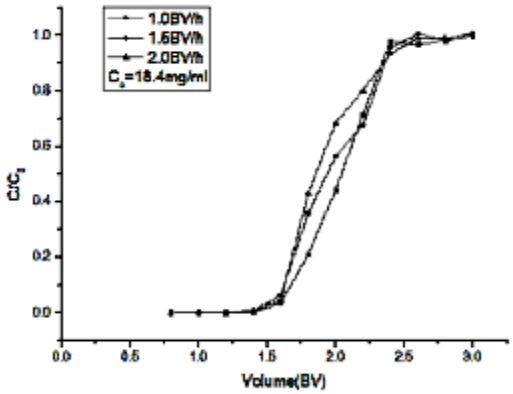

Fig. 3(b). Effect of (a) concentration and (b) flow rate on the breakthrough curves in resin D101

Table 2. Langmuir and Freundlich parameters of sucralose-6-ester on D101

Langmiur model	20°C	30°C	40°C
equation	$q_e = \frac{55.2C_e}{2.299 + C_e}$	$q_e = \frac{54.6C_e}{2.985 + C_e}$	$q_e = \frac{53.6C_e}{4.391 + C_e}$
R^2	0.9890	0.9856	0.9838
q_m(mg/g)	55.2	54.6	53.6
K_L	2.299	2.985	4.391
Freundlich model			
equation	$q_e=25.04C_e^{1/4.04}$	$q_e=21.60C_e^{1/3.56}$	$q_e=16.42C_e^{1/2.89}$
R^2	0.9978	0.9849	0.9910
K_F(mg/g)	25.04	21.60	16.42
1/n	0.2475	0.2809	0.3460

capacity increased along with the increase of the initial concentration, and appropriately high concentration is more conducive to the dynamic adsorption. Figure 3(b) indicated that varying the flow rate from 1.0 to 2.0 BV/h makes almost no difference to the adsorption performance. Therefore, 18.4 mg/ml and 2.0 BV/h was chosen for further experiments while the adsorption capacity is 29.4 mg(sucralose-6-ester)/ml(resin).

Elution process

The elution process takes gradient elution: 0.5 BV of distilled water, 0.5 BV of 3% ethanol, and then 1.0 BV of 10% ethanol at a flow rate of 1.0 BV/h. The distilled water and 3% ethanol/water solution was used to remove the relatively large polar impurities like the monochlorosucrose-6-ester, dichlorosucrose-6-ester etc., and then 10% ethanol/water solution was used to elute sucralose-6-ester.

Figure 4 shows the elution curve of sucralose-6-ester from the D101 resin in the separation process. The results revealed that the strong polar impurities were removed with 0.5 BV of distilled water and 0.5 BV of 3% ethanol, followed by 1.0 BV of 10% ethanol to elute sucralose-6-ester. The fractions were collected and merged. The purity of sucralose-6-ester was measured to be 41.8%, and the recovery of the yield was 56.3% in the first separation cycle. To further purify sucralose-6-ester, second separation cycle was taken, the results showed the purity was 64.7% and the recovery was 58.2%.

Effect of Separation

The effect of D101 resin on separating sucralose-6-ester was examined by HPLC. Fig. 5 showed the effect of separation before and after treatment by D101 resin. It can be seen that some impurities have been removed by D101 resin, and the purity of sucralose-6-ester was significantly increased.

Re-usability of the Resin D101

The reusability of the resin plays an important role in the separation process. The column was subjected to a cleaning procedure after each separation cycle to the next run. This cleaning procedure consisted of the following procedures: 1 BV of 30% ethanol at the rate of 1 BV/h, then 1 BV 60% and 1 BV90% ethanol at the rate of 2 BV/h to wash the column, followed by washing with a large amount of distilled water.

Table 3 illustrates the breakthrough capacities on the resin D101 after repeated use. It showed after 5 cycles of use, the adsorption capacities remained the same.

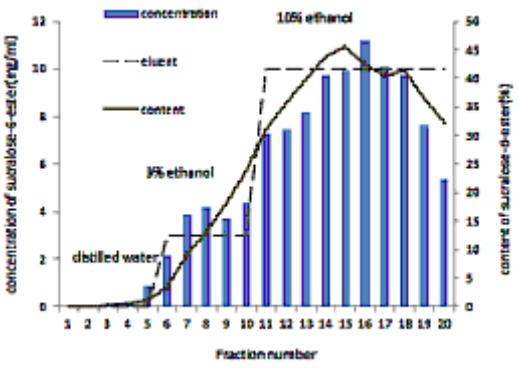

Fig. 4. The elution curve of sucralose-6-ester from the D101 resin column

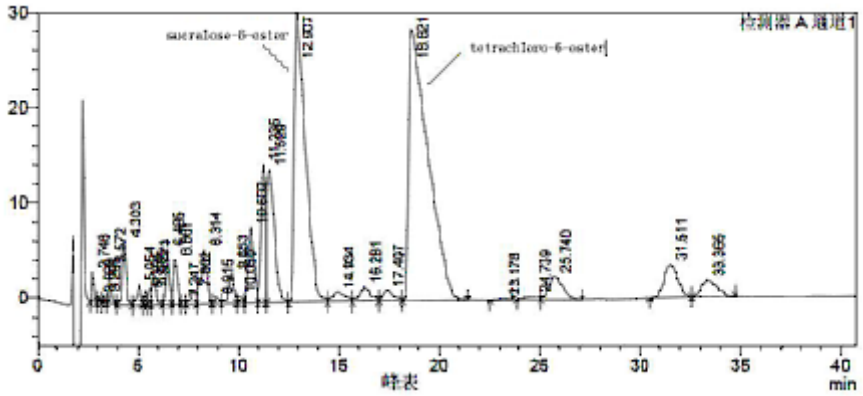

Fig. 5(a). HPLC of sample solution before(a) and after (b) treatment with D101 resin

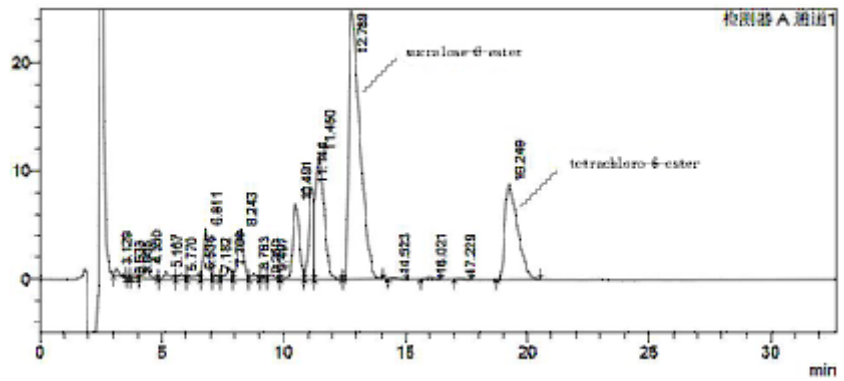

Fig. 5(b). HPLC of sample solution before(a) and after (b) treatment with D101 resin

Table 3. The adsorption capacities on resin D101 after repeated uses

Cycle	Adsorption capacity of sucralose-6-ester(mg/ml resin)
1st cycle	29.4
2nd cycle	28.6
3rd cycle	30.0
4th cycle	29.3
5th cycle	29.0

CONCLUSIONS

The separation process of sucralose-6-ester with macroporous resin has been successfully developed in this study. The D101 resin offered the best separation performance because of its highest desorption ratio and selectivity. The adsorption isomer data fit well both to the Langmuir model and Freundlich model. The optimum parameters were obtained to ensure the high efficiency of separation. The purity of sucralose-6-ester was increased from 21.3% to 41.8% at the recovery of 56.3% in the first separation, and to 64.7% at the recovery of 58.2% in the second separation. The D101 resin column can be used for at least 5 times without reducing its adsorption property.

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