

Research methods used to control the plasticization and retrogradation process of starch

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The paper presents the possibility of using FTIR spectroscopy, X-ray diffraction (XRD), gel permeation chromatography (GPC) and differential scanning calorimetry (DSC) to control the starch plasticization process. The changes that occur in the starch structure after its plasticization and retrogradation process after a certain time are discussed.

Keywords – bioplastic, thermoplastic starch, plasticization, retrogradation, FTIR, XRD, GPC, DSC

Introduction

Biodegradable plastics can be obtained from cheap, widely available renewable plant raw materials, which makes them "double green". This means that, apart from the advantage of being biodegradable, they are obtained from raw materials get from renewable raw materials. A polymer obtained from renewable sources, which can be successfully used in the biodegradable plastics industry, is starch. This is confirmed by market data, according to which starch-based blends accounted for approximately 18% of the biopolymer market in the European Union in 2022 [1, 2].

Starch is a natural polymer composed of linear amylose chains and branched amylopectin. In the plastics industry, starch is used in its native form as a filler for other polymers or it must be processed into a thermoplastic form. This is due to the thermal properties of native starch, which prevent its processing, because its melting point (220 - 240°C) is higher than the degradation temperature (about 220°C). This means that starch in its pure form cannot be processed and it is necessary to obtain the so-called thermoplastic starch (TPS). For this purpose, starch must undergo a plasticization process, i.e. destroy the crystalline structure of starch grains by applying shear forces and appropriate temperature conditions in the presence of plasticizers. This is effected in the extrusion process which leads to obtain an amorphous, thermoplastic material - thermoplastic starch which can be further processed with classic techniques used in plastics processing [3, 4].

One of the starch disadvantage is the susceptibility to the retrogradation process, which consists in the fact that the starch returns to its original, crystalline structure and the effect is "wetting" of plasticizers. This phenomenon limits numerous applications of starch materials. Therefore, it is necessary to select an appropriate plasticizing composition that will both plasticize

the starch effectively and inhibit the progress of the thermoplastic starch retrogradation process [5]. If appropriate analytical methods are applied, the plasticization and the possible progress of the starch retrogradation process can be controlled. The paper presents the results of analyzes using infrared spectroscopy (FTIR), X-ray diffraction (XRD), gel permeation chromatography (GPC) and differential scanning calorimetry (DSC). Changes that occur in the structure of native starch in the plasticization process with glycerol, as well as structural changes in thermoplastic starch as a result of conditioning TPS granules at 25°C and RH=50%, are discussed.

Experimental part

Preparation of thermoplastic starch

The native potato starch was mixed with glycerol as a plasticizer using a high-speed mixer. Then, homogeneous mixture was extruded using a co-rotating twin-screw extruder from LABTECH Engineering with L:D=48:1 parameters and screw diameter of 20 mm, in temperature range of 70 - 140°C and screw speeds in the range of 90 - 120 rpm. The threads of the material were air cooled and granulated using a side-cutting granulator, and thermoplastic starch granule about 3 mm long were obtained [6].

FTIR Spectroscopy

Infrared spectra were measured using a Nicolet iS10 spectrometer produced by Thermo Scientific. The spectra were recorded using the appropriate ATR technique (Attenuated Total Reflection) at room temperature with 4 cm⁻¹ resolution in the mid-infrared spectral range of 650 - 4000 cm⁻¹.

The process of starch plasticization leads to changes in its structure. As a result of the plasticizer effect, shear forces and temperature during the extrusion process, the intramolecular and intermolecular hydrogen bonds in the starch chains between the hydroxyl groups are broken. In place of these bonds, hydrogen bonds between the -OH groups of the starch and the plasticizer are formed. Table 1 lists the wavenumber values for the characteristic bands of native and thermoplastic starch.

Table 1. Characteristic bands in the FTIR spectrum of native and thermoplastic starch.

| Composition | Hydrogen bonds | C-O in C-O-H starch | C-O in C-O-C starch |
|----------------------|-----------------------|--|----------------------|
| Native starch | 3303 cm ⁻¹ | 1151 cm ⁻¹ 1083 cm ⁻¹ | 996 cm ⁻¹ |
| Thermoplastic starch | 3287 cm ⁻¹ | 1150 cm ⁻¹ 1079 cm ⁻¹ | 995 cm ⁻¹ |

In the FTIR spectra of TPS granules, a shift in the characteristic bands compared to native starch is observed. The hydrogen bond band shifted towards lower wavenumbers from 3303 cm⁻¹ to 3287 cm⁻¹. According to the harmonic oscillator model, which is used in FTIR spectroscopy, the wavelength of the band for a particular bond, decreases with the increase of the molecular interaction force. The reduction in the wavenumber value suggests that the newly formed hydrogen bonds between the plasticizer and starch are stronger than the intramolecular and intermolecular hydrogen bonds between the hydroxyl groups of starch [7].

Characteristic changes in the thermoplastic starch spectrum in comparison with native starch also occur in the range of 900 - 1200 cm⁻¹. In this range, the FTIR spectrum of native and

thermoplastic starch shows bands associated with C-O stretching vibrations in the C-O-H system and C-O in the C-O-C system in starch chains. Both the C-O-H and C-O-C groups present in the starch structure can form hydrogen bonds with the plasticizer [8]. These bands are also shifted towards lower wavenumbers which confirms the formation of a more stable thermoplastic starch structure.

Gel Permeation Chromatography (GPC)

Molecular weight measurement of the samples was performed using Malvern gel chromatograph equipped with a refractometer (RI), the light scattering RALS (90°) and LALS (7°) and a viscometer (VS) detectors combined. This four detectors were calibrated with pullulan standard (118 kDa). The separation was performed using three aqueous SEC chromatography columns (Malvern: A2000, A5000, A6000). The detectors and columns were thermostated at 35°C. The mobile phase was 0.1M NaNO₃ and flow rate during measurement was 0.5 ml/min. For the analysis, the weighed sample of starch was dissolved in DMSO/H₂O (90:10). Concentration of the sample was 2.5 mg/ml. The sample in DMSO/H₂O solution was placed in the heating-mixing module at 100°C and stirred at a constant speed for 120 min. After cooling and before the analysis, the dissolved sample was filtered with 0.22 µm filters. The injection volume of the sample was 100 µl. The refractive index of the solvent was 1.3340 and the dn/dc value of native starch was 0.16. For thermoplastic starch, dn/dc was determined using an RI detector and it is 0.10. Calculations of molecular weight, intrinsic viscosity and Mark-Houwink coefficient were done using the OmniSEC 5.0 software provided by Malvern [9].

The results of molecular weight measurements and parameters determining the structure of polymers obtained by gel permeation chromatography are summarized in Table 2.

Table 2. Results of GPC analyzes of native and thermoplastic starch

| Composition | M _w [MDa] | IV [dL/g] | [a] Mark-Houwink constants |
|----------------------|-------------------------|--------------|-------------------------------|
| Native starch | 33,63 | 1,990 | 0,190 |
| Thermoplastic starch | 3,52 | 0,282 | 0,102 |

The average molecular weight of native starch is approx. 33 MDa. The applied shear forces during extrusion and the temperature as well as the plasticizer effect in the plasticization process result in a significant reduction of the molecular weight of the obtained thermoplastic starch (3.52 MDa) as compared to native starch. Thermal degradation and mechanical degradation of polymer chains caused by shear forces affect the appearance of the shorter chains which leads to a lower molecular weight of starch. If the molecular weight of the obtained thermoplastic starch is lower, efficiency of the plasticization process is the greater [9, 10].

The intrinsic viscosity depends on general shape of molecules (rod > coil > sphere), chain length (according to the Mark-Houwink equation) and branching of polymer chains (linear > branched). The intrinsic viscosity (IV) of native starch is 1.990 dL/g, and after the plasticization process this value is significantly reduced to about 0.3 dL/g. This result confirms the degradation and shortening of native starch polymer chains.

The Mark-Houwink constants of native and thermoplastic starch are characterized by a value of less than 0.5. This means that starch molecules form compact and spherical structures. If the value of the constant is smaller, the structure of the polymer is more branched and molecules create more compact and spherical forms [11, 12].

X-ray diffraction (XRD)

Diffractograms were recorded using a PANalytical Empyrean diffractometer equipped with PIXcel3D detector and nickel filtered Cu-K α radiation. The scattering angles (2θ) ranges were from 5 - 40° at 0.01° step size. Moreover, thermoplastic starch samples were milled after having been frozen in liquid nitrogen and placed in a suitable holder in powder form.

Native starch has a partially crystalline structure: the crystalline phase constitutes approx. 15 - 45% of the total structure depending on its botanical source with the rest being the amorphous phase [10, 11]. The crystalline structure of native starch is destroyed during plasticization in the extrusion process, and in its place a semicrystalline structure is formed resulting from the recrystallization of amylose. The crystalline phase is a secondary crystal structure V_H or E_H which is formed after cooling of the granulate. If the conditions of the extrusion process do not ensure a complete disruption to the structure of the native starch, then the residual crystalline lattices present in the native starch also exist in the TPS granulate.

Figure 1 shows the diffractograms of native starch and starch plasticized with glycerol. Table 3 shows the results of XRD analyzes of native and thermoplastic starch immediately after extrusion and after conditioning at 25°C and RH=50%. Analysis of the crystal structure type was performed based on literature data [10], and the crystallinity of native and thermoplastic starch granulates was determined according to the Nara and Komiya methods [13].

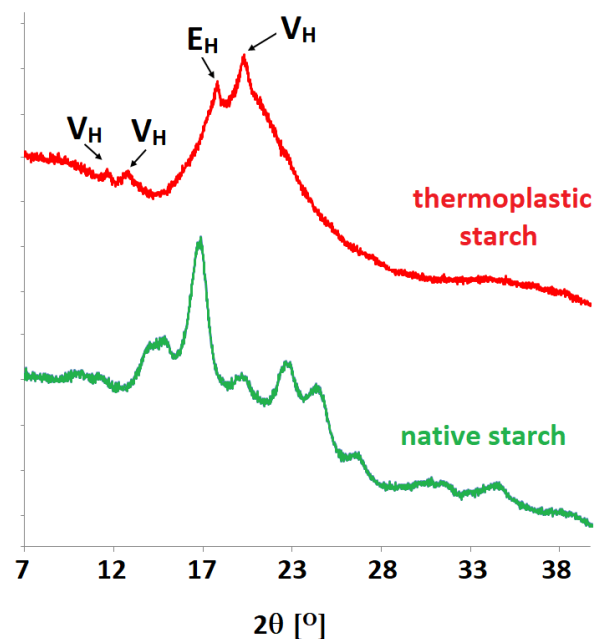


Fig.1. Diffractograms of native starch and starch plasticized with glycerol.

The XRD analysis of native potato starch confirmed the presence of the B-type crystal structure, whereas the crystalline phase content is 45.5%. As a result of starch plasticization with glycerol, crystalline regions of starch granules undergo destructure and therefore, there are no peaks characteristic of B-type structure on diffraction pattern of the thermoplastic starch. However, there are diffraction peaks associated with the formation of secondary crystal structures of the E_H- and V_H-type. Destruction of the crystalline phase of native starch in the plasticization process resulted in a decrease in the content of the crystalline phase in the obtained thermoplastic starch to 34.7%.

Table 3. Analysis of the types of native and thermoplastic starch structure

| | After extrusion | | | After 7 days (25°C, RH=50%) | | After 30 days (25°C, RH=50%) | |
|---------------|---|--|----------------------|--------------------------------|----------------------|---------------------------------|----------------------|
| | Diffraction peaks [°2θ] | Starch structure | Starch crystallinity | Starch structure | Starch crystallinity | Starch structure | Starch crystallinity |
| Native starch | 5.7; 15.2(s); 17.2(vs); 19.8(s); 22.2(s); 24.1; 26.3; 31.0; 34.4 | B-type | 45.5% | - | - | - | - |
| TPS | 7.9(br); 13.1(w); 19.8(vs) 18.2(vs) | V _H -type E _H -type | 34.7% | V _H -type B-type | 25.7% | V _H -type B-type | 33.0% |

Analysis of the diffraction pattern of the TPS granules containing 30% glycerol after 7 days of conditioning at RH=50% and at 25°C confirms the presence of the V_H-type crystal structure. The non-stable E_H-type structure present in the granulate directly after extrusion is transformed into a durable and more stable V_H-type structure. The total crystallinity of the sample decreases to 25.7%, which may indicate that the absorbed moisture causes further plasticization of the material and its amorphization. Simultaneously, peaks originating from the B-type structure appear in the diffractogram, which may indicate the beginning of the retrogradation process. Some of the amorphous regions of the TPS granules return to the original B-type structure under these temperature and humidity conditions which is accompanied by the migration of glycerol to the surface of the material. As a result of further storage of this granulate in the increased humidity and temperature conditions, the content of B-type structure increases, and thus, the total crystallinity of the sample increases to 33.0%. This confirms the further progress of the starch retrogradation process [8, 14].

Differential Scanning Calorimetry (DSC)

DSC measurements were performed on a TA Instruments Q20 apparatus with a cooling system down to -90 °C in a nitrogen atmosphere. The calibration was performed using metallic indium as a standard. Test samples weighing about 10 mg were placed in a hermetic aluminum pan and then in a measuring cell. The sample was tested according to the following temperature program:

- cooling down to -90°C,
- heating to 180°C at a rate of 10°C/min,
- re-cooling to -90°C,
- heating to 180°C at a rate of 10°C/min.

The obtained results are presented as heat flow per unit of time depending on the temperature.

The plasticization process of starch also leads to changes in the thermal properties of the material. Thermal analysis of the thermoplastic polymer is mainly based on a study by DSC. The most important parameter is the glass transition temperature T_g which is a characteristic feature for each polymer. Below this temperature, thermoplastic materials are hard and brittle. Above the glass transition temperature, polymers have the properties of viscoelastic materials. The analysis of the DSC polymer curve should be performed based on the second heating cycle of the sample, due to the fact that each polymer sample has its so-called "thermal history. Figure 2 shows the DSC curve of starch plasticized with glycerol.

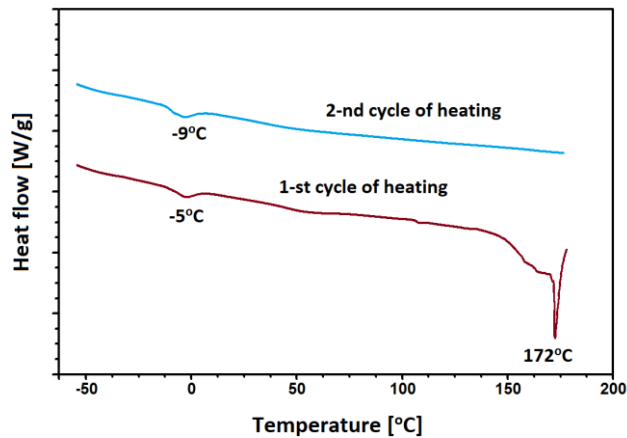


Fig.2. DSC curve of thermoplastic starch.

In the first heating cycle, a peak appears on the curve of thermoplastic starch at 172°C, associated with the melting of crystal structures that were formed during cooling and storage of the granulate. In addition, at -5°C, an endothermic transformation occurs, which is related to phenomenon of thermoplastic starch glass transition. In the second heating cycle, one endothermic transformation related to the glass transition temperature of TPS starch is already observed. The glass transition temperature in the DSC thermographs is related to the thermal transformation of the amorphous phase of polymer. This confirms that a completely amorphous starch structure was obtained in the plasticization process [3, 15].

Conclusions

Natural polymers such as the native starch and the thermoplastic starch are materials difficult to analyze. Results of tests are often ambiguous and difficult to interpret. Therefore, an important aspect in the quality control of granules based on the thermoplastic starch is selecting an appropriate analytical method and optimize measurement conditions.

The conducted tests confirm that glycerol is an effective starch plasticizer, and the effectiveness of the plasticization process can be successfully assessed using methods such as: FTIR, GPC, XRD and DSC. Additionally, the possible progress of the retrogradation process can be confirmed by the XRD method.

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