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Dedicated to Professor Liviu Literat
On the occasion of his 85th birthday

STRUCTURAL FT-IR AND ^{13}C CP/MAS NMR INVESTIGATION OF NATIVE STARCH WITH PLASTICIZERS BEFORE AND POST EXTRUSION PROCESS

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ABSTRACT. Structural aspects of native corn starch samples with plasticizers are investigated by FT-IR, ^{13}C CP/MAS NMR and XRD methods before and after extrusion process. The crystalline A, B and V – types structural phases prevail before extrusion in the prepared samples. The amorphous phase becomes dominant in both recent prepared formula and control specimen sample after extrusion.

Keywords: *starch, extrusion, FT-IR, CP/MAS NMR, XRD*

INTRODUCTION

Starch in one most studied biopolymers and many structural aspects involving the arrangement of the two component molecules, amylose and amylopectin, inside native starch granules and starchy materials are strongly investigated in the last years [1-4]. The both component molecules consist of α(1-4) linked D-glucose units, amylose being essentially linear, whereas amylopectin is a highly branched polymer due to 5-6% of α(1-6) links [3].

The native starch has a granular structure with an internal architecture characterized by concentric rings representing semi-crystalline shell (thickness 120-400nm) separated by essentially amorphous regions. There is much

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evidence that the semi-crystalline shells consist of regular alternating amorphous and crystalline lamellae repeating at 9-10 nm. In this structural organization, parallel double helices of amylopectin side chains are assembled into radially oriented clusters [4].

The amylose to amylopectin ratio influences strongly the physical-chemical characteristics an properties of starch, as viscosity, gelation power, adhesion etc [5].

The vibrational spectroscopic techniques, such as IR spectroscopy and Raman spectroscopy are successfully used in the quantitative investigation of control analysis of starch for the food industry, characterization of some medical polysaccharides and quantitative investigation of amylose and amylopectin content in different starch samples [6-11].

The vibrational spectroscopies were also used to investigate the retrogradation and gelation processes of some types of starches [6-8] and for characterization of some medical polysaccharides, including amylose and amylopectin [12].

Beside the vibrational methods the nuclear magnetic resonance (NMR) spectroscopy, especially nuclear magnetic relaxation method is a powerful method for studying the morphology, compatibility, chain conformation and dynamics of polymer systems [13-16].

NMR is sensitive to short-range order and according to the theory of NMR signals developed by Kulagina et al. [13] is possible by the analysis of NMR line area to determine the fractions of crystalline and amorphous phases in solid biodegradable polymers.

According with this method, the cross-polarization magic-angle spinning NMR (CP/MAS NMR) technique is also successfully used to crystallinity degree evalution by inspecting the width of the resonance lines [7, 17-19].

Thermoplastic extrusion is a thermo-mechanical processing used to disrupt and transform the semi-crystalline structure of starch granules to form a homogeneous and amorphous material. This transformation is usually accomplished by small amounts of molecular substances commonly known as plasticizers. The most used plasticizes are water, a volatile plasticizer and glycerol, a non-volatile plasticizer [20, 21].

The plasticizing – antiplasticizing effects of water and glycerol contents on starch samples and also the amylose / amylopectin ratious were recent investigated by various methods [4, 9, 16, 20].

The following samples, blend formula (68% native starch, 17% glycerol, 15% water) before extrusion (a), extruded (previous blend (a) formula) sample (b) and a control extruded specimen (c) were investigated by FT-IR and ^{13}C CP/MAS NMR spectroscopy in order to establish the prevalence of crystalline or amorphous domains after the extruding process.

RESULTS AND DISCUSSION

Fig.1. shows the IR spectra of the blend formula (a) and extruded starch samples (b, c). The two bands from 3300 cm^{-1} and 1650 cm^{-1} are ascribed to water stretching and bending vibrations and also to different OH groups present in starch and glycerol molecules [7, 8]. The other absorbtions are originated from the vibrational modes of the amylose and amylopectin, the principal components of starch [7].

Thus the two absorption bands from at 2930 cm^{-1} and 2860 cm^{-1} are assigned to vibrations of CH_2 groups [7]. The absorbtions from 1338 cm^{-1} region are due to bending modes of O-C-H, C-C-H and C-O-H angles from amylose and amylopectin formations [22].

The strong absorbtion peaks appeared in the 1150 – 900 cm^{-1} region are assigned to C-C and C-O stretching vibrations [23]. The bands from 1000 cm^{-1} region are sensitive to changes in crystallinity and the intensity of 1000 cm^{-1} band determines the orientation in intermolecular H – bonding of CH and CH_2 in CH_2OH [7, 24].

The band from 1106 cm^{-1} of the sample (a) is characteristic for glycerol and assigned to C-O stretching vibrations [25, 26]. As it is suggested by NMR spectrum of this sample (^{13}C line from 64 ppm) a complex between native starch and glycerol stabilized by hydrogen bonds appears. The disappearance of this band after extrusion may be due to the homogenization of the structural network.

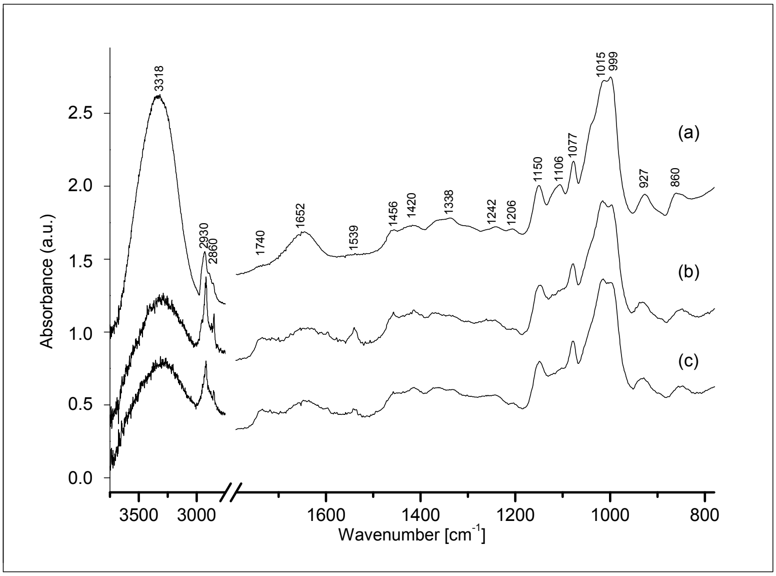

Figure 1. FT-IR spectra of blend formula before (a) and after extrusion (b) and of a control extruded specimen sample (c).

Many correlations between changes in the some band intensities from the 1000 cm^{-1} region (994, 1000, 1022, 1040, 1047, 1053 cm^{-1}) and the crystalline and amorphous phases (forms) of different starches are given in papers [4, 7, 9].

According to Capron et al. [4] the intense absorption from 998 cm^{-1} may be assigned to hydrated crystalline domains whereas the band at 1015 cm^{-1} reveals the amorphous contribution of plasticizers in starch formulas.

By analyzing the IR spectra (Figs. 1a, 2a) of blend formula before extrusion results that 999 cm^{-1} band correlated with crystalline domains is more intense than the 1015 cm^{-1} band characteristic of amorphous starch samples.

In the same time it can be observed (Figs. 1, 2b, c) that for extruded samples, the 1015 cm^{-1} band is more intense than 999 cm^{-1} band, thus resulting that in extruded starch samples (obtained by us (b) and control specimen (c)) the amorphous domains prevail.

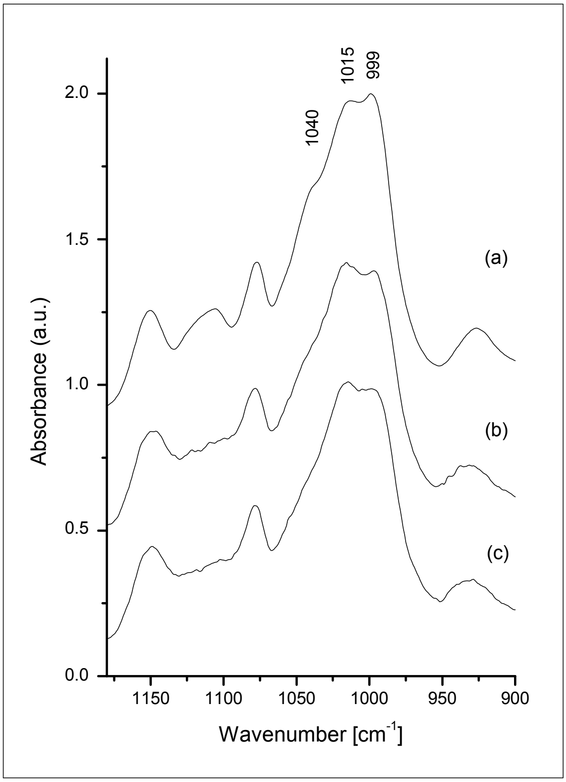

Figure 2. Extended 1000 cm^{-1} regions of the IR spectra shown in Figure 1.

The appearance of the weak band at 1539 cm^{-1} in Fig. 1b, c may be assigned to C-O vibrations from conjugated carbonyl and carboxyl groups after extruding or cooking processes (associated to the retrogradation in maize tortillas) of starch samples [7]. This fact may be ascribed to the partial amylose separation of amylopectin after the thermal treatment (gelatinization and retrogradation) of starch and also to the stress during extrusion process [2].

^{13}C CP/MAS NMR spectra of the investigated samples are shown in Fig. 3. The structural crystalline behavior of a starch granule is due to amylopectin, but also to the amylose/amylopectin ratio. According to Thérien-Aubin and Zhu [19], there are three types of crystalline starch structures: A, B, which are both formed by double helices of amylose and amylopectin, and V-type, composed of single helices.

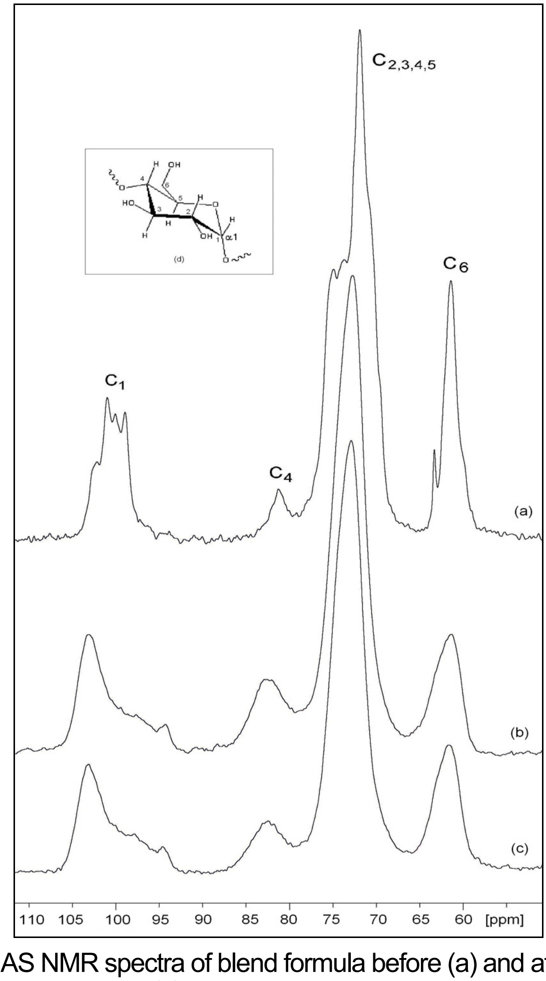

Figure 3. ^{13}C CP/MAS NMR spectra of blend formula before (a) and after extrusion (b) and of a control extruded specimen (c) recorded at room temperature and 14 kHz spinning speed. The insert (d) represents the chemical structure of α(1-4) glucopyranose.

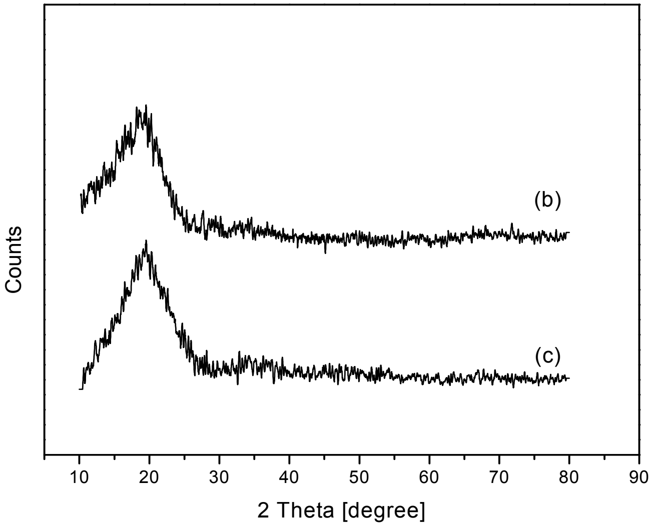

Figure 4. X-Ray Diffractograms of extruded samples (b, c).

Solid-state NMR spectroscopy, in particular ^{13}C CP/MAS NMR, is accurately used in distinguishing these three structural motifs (phases) [7,17-19]. Also, the degree of crystallinity can be easily probed by inspecting the shape and the width of the NMR resonance lines.

The most pronounced crystalline behavior is put in evidence in the spectrum of sample (a), blend formula (65% native starch, 17% glycerol, 15% water) before extrusion (Fig 3a). Thus, the ^{13}C CP/MAS spectrum of this sample shows the major presence of A, B and V- types of crystalline structures. In the region of the characteristic C_1 NMR resonance (glucose units [19]), an overlap of the triplet centered around 101 ppm specific to the A type – crystalline former with a doublet at 101 ppm characteristic of B type crystalline structure and a singlet at 104 ppm for the V- type structure appears.

The strong $C_{2,3,4,5}$ quadruplet signal from 73 ppm is also characteristic of A – type phase. We mention also that in the intense C_6 absorbtion region (62 ppm) the contributions of A, B, and V – types crystalline phases manifest, too.

The sharp ^{13}C line from 64 ppm belongs to glycerol, and suggest the formation of a complex between native starch and glycerol, most probably stabilized by hydrogen bonds.

The presence of a small amorphous phase in this sample (a) is suggested by the weak and broad NMR resonances at 104 ppm (C_1), 83 ppm (C_4) and 61 ppm (C_6).

In contrast with these results, the (b) and (c) NMR spectra of the extruded samples show the domination of the amorphous phase character. Both spectra are similar with ^{13}C CP/MAS NMR spectra reported by other authors for amorphous starch samples [18, 19].

The prevalence of amorphous domains after the extrusion suggested by IR and ^{13}C CP/MAS NMR data is also confirmed by XRD patterns (Fig.4) of extruded samples (b, c).

The absence of sharp peaks in the diffraction pattern of the extruded samples shows only the amorphous character without any crystalline form [27].

EXPERIMENTAL SECTION

The normal corn starch used in this study was obtained from SC Amylon Sibiu, Romania, having an water content on wet basis (wt.b) of 10.76 %, a density of 0.561 g/cm^3 and an amylose content of 21%. The glycerol used in formulas was purchased from SC Nordic Invest SRL Cluj Napoca and had a concentration of 99.5% and a density of 1.262 g/cm^3. The water used was from the water supply system.

FT-IR/ATR spectra were recorded at room temperature on a conventional Equinox 55 (Bruker, Germany) spectrometer equipped with a DTGC detector, coupled with an ATR sampling device (Miracle, Pike Techn.). The resolution was of 2 cm^{-1}.

^{13}C solid state cross-polarization magic-angle-spinning (CP/MAS) NMR spectra were recorded on a Bruker Avance III wide-bore spectrometer operating at 500 MHz Larmor frequency for ^1H, and 125 MHz for ^{13}C, respectively, using a 4 mm double resonance probehead. Standard ^{13}C CP/MAS experiments were performed at room temperature, using a spinning frequency of 14 kHz and a ^1H 90° pulse of 2.3 μs. The spectra were acquired under two-pulse phase-modulated (TPPM) 1H decoupling at 100 kHz by averaging 4000 scans, with a recycle delay of 5 s, and a 2 ms contact pulse. ^{13}C CP/MAS spectra are calibrated according to the $^{13}CH_3$ line in TMS through an indirect procedure which uses the carboxyl NMR line in α-glycine.

A laboratory twin-screw extruder with co-rotating intermeshing, self-wiping screws ZK 25, Collin, Germany) was used to conduct the extrusions. The extruder has 25 mm screw diameter, 30:1 length to diameter ratio, max. 400 rpm screw speed and six independent electric heating and cooling areas.

The starch was fed into the extruder hopper with a twin-screw volumetric feeder, DSV 020D, Definitive Inovation, Italiy.

The dosing pump used to feed the plasticizers from the plasticizers tank was a peristaltic pump SP 311/12, VELP, Italiy.

The plasticizers were added into the working area through a pipe connection located at 170 mm from axis of the supply hopper.

The screw speed was set at 150 rpm and the barrel temperatures were maintained during the experiment at 30, 50, 80, 100 and 120 ^{0}C, respectively, from the feeding port to the die section.

A circular die plate with one hole was used. The diameter of the hole in the die is 3 mm.

The extruding product was collected and cooled to room temperature. Each experimental samples were taken after the extruder had reached steady state.

X – ray diffractograms were obtained with a Bruker D8 Advanced X – ray diffractometer with a graphite monochromator for Cu - K_α radiation with λ = 1.54 Å.

CONCLUSION

Both amorphous and crystalline structural phases are present in various proportions in the investigated starch samples.

The crystalline structural phases (A, B, V – types) prevail in the blend formula (68% native starch, 17% glycerol, 15% water) before extrusion.

After extrusion process the amorphous phase becomes dominant in both, recent prepared formula and control specimen sample.

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