Study of succinic anhydride amidation by 2-aminothiazol

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Abstract. The reaction between succinic anhydride and 2-aminothiazole was carried out in polar organic solvents (acetone, 1,4-dioxane, isopropanol) and in a mechanochemical reactor. By of X-ray phase analysis, IR, ¹H, ¹³C NMR spectroscopy it was found that in both techniques of carrying out the reaction, the only product was succinamide. It was shown that the process efficiency in the mechanochemical reaction conditions is higher in comparison with the traditional solution’s synthesis. The solid-phase interaction development was proceeded during 45 days after load removed, it was especially intense in the first 10 days.

1. Introduction

The structural fragment of 2-aminothiazole (AT) is present in a number of biologically active compounds with antibacterial action (for example, third generation cephalosporins), therefore, AT can be considered as a model compound in development of methods for obtaining new antimicrobial drugs. In particular, promising drug can be AT, substituted at the amino group by the residue of succinic acid, which is widely involved in the body metabolic processes [1]. So, succinamide, formed during the reaction of AT with succinic anhydride (SAn) in acetone, according to [2], exhibits pronounced antihypoxic activity. Wherein, the authors of [2] do not provide methodological conditions for obtaining the product, data on its yield and spectral characteristics. Succinic anhydride, in turn, is a low-molecular-weight analogue of physiologically active maleic anhydride copolymers [3], therefore, the study of the reaction of AT with SAn is of interest as a model one for the obtaining of conjugates for the antimicrobial drugs with modified release.

This investigation of the interaction between AT and SAn aimed to detect the structure of the amidation product and to find the effect of the reaction conditions on the product yield.

2. Experimental

AT was purchased from Acros Organics (97 %), SAn was of analytic reagent grade and used without further purification. The purity of the starting materials was confirmed by ¹H NMR and IR spectroscopy. Solvents (1,4-dioxane (DO), isopropyl alcohol (IPA), acetone) were purified by distillation, water was twice distillated (bidistillate).

The reaction of SAn with AT was carried out in solution and in a mechanochemical reactor at reagents ratio of 1:1 mol/mol. In the first case, 0.001 mol each of SAn and AT (0.1007 and 0.1001 g, respectively) were dissolved in 3 ml of solvent (acetone, DO, IPA or water) in a 30-ml round-bottom flask; the solution was kept at a given temperature. The reaction temperature of 0 and 10 °C was kept constant using an ice-water mixture, from 20 to 60 °C – a water bath. In the latter case, the flask was
equipped with a reflux condenser. After a certain period of time the reaction mixture was cooled to room temperature, unreacted SAn and AT were filtered off, the solvent was distilled off at a vacuum of ~ 50 mm Hg. Water (8 ml) was added to the obtained dry residue, vigorously stirred (10 min), centrifuged (3000 rpm, 10 min), the liquid was decanted, the residue was dried to constant weight in an oven at 50 °C.

Mechanical treatment (MT) of SAn, AT or their mixtures was carried out in an MLW KM-1 ball mill (3000 rpm, ball weight 0.2 kg, applied load 20 N, sample weight ~ 0.5 g) and in a 75T-DrM vibration grinder (working body weight 1.13 kg; vibration acceleration 300-490 m·s⁻², vibration velocity 0.942-1.57 m·s⁻¹, applied load 339-550 N, sample weight ~ 4.5 g). The product was isolated from the dispersed mixture as follows: water (10 ml) was added to the sample of ~ 0.1 g, then it was stirred, centrifuged, separated from the solution with unreacted SAn and AT, dried as described above.

IR and NMR spectra were recorded using Specord 75IR spectrophotometer (range 4000-400 cm⁻¹, nujol mull) and Bruker Avance II spectrometer (400 or 100 MHz for recording ¹H or ¹³C NMR spectra) at 25 °C in DMSO-d₆. X-ray phase analysis was performed on a DRON-3 diffractometer with CuKα-radiation, wavelength α = 1.54181 Å (U = 30 kV, I = 20 mA).

3. Results and discussion

Four polar solvents were chosen as the medium for the synthesis in solution: water, DO, IPA, acetone. When the reaction was carried out in organic solvents, the product was a small-crystalline white powder with a melting point of 193-195 °C, which were practically insoluble in water. When using water as a solvent, the product could not be isolated.

After carrying out the reaction in DO, significant changes were noticeable in the IR spectrum of the obtained product in comparison with the spectra of the initial reagents (Figure 1): the NH amino group stretching vibrations bands in the region of 3410-3290 cm⁻¹ and the SAn anhydride group at 1860 and 1790 cm⁻¹ were complete disappeared; new absorption bands at 3460 (NH amide) and 1660 cm⁻¹ (C=O amide) were appeared. It should be noted that IR spectra were the same for products, which were obtained in different solvents.

In ¹H NMR spectra of the initial reagents were presented a singlet at 2.90 ppm (–CH₂– succinic anhydride); doublets at 6.53, 6.93 ppm (=CH– thiazole protons); singlet at 6.86 ppm, appropriating protons of 2-aminothiazole NH₂ group [4]. In the synthesized substance spectrum (Figure 2a), five proton signals are observed, of which two signals are multiplets 1, 2 and two ones are doublets 3, 4 with centers at 2.55, 2.65, 7.17, and 7.44 ppm, respectively. The integral intensities of these signals are in a ratio of 2:2:1:1, which makes it possible to assign signals 1 and 2 to the –CH₂–CH₂– group protons resonance of the succinic acid fragment, signals 3 and 4 to the –CH=CH– group protons resonance of the AT fragment. The signal of the AT amino group is absent, while a wide signal 5 appears at 11.94-12.33 ppm with a maximum at 12.11 ppm; the ratio of signals 3 and 5 integral intensities is 1:1.7. The type, position, and intensity of signal 5 are suggesting that it belong the NH and COOH groups protons in succinamide.

The ¹H NMR results complement by the ¹³C NMR data. The obtained ¹³C NMR spectrum (Figure 2b) fully corresponds to the spectrum of (2Z)-4-oxo-4-(1,3-thiazol-2-ylamino)-but-2-enoic acid (semiamide of succinic acid and AT), which was the theoretical modeling result by the ACDLABS software package [5].
Figure 1. IR spectra of SAn (1), AT (2), the product of SAn with AT interaction in solution (DO, 1 h, 60 °C) (3) and in a mechanoreactor (1 h, 20 N) (4).

Figure 2. Fragments of the spectrum $^1$H NMR (a) and $^{13}$C NMR (b) of the product of the reaction between succinic anhydride and 2-aminothiazole in acetone (1 h, 20 °C).

Thus, the NMR and IR spectroscopy data were indicated that a result interaction of the SAn and AT in polar solvents was formation of succinamide. The maximum product yield in DO (52 wt.%) and acetone (36 wt.%) was achieved at 40 and 10 °C, respectively (Figure 3, curve 1); an increase of the time synthesis at the above temperatures does not affected at the reaction course (Figure 3, curve 2). In IPA medium, the SA yield does not exceed 15 wt.%.
Figure 3. Dependence of the succinamide yield from temperature (1) for 1 h reaction and from synthesis time (2) for temperature 40 °C (a), 10 °C (b). Solvent: 1,4-dioxane (a), acetone (b).

For elimination the toxic solvents use, the interaction of the SAn and AT under the mechanochemical reaction conditions was studied. As shown earlier [6], mechanosynthesis, in comparison with the reaction in the liquid phase, can give a higher yield and conversion rate, and this approach is becoming more significant in the pharmaceutical industry [7].

The NMR and IR spectroscopy results showed that the starting anhydride and amine do not undergo chemical changes during mechanical treatment in a ball mill (1 h, 20 N). Their crystal structure is also preserved: there are no new reflections on the substance’s diffractograms (Figure 4) after their being in the mechanoreactor.

Figure 4. X-ray diffractogram of AT (a) and SAn (b) before (1) and after mechanical treatment in a ball mill for 1 h at 20 N (2).
At the same time, a significant decrease in the X-ray peaks intensity and their weak broadening were observed for SAn, which indicated on a decrease the anhydride crystals size and an increase in the microstresses level.

The X-ray diffractogram of a sample which separated after SAn and AT simultaneous grinding shows significant changes in comparison with X-ray diffractogram of physical mixture anhydride–amine (Figure 5): on the first one there are low intensity numerous reflections in the range from 17.0° to 24.6°, which obviously refer to amorphized amine crystals, and intense reflections at 25.1°, 26.4°, and 29.7°, which unambiguously point to new substance phases formation.

It was found completely agreement in the IR spectra (curves 3, 4 in Figure 1), as well as the position and form of signals in 1H NMR spectra, the value of melting points (193-195 °C) for the products which were obtained after interaction between SAn and AT both in organic solvents and in a mechanoreactor. This allows us to say that the mechanochemical reaction between succinic anhydride and 2-aminothiazole similarly reaction in solution, proceeds with the formation of a semi-amide according to the scheme:

\[
\begin{align*}
\text{N} & \quad \text{H}  \\
\text{O} & \quad \text{S}  \\
\text{O} & \quad \text{O}  \\
\text{O} & \quad \text{O}  \\
\text{H} & \quad \text{N}  \\
\text{S} & \quad \text{N}  \\
\text{H} & \quad \text{O}  \\
\end{align*}
\]

The degree of the reagent mixture conversion to semi-amide in a ball mill was increased with an increase in the processing time up to 2 h; a further increase in the duration of mechanoactivation somewhat reduced the efficiency of amidation (Figure 6a, curve 1).

Since mechanically activated processes are characterized by the separation in time of mechanical action and the formation of chemical bonds [8], the semi-amide content in mechanically treated mixtures was assessed not only soon after co-grinding of the reagents (after 1 h), but also during a long period after it, namely, for up to 45 days. It was established the influence of the mixture holding time after MT on degree of amidation. As seen in Figure 5a, in all the compositions under study, semi-amide accumulation took place during the entire selected period from the moment of mechanochemical treatment; the greatest increase in succinamide quantity occurred during the first 10 days after MT. Subsequently, the product accumulation occured to an insignificant extent, its
maximum amount (63-67 wt.%) was formed 45 days after co-grinding the mixture of SAn and AT in a ball mill for 2-3 h.

A similar relationship can be traced with an increase in the acting load. Thus, in a vibration attritor samples were ground under the action of a force equal to 339-550 N, i.e. ~ 20 times higher than in a ball mill, and the mechanoactivation of a mixture of SAn and AT in this conditions for 2 min allowed to isolate only 4 wt.% succinamide from the samples immediately after removing from the chamber of grinder (after 1 h). The formation of chemical bonds between SAn and AT proceeded to a greater extent after the removal of the load (Figure 6b). An increase in the applied force makes it possible to obtain practically the same amount of succinamide (65 wt.%) as when using a ball mill, but in a shorter time period, within 20 days.

4. Conclusions
The results obtained allow us to say that the interaction of succinic anhydride with 2-aminothiazole both in polar organic solvents and in a mechanochemical reactor leads to the formation of succinamide. In the solution, the largest amount of the product (≥ 50 wt.%) is formed in the 1,4-dioxane medium at temperatures of 20-40 °C. The solid-phase interaction passes through the stage of amorphization of the reagents, depends on the time of mechanical treatment, proceeds in time after the termination of the external force, and accelerates with an increase in the magnitude of the applied load. The efficiency of the process under the conditions of a mechanochemical reaction is higher in comparison with traditional synthesis in solution.

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