Preformulation Studies for Albendazole

A DSC and FTIR analysis of binary mixtures with excipients

CRISTINA TRANDAFIRESCU[1], CODRUTA SOICA[1], ADRIANA LEDETI[2], FLORIN BORCAN[2]*, LENUTA-MARIA SUTA[3], MARIUS MURARIU[4], CRISTINA DEHELEAN[5], DANIELA IONESCU[5], IONUT LEDETI[6]

[1] University of Medicine and Pharmacy Victor Babes, Faculty of Pharmacy, Department of Pharmaceutical Chemistry, 2 Eftimie Murgu Sq., 300041, Timisoara, Romania

[2] University of Medicine and Pharmacy Victor Babeº, Faculty of Pharmacy, Department of Analytical Chemistry, 2 Eftimie Murgu Sq., 300041, Timisoara, Romania

[3] University of Medicine and Pharmacy Victor Babes, Faculty of Pharmacy, Department of Pharmaceutical Technology, 2 Eftimie Murgu Sq., 300041, Timisoara, Romania

[4] University of Medicine and Pharmacy Victor Babes, Faculty of Pharmacy, Department of Toxicology, 2 Eftimie Murgu Sq., 300041, Timisoara, Romania

[5] University of Medicine and Pharmacy Victor Babes, Faculty of Medicine, Department of Surgery II, First Surgical Clinic, 2 Eftimie Murgu Sq., 300041, Timisoara, Romania

[6] University of Medicine and Pharmacy Victor Babes, Faculty of Pharmacy, Department of Physical Chemistry, 2 Eftimie Murgu Sq., 300041, Timisoara, Romania

The present study focuses on the evaluation of compatibility/incompatibility between the components of binary mixtures containing albendazole and six different pharmaceutical excipients currently used in solid formulations: colloidal silica (SiO_2), talc (T), magnesium stearate (MgST), starch (St), mannitol (Man) and polyvinylpyrrolidone (PVPK30). The binary mixtures were comparatively analyzed vs. pure components by means of DSC and FTIR spectroscopy.

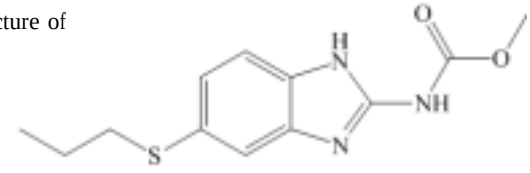
Keywords: albendazole, solid state, excipients, DSC, FTIR

Gallbladder injuries are common surgical specimens and most procedures are performed for cholelithiasis and cholecystitis [1]. Bile and bile acids exhibit antibacterial properties [2]; however, parasitic infection of the gallbladder may be associated with *Ascaris lumbricoides* [1,3] and *Giardia lamblia* parasitoses [4]. Biliary ascariasis is usually detected incidentally during surgery for cholelithiasis [5]. The growth and the encystations of *G. lamblia* depend on the dietary lipids, cholesterol, fatty acids and bile salts [6]. Giardia cysts can infiltrate the gallbladder and, subsequently, the common bile duct, which may lead to the modification of the bile in terms of composition and dynamic properties as well as pathological changes of the digestion [7]. Gallstones represent a risk factor for the development of gallbladder cancer [8] which was associated in one case report with trophozoites of *G. lamblia* [9].

Albendazole, methyl [5-propylthio)-1-H-benzimidazol-2-yl] carbamic acid methyl ester (ABZ) is a broad-spectrum benzimidazole derivative that acts against intestinal and systemic parasitoses [10]. ABZ exhibits anthelmintic activity on nematodes, cestodes and trematodes, being widely used in the treatment of human and veterinary medicine [11-14]. The anthelmintic mechanism of ABZ activity is based on its binding to parasite tubulin which leads to a disruption of the tubulin-microtubule dynamic equilibrium and, subsequently, to an alteration of basic cell functions [15]. ABZ also exerts its antiprotozoan activity in *Giardia lamblia* infections [16]; it interacts with the colchicine site of tubuline in the microtubules, leading to the disruption of their assembly [16].

Albendazole (MW 265.33 g/mol) is a white or slightly yellowish powder, with poor flow property and compressibility [17,18]. The drug is practically insoluble in water (1μg/mL at 25 °C, logP is 13.94) [18, 19]. ABZ has two pKa values, 2.68 and 11.83, respectively; at pH 1.2, the solubility of ABZ is 900 ìg/mLwhile at pH above 5 the solubility decreases at values under 1μg/mL [18]. The structure of ABZ is presented in figure 1.

Fig. 1. Structure of ABZ



ABZ forms colorless crystals with melting point of 481-483 K and has two polymorphs: ABZ Form I (metastable at ambient temperature, assigned to the commercialized drug) and ABZ Form II (enantiotropically related to form I); both forms are physically stable [19]. ABZ is a zwitterionic compound with log K_{ow} between 1.5 and 4 [20].

After oral administration ABZ is poorly and erratically absorbed from the intestinal tract, due to its slow dissolution rate and is converted by first-pass hepatic metabolism into its less active metabolite, albendazole-sulphoxide (ABZ-SO) [15,21]. ABZ-SO achieves variable concentrations in blood, bile, liver tissue and crosses the brain-blood barrier [15].

ABZ is classified by the Biopharmaceutics Classification System as a class II drug (BCS class II), having an extremely poor aqueous solubility which limits its oral absorption and

* email: fborcan@umft.ro

a high permeability through the biological membranes [10, 18, 22].

Formulation and preformulation studies include diverse thermoanalytical techniques for the identification of suitable excipients in order to obtain a dosage form in which the undesired interactions responsible for the drug–excipient incompatibility are minimized. The most frequently used analytical techniques for compatibility screening studies are thermal methods such as differential scanning calorimetry (DSC) [23,24], thermogravimetry and derivative thermogravimetry (TG/DTG) [25], hot stage microscopy (HSM) [26] and other instrumental methods, such as Fourier-transformed infrared spectroscopy (FTIR) [23-25], scanning electron microscopy (SEM) [27,28] and powder X-ray diffraction (PXRD) [29]. Thermal analysis is an important tool in the assessment of pharmaceutical mixtures containing one or more active substances, such as co-crystals which became widely investigated due to their increased bioavailability and enhanced pharmacological effects [30-32].

DSC is a thermoanalytical technique widely used to highlight the thermal transitions and the morphological changes of active pharmaceutical ingredients (API) and of their mixtures with different excipients, as well. By DSC, the melting peak of API is primarily analyzed. In case changes are observed in terms of shape and maximal value of the melting peak additional analytical techniques (usually FTIR) are used to support the incompatibilities observed on the DSC or heat flow curve [33, 34].

Following these considerations, in this study we set our objective to analyze several binary mixtures containing albendazole (ABZ) in association with various excipients.

Experimental part

Albendazole (ABZ) was obtained from Biesterfeld Siemsglüss, Hamburg, Germany (pharmaceutical grade) and used as received.The pharmaceutical excipients (pharmaceutical grade) were used as received as follows: colloidal silica (Aerosil 200Evonik Degussa, Germany), talc (Luzenac Pharma, Italy), magnesium stearate (Fluka, Germany), starch (Grain Processing Corporation, USA), mannitol (Merck, Germany), polyvinylpyrrolidone PVP K-30 (Sigma-Aldrich, Germany).

The binary mixtures of ABZ and excipient consisted of equal masses of ABZ and each excipient, respectively. Physical mixtures were prepared by simple mixing of the two substances in an agate mortar with pestle, for approximately 5 min. The 1:1 mass ratio was chosen in order to maximize the probability of observing potential interaction(s).

Samples were characterized using a Mettler-Toledo DSC1 instrument (Mettler-Toledo, Switzerland). Small amounts of samples (between 3.5 and 4.1 mg) were placed in aluminum crucibles with pierced caps and heated between 25-300°C in an inert argon atmosphere (100 mL/min flow) with a heating rate β=5°C/min. A reference material (empty aluminum crucible with pierced cap) simultaneously undergoes the same programmed time/temperature routine.

Attenuated Total Reflection Fourier Transform Infrared Spectroscopy (ATR-FTIR) spectra of the samples were obtained in attenuated total reflectance (ATR) mode on a Bruker Vertex 70 (BrukerDaltonik GmbH, Germany) spectrometer equipped with a Platinium ATR, Bruker Diamond Type A225/Q. Spectra were collected in the 4000-400 cm^{-1} spectral range, with a resolution of 1 cm^{-1} and with 32 co-added scans.

Results and discussions

ATR-FTIR spectroscopy

The ATR-FTIR spectroscopy was chosen as spectroscopic investigational tool since it has some advantages over classical KBr pelleting technique: the samples can be recovered and used for further investigations, a significant aspect especially for compounds with limited availability or expensive ones; however, the main advantage is represented by the fact that the sample is not subjected to pressing which can influence the polymorph state or solid packaging properties.

The main characteristic bands observed in the ATR-FTIR spectrum of ABZ are presented as follows: broad band in the spectral range 3413-3188 cm^{-1} with peak at 3317 cm^{-1} due to the stretching of N-H amine groups and overlapped with the vibrations of N-H bond from carbamate moiety; the sharp bands with reduced intensities due to the stretching of alkane-type C-H bonds from the propyl moiety appear around 2958 and 2868 cm^{-1}; the bending of the C=O bond from the carbamate moiety appears as a sharp band at 1712 cm^{-1}.

The aromatic benzoimidazolyl system is identified by several characteristic vibrations, including two intense overlapped bands in the 1610-1630 cm^{-1} spectral range, with peaks at 1629 and 1618 cm^{-1}. Another characteristic band for the aromatic system is the sharp band at 1525 cm^{-1} as well as the overlapped bands at 1441 and 1419 cm^{-1}. Other characteristic bands appear in the 960-760 cm^{-1} fingerprint region, but are difficult to be asset to certain moieties due to the overlapping with $C_{aliphatic}$-S and $C_{aromatic}$-S vibrations (705-670 cm^{-1} and 1090 cm^{-1}, respectively) and with numerous combination bands, as well.

As previously stated in our studies regarding the compatibilities of different APIs with excipients [29,33-35], the search for potential interactions should be carried out in the spectral regions of *reactive* functional groups since their modification is the most probable. The FTIR spectra of ABZ, excipients and binary mixtures are presented in figure 2.

Since our research group previously published some papers where the UATR-FTIR profiles of excipients were described [29,33], in the present study we will focus solely on the modifications of the position (shifting to lower or higher wavenumbers) or intensities for ABZ characteristic bands in binary mixtures *vs.* pure compound (table 1).

One can notice that the spectroscopic behavior of ABZ is similar in binary mixture and as pure sample, indicating that under ambient conditions the interactions are not clearly occurring. However, some modifications were recorded for the position of the band corresponding to the stretching of N-H group in the case of samples containing SiO_2, talc, PVPK30 and mannitol. Other functional bands appear at similar wavenumbers (a shifting of $\pm$3 cm^{-1} is not a clear indication of interaction(s)).

DSC Analysis

After performing ATR-FTIR analysis under ambient conditions, we analyzed the effect of thermal stress over the components of the binary mixtures, namely if it triggers interactions between API and excipients. In figure 3 the DSC curves determined for ABZ, binary mixture and pure excipient are represented.

The main approach in the analysis of the thermal treatment effect over binary mixtures consists in the

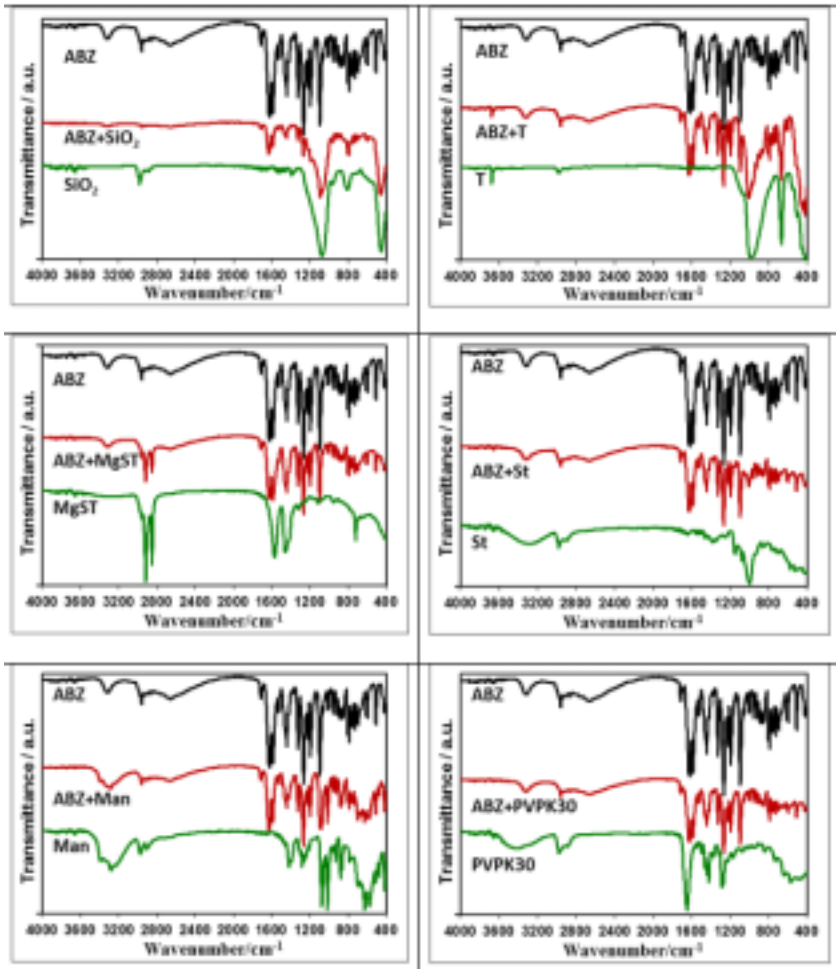


Fig.2.The ATR-FTIR spectra of albendazole (ABZ), binary mixtures and selected excipients in the 4000-400 spectral range

Table 1
THE ABZ BANDS IN BINARY MIXTURES WITH SELECTED EXCIPIENTS

Sample	Analysis of ATR-FTIR spectral regions (cm^{-1})		
	4000-2500	1800-1000	1000-400
ABZ	3317; 2958; 2868; 2659	1712; 1629; 1618; 1587; 1525; 1440; 1419; 1325; 1265; 1222; 1193; 1120; 1095; 1058; 1006	958; 923; 887; 864; 806; 790; 771; 759; 729; 696; 611; 597; 511; 422
ABZ+SiO_2	3325; 2958; 2868; 2659	1714; 1633; 1622; 1589; 1442; 1419; 1327; 1269; 1222; 1193; 1093; 1006	958; 923; 889; 864; 806; 792; 771; 761; 730; 696; 613; 597
ABZ+T	3323; 2958; 2868; 2661	1714; 1629; 1620; 1587; 1525; 1440; 1419; 1325; 1267; 1222; 1193; 1120; 1095; 1006	958; 923; 887; 864; 806; 790; 771; 761; 730; 696; 420
ABZ+MgST	3313; 2958; 2869; 2659	1714; 1631; 1620; 1587; 1525; 1440; 1419; 1327; 1267; 1222; 1195; 1120; 1095; 1006	958; 923; 887; 864; 806; 790; 771; 761; 730; 696; 420
ABZ+St	3317; 2958; 2869; 2661	1714; 1629; 1620; 1587; 1525; 1440; 1419; 1325; 1267; 1222; 1193; 1120; 1095; 1006	958; 923; 887; 864; 806; 790; 771; 759; 730; 696; 420
ABZ+Man	3286; 2958; 2868; 2661	1712; 1631; 1621; 1589; 1525; 1440; 1419; 1325; 1267; 1222; 1195; 1120; 1095	958; 923; 881; 864; 806; 790; 771; 761; 730; 694; 416
ABZ+PVPK30	3321; 2958; 2868; 2659	1712; 1630; 1620; 1587; 1523; 1440; 1421; 1325; 1267; 1222; 1193; 1122; 1096	958; 923; 887; 864; 806; 790; 771; 759; 730; 697; 422

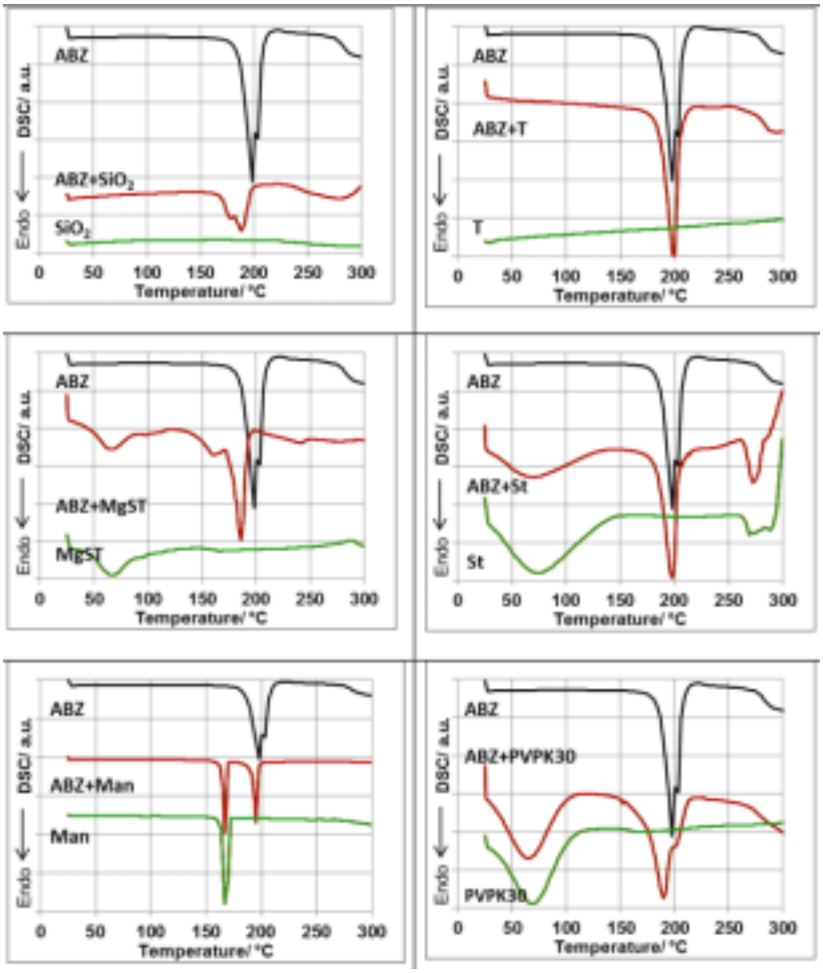


Fig.3. DSC curves for the analyzed binary samples in comparison with ABZ and each excipient, respectively

Sample	Thermal events revealed by the DSC curves (cm^{-1})			Interaction
	T_{onset} (°C)	T_{peak} (°C)	T_{offset}(°C)	
ABZ	169	198; 203	215	
ABZ+SiO_2	161	176; 186	199	++
ABZ+T	157	198	213	--
ABZ+MgST	35*; 130;	60*; 155; 185	84*; 195	++
ABZ+St	30*; 169; 258§	65*; 197; 271§	131*; 213; 282§	--
ABZ+Man	150*; 178	163*; 189;	174*; 203	++
ABZ+PVPK30	30*; 146	60*; 188; 197	105*; 213	++

Table 2
THE DSC DATA OBTAINED FOR BINARY MIXTURES VS. PURE COMPOUNDS. THE PEAKS MARKED WITH * AND § ARE DUE TO THE EXCIPIENT.

assessment of the modifications that appear in the melting point value of ABZ.

The DSC curve of ABZ reveals that two overlapped endothermal events are occurring within the 169-215°C temperature range. One can notice the appearance of two peaks at 198 and 203°C, respectively, the last corresponding to the melting process of the active substance. The overlapping of these two endothermal events is not associated with ABZ degradation or with the presence of impurities, but with a clear proof of the existence of enantiomeric compounds or polymorphic forms [36]. Due to the overlapping of thermal events, a correct estimation of the fusion enthalpy cannot be achieved. The DSC data obtained for binary mixtures are presented in table 2.

After a comparative analysis of the ABZ behavior in binary mixtures *vs.* pure ABZ, some major modifications were noticed in the thermal events of the samples. In case of ABZ+T and ABZ+St the DSC profiles are similar to the one of ABZ alone thus suggesting that thermal stress did not induce any interactions between the components; therefore they can be considered compatible. In all other situations the melting point of ABZ is drastically decreased in comparison to the pure drug possibly suggesting the formation of eutectic mixtures or chemical interactions between reactive functional groups. These excipients should be carefully analyzed in the final pharmaceutical formulations since they can clearly interact with ABZ.

Conclusions

In this paper we investigated the compatibility of albendazole with colloidal silica, talc, magnesium stearate, starch, mannitol and polyvinylpyrrolidone using ATR-FTIR spectroscopy, for the analysis of samples prepared under ambient conditions, followed by the effect of thermal stress on binary samples.

The DSC curves of the mixtures of albendazole with six excipients, respectively, clearly indicated the presence of

some interactions between ABZ and polyvinylpyrrolidone, mannitol, magnesium stearate and colloidal silica.

These observations are of valuable applications in the pharmaceutical technology field especially in the development of new generic formulations where a careful selection of excipients may lead to oral pharmaceutical formulations with improved pharmacokinetic properties, stability and longer shelf-life.

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