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At-line analysis of pharmaceutical nanofiber-products using ATR-FTIR spectroscopy

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Introduction

Attenuated total reflectance (ATR) is a quick and non-destructive sampling technique for obtaining the mid IR spectrum of a material's surface[1]. In a typical ATR configuration (Figure 1), the infrared beam enters a high refractive index ATR crystal and is internally reflected. This results in an evanescent wave, which penetrates the sample in contact with the ATR crystal and produces a reflection/absorption spectrum[2]. ATR crystal is called the internal reflection element (e.g., zinc selenide, diamond, silicon or germanium)[3] and all types of samples (e.g. solids, liquids, powders, pastes, pellets, slurries, fibers etc.) can be placed undiluted on the ATR crystal. The measurement then is typically performed within seconds[4].

Attenuated total reflectance Fourier transform infrared (ATR-FTIR) spectroscopy has showed the benefits in direct determination of niflumic acid in pharmaceutical gel[5], some anti-inflammatory in tablets and hard gelatin capsules[6], drug (ketoconazole) release from suspension[7], sorbitol and sodium glutamate in yellow fever vaccine[8], methylamphetamine seizures[2], sibutramine adulteration in tea and coffee[9] and metformin in a human single hair fiber[10]. To date, drug-loaded nanofibers are novel pharmaceutical products that have many advantages for drug delivery and release[11,12]. However, there is no report of ATR-FTIR spectroscopy used as quantitative tools for determination of active pharmaceutical ingredients in nanofibers. Thus, the aim of this work was to study the potential of ATR-FTIR method for quantitative analysis of the active constituent loaded in pharmaceutical-nanofiber products.

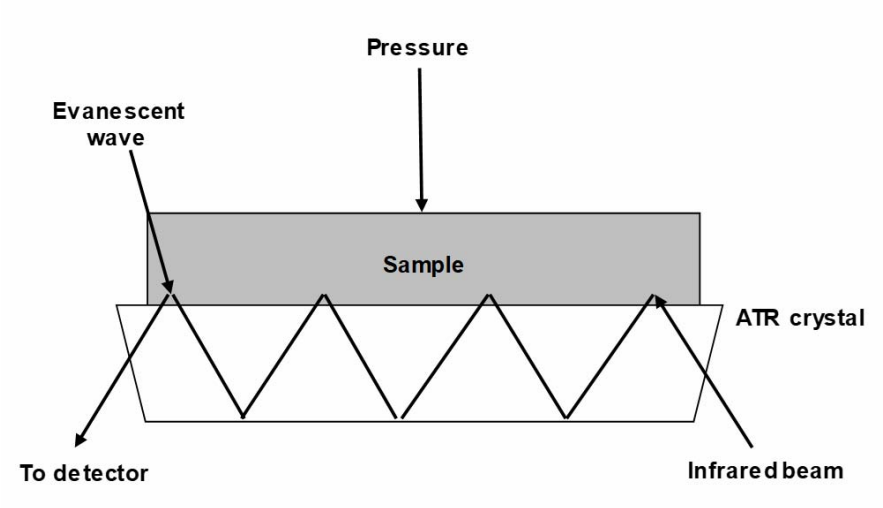

Figure 1. General ATR-FTIR spectroscopy setup

Methods

Preparation of electrospun α-arbutin nanofibers

α-Arbutin (Figure 2) loaded in cellulose acetate (Figure 2) nanofibers were prepared using electrospinning process described in the previous work[13]. The calibration set has concentrations of 1.5, 1.8, 2.1, 2.4, 2.7, 3.0, 3.3, 3.6, 3.9, 4.2, 4.5 and 4.8% (w/w) of α-arbutin in cellulose acetate nanofibers, respectively. In this work, 3.0% w/w α–arbutin in cellulose acetate nanofiber mat (Figure 2) was prepared as the prototype sample for analysis. The morphology and diameter of α-arbutin nanofiber samples were examined using scanning electron microscopy (SEM, Quanta 200, FEI with genesis EDAX).

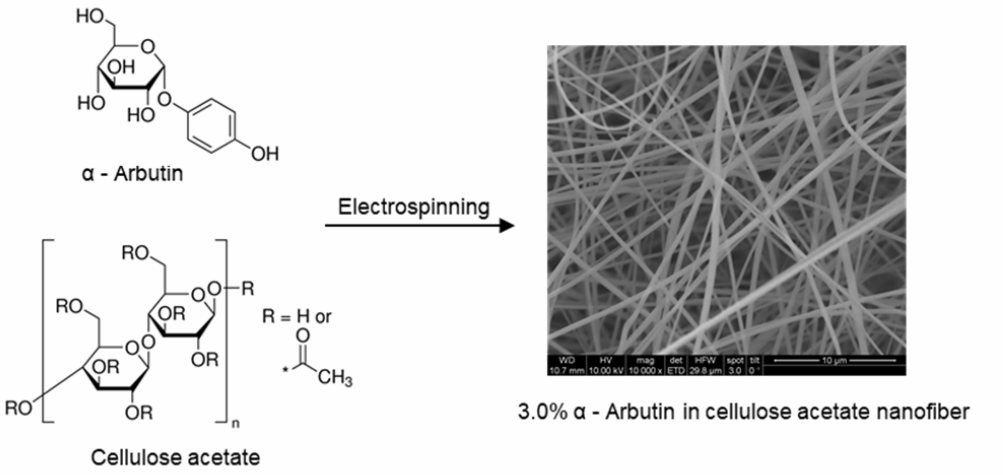

Figure 2. Structures of α-arbutin and cellulose acetate and an SEM image of 3.0% α-arbutin in cellulose acetate nanofibers with 10K magnification

ATR-FTIR spectroscopic method

ATR-FTIR spectra of all samples were collected on a Bruker Tensor 27 Fourier transform infrared spectrometer with a Mercury-Cadmium-Telluride (MCT) liquid N_2 cooled detector and configured with a single reflection diamond ATR accessory (Platinum ATR, Bruker). Each spectrum was recorded at ambient temperature in the region of 500-4000 cm^{-1}, in absorbance mode, by an average of 32 scans with a resolution of 4 cm^{-1}. Spectra were acquired using OPUS software version 6.2 provided by Bruker. Before scanning each sample, a background spectrum was taken by recording the clean and dry ATR crystal against air. Raw spectral data were pre-processed using a baseline correction followed by standard normal variate (SNV) scaling and mean center correction, then partial least squares (PLS) modelling was applied using Matlab 7.0 (The Mathworks). Cross-validation using the leave one out technique was performed to estimate the performance of the constructed models. The predicted concentrations of the components in each sample were compared with the actual concentrations in the calibration samples and root mean square error of cross-validation (RMSCV) was calculated to select the optimal PLS latent variables (LV).

Results

The SEM images of all α-arbutin in cellulose acetate nanofibers showed the uniaxial alignment with the average diameter of in the range of 500–600 nm. The ATR-FTIR spectra in the range of 500-2,000 nm, of α-arbutin, cellulose acetate and 3.0% α-arbutin in cellulose acetate nanofibers are presented in Figure 3. α-arbutin has the specific bands at 779, 831, 1010, 1078, 1214 and 1514 cm^{-1} while cellulose acetate has the specific band of at 1738 cm^{-1}. The bands in spectrum at 779, 831 and 1514 cm^{-1} of 3.0% α-arbutin in cellulose acetate nanofibers demonstrated that the drug encapsulated in the nanofibers. ATR-FTIR spectra of standard nanofibers were collected and built the quantification model. For the PLS model the dataset was cut off at 1700 cm^{-1}, because at higher wavenumbers no significant contribution of sample can be expected. The spectrum was pre-processed using SNV scaling and mean centering. The values of PLS model parameters are presented in Table 1.

Table 1. Parameters of the PLS model based on ATR-FTIR spectroscopy

Parameters		
Number of latent variables	3	4
R^2	0.9860	0.9937
REMSECV	0.4701	0.3973

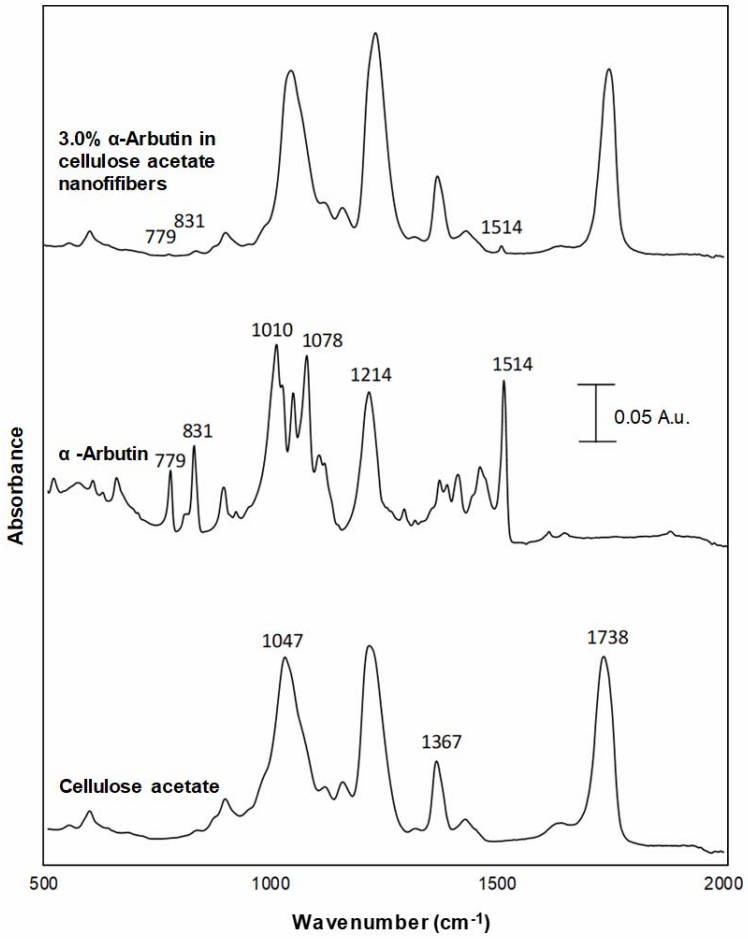

Figure 3. ATR-FTIR spectra in the range of 500-2,000 nm, of 3.0% α-arbutin in cellulose acetate nanofibers (top), α-arbutin (middle) and cellulose acetate (low)

Discussion

FTIR-measurements performed in ATR mode are very comfortable. The analytical procedure requires no sample preparation and reagent-free measurement. After the background spectrum was measured, the intact nanofiber sample was cut in a small piece and placed on the stage, then compressed on ATR crystal and scanned against the background of the clean crystal. This process is rapid and the ATR-FTIR spectrum was obtained within a few minutes.

When integrating the specific bands for each of the two components and calculating the ratio of α-arbutin/cellulose acetate. The resulting datapoints can be fitted linearly with a correlation coefficient of 0.993, which proves that α-arbutin should be distributed evenly amongst cellulose acetate nanofibers, since the measurement area with the ATR is big enough to include several nanofibers. The band at 1514 cm^{-1} provides the most information of the α-arbutin content in the fibers when measured with ATR-FTIR. PLS models built from ATR-FTIR spectra allowed the determination of α-arbutin concentrations in nanofiber samples without any sample preparation. The results showed a good correlation between actual and found values of this model when 4 components were chosen with the RMSECV of 0.3973 and the cross-validation correlation coefficient (R^2) value of 0.9937 (Table 1). However, the method validation of ATR-FTIR method should be conducted to facilitate the integrity of the method.

Conclusion

The ATR-FTIR spectroscopy in combination with PLS calibration demonstrated the potential for quantifying α-arbutin content in cellulose acetate nanofibers since it was efficient, easy-to-use and fast. The method could be a strong candidate as a process analytical technology (PAT) tool for pharmaceutical nanofiber-manufacturing process allowed at-line detection of drug concentration in real-time because of its non-destructive, non-invasive and minimal time-consuming aspects.

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