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# Prediction of drug dissolution profiles from tablets using NIR diffuse reflectance spectroscopy: A rapid and nondestructive method

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**Abstract**

A comparison between dissolution profiles obtained by using a dissolution apparatus (conventional method) and the NIR diffuse reflectance spectra of a series of clonazepam-containing batches is reported. Ten different formulations with fixed amount of clonazepam and varying proportions of excipients were analyzed at seven dissolution times and three different media. The percentages of dissolution of each sample were correlated with the NIR spectra of three tablets of each batch, through a multivariate analysis using the PLS regression algorithm. The squared correlation coefficients for the plots of percentages of dissolution from the equipment laboratory (dissolution apparatus and HPLC determination) versus the predicted values, in the leave-one-out cross-validation, varied from 0.80 to 0.92, indicating that the NIR diffuse reflectance spectroscopy method is an alternative, nondestructive tool for measurement of drug dissolution from tablets.


*Keywords:* NIR diffuse reflectance; Dissolution profiles; Clonazepam-containing tablets

## 1. Introduction

One of the most common practices in the pharmaceutical industry is the determination of dissolution profiles in drug tablets. This is currently performed using a dissolution apparatus and often requires a long time analysis. Efforts have been done to implement lesser cost and time-consuming methods in order to predict dissolution from tablets, specially using near-infrared spectroscopy [1,2]. In a recent study [2], the dissolution profile from tablets of different hardness were successfully evaluated, but other applications of NIR have special place in the pharmaceutical research. Classical examples are the use of diffuse reflectance or transmittance measurements to quantify actives in tablets [3], to determine content uniformity in tablets [4], and others concerning to pharmaceutical and chemical sciences [5–9].

Treatment of NIR spectra for quantitative analysis, as well as other spectroscopic data (such as UV [10,11] and fluorescence [12] spectra), is usually done by multivariate analysis in order to correlate them with the intended dependent variable. Partial least squares (PLS), principal component regression (PCR) and classical least squares (CLS) are some of the regression methods frequently used for quantitative purposes [10–12]. In our study, we have applied the PLS regression method to correlate the NIR diffuse reflectance spectra with the percentages of dissolution obtained through the conventional method, since it is an well established method for quantitative multicomponent analysis.

Thus, the investigation to be tested in this paper is to encounter a linear relationship between percentage of drug dissolved from tablets and NIR absorbance, using clonazepam-containing tablets with different proportions of excipients as samples. By calibrating the NIR spectra using the dissolution results from the laboratory equipment as dependent variables, it is possible to predict, in all media

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tested, the dissolution profiles of the tablets, which are the percentage of drug dissolution in determined collection times.

## 2. Experimental

### 2.1. Materials

Ten batches of clonazepam-containing tablets were used, containing the following formulations: clonazepam (1.25%), pregelatinized starch (2–10%), cellulose microcrystalline 102 (5–72.95%), lactose 50/70 (32.45–85.95%), magnesium stearate (0.8%), sodium lauryl sulfate (0–1.5%), colloidal silicon dioxide (0–1%) and lactose 200 (0–60%).

### 2.2. Dissolution

Three tablets from each batch were evaluated for dissolution by using a Hanson dissolution apparatus. The dissolution testing was performed in 900 mL distilled water at $37 \pm 0.5\,^{\circ}C$, and the percentage of drug dissolution from each tablet was measured at different time intervals (3, 7, 11, 15, 20, 30 and 45 min), at three different media (pH 3.0, 4.5 and 6.8), using a Waters HPLC with an UV–vis detector ($\lambda_{max} = 254$ nm). USP apparatus and conditions were used to run dissolution and acetate (pH 3.0 and 4.5) and phosphate (pH 6.8) buffers were used for the present purpose. The HPLC conditions were: an isocratic system with a Waters® 515 HPLC Pump, 2487 Dual Absorbance Detector, 2475 Multi Fluorescence Detector, 717 Plus Autosampler and Temperature Control Module. The column used was a reversed phase column Symmetry C18 (Waters 4.6 mm × 250 mm, 5 μm). The mobile phase was composed of buffer ammonium phosphate dibasic pH 8.0/methanol/tetrahydrofuran (600:520:130), filtered through 0.45 μm membrane (Millipore®) and degassed during 5 min. The flow rate was 1.5 mL/min, injection volume 50 μL, temperature 45 °C. The detector was adjusted to 254. The running time for analysis was 8 min. Data were obtained using the software Millennium 132 Chromatography Manager.

### 2.3. NIR analysis

NIR reflectance spectra were measured using a Perkin-Elmer Model Spectrum One NTS spectrophotometer. Each measured spectrum was the average of 32 scans, obtained with resolution of 8 $cm^{-1}$ and over the range of 4000–10000 $cm^{-1}$. Three tablets from each batch were taken to be included in the calibration set and, consequently, to be used as samples in the cross-validation tests, and their spectra were processed using the spectrum software (Perkin-Elmer). No sample preparation was needed, being the entire tablet analyzed through diffuse reflectance. Spectra were also treated and correlated with the dissolution results by using the Pirouette 3.11 program [13].

## 3. Results and discussion

The goal of this work is to develop a methodology that allows us to predict formulations with adequate dissolution profiles in a simple, rapid and reliable manner. Since NIR spectroscopy has been applied for diverse purposes worldwide, it is supposed to be applied successfully in prediction of dissolution percentages, in selected media and collection times, to tablets of a series of clonazepam-containing batches. For that a training set, namely the calibration compounds, was used to build the model, which was tested through leave-one-out cross-validation. The calibration set was composed of 30 samples, where 3 tablets of each of the 10 batches were used to have their NIR spectra correlated with the percentages of dissolution obtained through HPLC, at three different pH's and seven sampling times. As mentioned above, the method was validated by leave-one-out cross-validation, i.e. each sample was left out from the calibration set once, the calibration set was remodeled and the dissolution percentage of the left out sample was predicted using the new calibration model. The results obtained from this analysis are discussed as follows.

NIR reflectance spectra (Fig. 1) were acquired as log (1/*R*), where *R* is the reflectance, and converted into percentage of transmittance (just as an usual way to illustrate plots), and the mean-centered spectra were used in the multivariate calculations without any further pretreatment. Each spectrum was correlated with the corresponding drug dissolution percentage, obtained for each of the seven collection times and at three dissolution media, which are shown in Table 1. PLS was the algorithm utilized for the multivariate regression and, then, 21 calibrations were obtained.

Good correlation results of predicted dissolution percentage by NIR versus dissolution percentage by laboratory equipment (dissolution apparatus and HPLC determination) were obtained and are shown in Table 2. The optimum number of factors found to represent the models was 10, since the residuals in both calibration and cross-validation barely changed when using more than 10 factors. The standard errors of validation varied from 2.75 to 4.96 for medium with pH 3.0, from 2.89 to 7.88 for

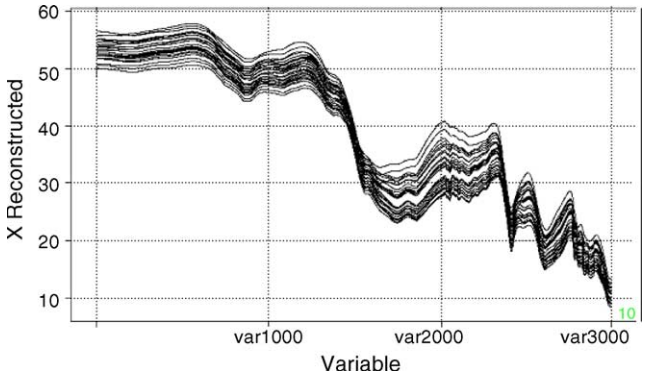


Fig. 1. Mean-centered spectra for the clonazepam-containing tablets.

Table 1
Dissolution profiles (%) for the 10 batches studied

| Batch | 3 min | 7 min | 11 min | 15 min | 20 min | 30 min | 45 min |
|---|---|---|---|---|---|---|---|
| pH 3.0 | | | | | | | |
| 01 | 6.52 | 14.48 | 20.70 | 24.52 | 29.03 | 35.74 | 42.37 |
| 02 | 0 | 7.16 | 12.12 | 16.03 | 19.55 | 26.16 | 34.31 |
| 03 | 12.43 | 27.92 | 35.82 | 40.12 | 45.76 | 51.46 | 57.69 |
| 04 | 7.48 | 17.91 | 24.45 | 28.55 | 32.22 | 35.98 | 38.77 |
| 05 | 8.82 | 20.50 | 25.70 | 29.32 | 32.26 | 35.56 | 39.87 |
| 06 | 19.32 | 37.29 | 47.29 | 52.68 | 56.54 | 60.80 | 63.18 |
| 07 | 19.45 | 33.86 | 41.00 | 45.38 | 48.84 | 53.55 | 57.57 |
| 08 | 10.58 | 22.05 | 30.22 | 35.62 | 40.21 | 46.13 | 50.13 |
| 09 | 12.40 | 20.84 | 26.34 | 29.80 | 32.86 | 38.50 | 43.25 |
| 10 | 17.87 | 41.52 | 51.56 | 56.32 | 60.74 | 64.28 | 65.72 |
| pH 4.5 | | | | | | | |
| 01 | 7.90 | 15.09 | 21.70 | 26.34 | 30.86 | 38.08 | 44.54 |
| 02 | 1.01 | 2.85 | 7.26 | 6.87 | 10.36 | 14.15 | 21.86 |
| 03 | 7.67 | 20.57 | 28.13 | 32.30 | 37.21 | 43.27 | 49.46 |
| 04 | 5.55 | 15.85 | 26.04 | 32.77 | 38.33 | 46.10 | 51.14 |
| 05 | 7.12 | 20.63 | 27.73 | 34.13 | 38.19 | 44.80 | 48.71 |
| 06 | 23.39 | 45.37 | 57.87 | 65.99 | 73.03 | 81.62 | 87.89 |
| 07 | 20.21 | 38.50 | 48.51 | 55.25 | 61.01 | 69.06 | 75.72 |
| 08 | 21.20 | 37.45 | 47.11 | 52.61 | 58.35 | 65.70 | 71.36 |
| 09 | 18.14 | 33.06 | 40.38 | 45.20 | 48.91 | 54.28 | 59.20 |
| 10 | 20.12 | 44.92 | 57.35 | 63.32 | 70.75 | 76.37 | 79.98 |
| pH 6.8 | | | | | | | |
| 01 | 10.41 | 18.30 | 24.16 | 27.99 | 31.81 | 37.24 | 43.12 |
| 02 | 0 | 0.91 | 3.52 | 5.50 | 7.59 | 11.71 | 16.36 |
| 03 | 4.94 | 15.59 | 22.47 | 26.32 | 30.18 | 37.25 | 41.91 |
| 04 | 6.39 | 18.74 | 23.97 | 29.27 | 33.81 | 39.86 | 46.59 |
| 05 | 10.67 | 21.39 | 25.15 | 29.64 | 33.85 | 39.41 | 45.04 |
| 06 | 30.08 | 51.87 | 61.68 | 66.63 | 71.12 | 75.52 | 79.18 |
| 07 | 25.21 | 42.72 | 50.49 | 55.70 | 59.21 | 64.44 | 68.67 |
| 08 | 22.22 | 45.42 | 57.03 | 62.63 | 66.85 | 72.34 | 76.60 |
| 09 | 18.15 | 35.69 | 43.18 | 48.56 | 52.54 | 57.54 | 61.82 |
| 10 | 32.22 | 60.23 | 70.53 | 76.01 | 80.02 | 83.25 | 85.81 |

Each dissolution percentage is an average of three HPLC measurements.

medium with pH 4.5 and from 2.96 to 6.34 for medium with pH 6.8. The cumulative variance using 10 factors was 99.99%. Fig. 2 illustrates and exemplifies some of the calibration/validation plots built using the methodology here proposed.

Tablet weight, diameter, thickness, hardness and disintegration were consistent in all batches. The NIR spectra varied sufficiently among the batches, as can easily be seen in Fig. 1, to represent any excipient modification in formulation, and this is markedly interesting by using this multivariate method instead of a univariate approach, since in this way the whole variance among spectra may be better appraised. Although loadings analysis has indicated higher weights for variables in the 4924–5052 $cm^{-1}$ (variables from 5076 to 4948) and 5330–6646 $cm^{-1}$ (variables from 4670 to 3354) regions, using factor number one, these selections have not improved the predictive ability of the model, according to leave-one-out cross-validation results. Evaluation with first and second derivatives, as well as multi-scattering correction, also did not provide any benefit on the prediction quality. Curves of predicted versus measured percent of dissolution were consistent for all assays, for both calibration and cross-validation tests, indicating that the dissolutions were carried out in a reproducible way in the three media studied, at the seven collection times, as well as the NIR measurements. All these, together with the similarity between predicted and experimental data, allow us to consider the presented method as an alternative approach for measurement of dissolution as percentage of drug dissolved tablets, namely the clonazepam-containing formulations described above.

Table 2
Squared correlation coefficients of calibration and cross-validation

| Time (min) | pH 3.0 | | pH 4.5 | | pH 6.8 | |
|---|---|---|---|---|---|---|
| | $R^2_{cal}$ | $R^2_{val}$ | $R^2_{cal}$ | $R^2_{val}$ | $R^2_{cal}$ | $R^2_{val}$ |
| 3 | 0.97 | 0.80 | 0.98 | 0.86 | 0.99 | 0.92 |
| 7 | 0.98 | 0.84 | 0.98 | 0.86 | 0.99 | 0.91 |
| 11 | 0.98 | 0.85 | 0.98 | 0.86 | 0.99 | 0.91 |
| 15 | 0.98 | 0.84 | 0.98 | 0.86 | 0.98 | 0.91 |
| 20 | 0.96 | 0.82 | 0.98 | 0.86 | 0.99 | 0.91 |
| 30 | 0.98 | 0.86 | 0.98 | 0.85 | 0.99 | 0.91 |
| 45 | 0.98 | 0.85 | 0.98 | 0.83 | 0.99 | 0.92 |

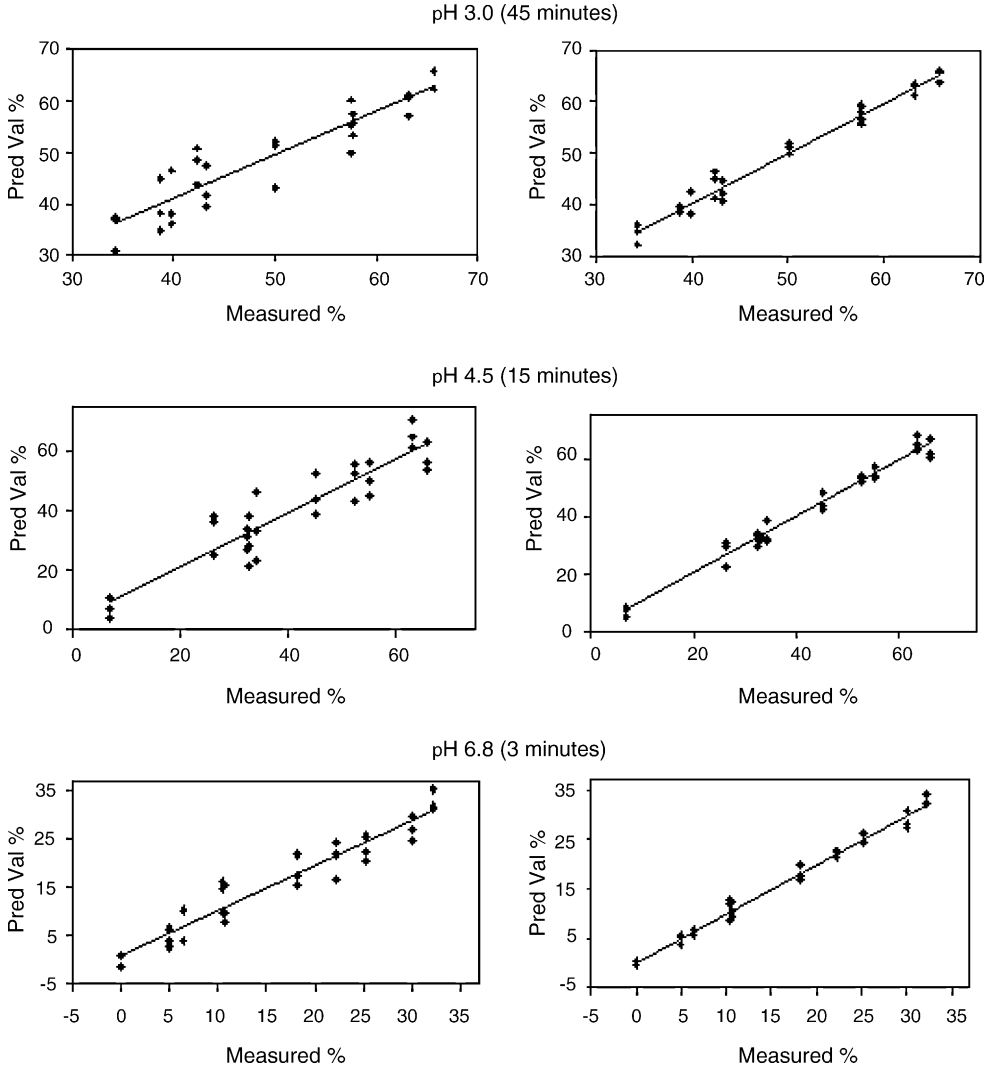


Fig. 2. Some calibration (left) and cross-validation curves, illustrating the correlation between the measured dissolution percentages (obtained through the traditional method) and the predicted ones (obtained by NIR).

External validation tests were also carried out in order to corroborate the cross-validation results. Such tests consisted in excluding one batch (three samples) from the original training set, proceed with calibration using the remaining nine batches and predict the percent of dissolution for the excluded batch. Batches 4, 6, 7, 9 and 10 gave correlation between predicted and experimental values higher than 0.98, while the other batches did not reach so a good prediction because one or more of their excipients are in extreme content. Thus, these results confirm the good predictive ability of the model.

A previously reported study of dissolution prediction using NIR [2], for tablets with varying hardness, given as different compression forces when manufacturing them, rendered squared correlation coefficients in validation tests from 0.72 to 0.86, indicating that our approach presents comparable, from equal to better, predictive ability, but now applied to the study of dissolution profiles of tablets differing in content of excipients.

## 4. Conclusions

The NIR diffuse reflectance procedure had the advantages over the reference technique of requiring no sample preparation and acquiring results in a fast manner. PLS demonstrated to be an adequate regression method in building the calibration models, without recourse to spectra transforms in addition to the mean-centering pretreatment. The presented method potentially provided sufficient assistance in the course of developing formulations with an adequate dissolution profile, since all of the 21 curves of predicted versus measured dissolution percentages were consistent and presented satisfactory correlation.

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