# Abstract
# A novel method to determine aminoglycosides in pharmaceutical solid formulations

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## Introduction

Aminoglycosides are a large class of antibiotics characterised by two or more aminosugar groups linked by glycosidic bonds to an aminocyclitol component. This class of compounds is water soluble, polar, and non-volatile with hydroxyl and amino groups in their structures. The lack of chromophore or fluorophore makes the analyses of these antibiotics very difficult by UV spectroscopy. Several analytical methods have been developed, mainly based on derivatisation reactions. The United States Pharmacopeia (USP) and the European Pharmacopeia (EP) describe a microbiological assay for the quantification of aminoglycosides in pharmaceutical products. Microbiological assays are very difficult to perform. They are expensive, time-consuming, lingering and the results are not very precise. For these reasons, simpler and more robust methods are needed for the quantification of this class of compounds.

Near infrared (NIR) spectroscopy has the potential to be used to quantify aminoglycosides without the need of derivatisation reactions or other types of sample pre-processing. This paper proposes a new method for the quantification of aminoglycosidic antibiotics based on NIR spectroscopy, using neomycin as an example. Neomycin is a broad spectrum aminoglycoside antibiotic used in gastrointestinal infection for the inhibition of Gram-negative and Gram-positive bacteria.

## Materials and methods

Two different types of samples were produced: synthetic and "doped" samples. Samples were based on a commercial formulation containing neomycin sulfate as active pharmaceutical ingredient (API), and three excipients: lactose, talc and magnesium stearate. The synthetic samples formulations were defined accordingly to an experimental design and produced by weighing the individual formulation components and mixing them in a glass mortar prior to the NIR analysis. To produce the "doped" samples the commercial tablets were macerated and mixed with neomycin sulfate to obtain the higher API concentration samples. The lower API concentration samples were obtained

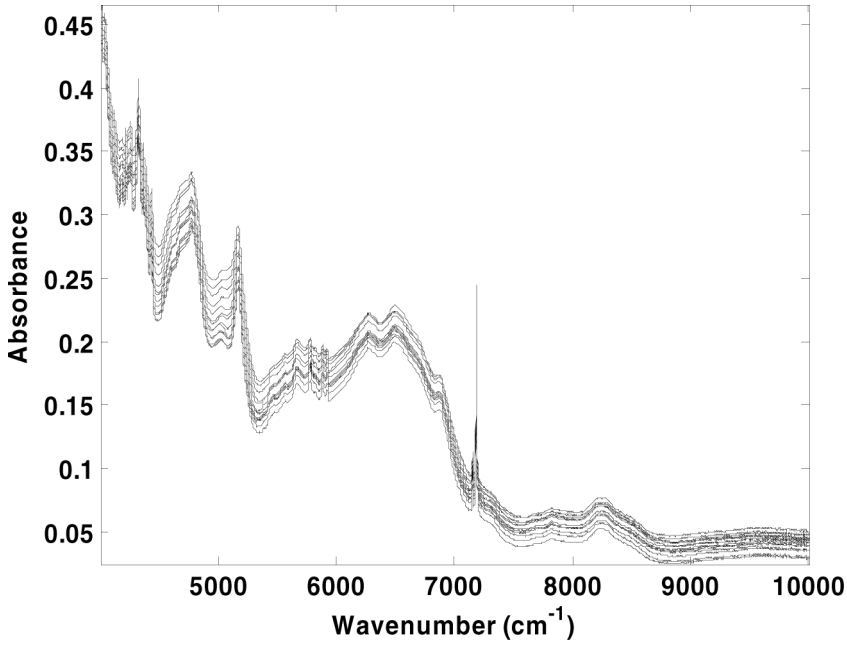


**Figure 1.** Synthetic samples NIR raw spectra.

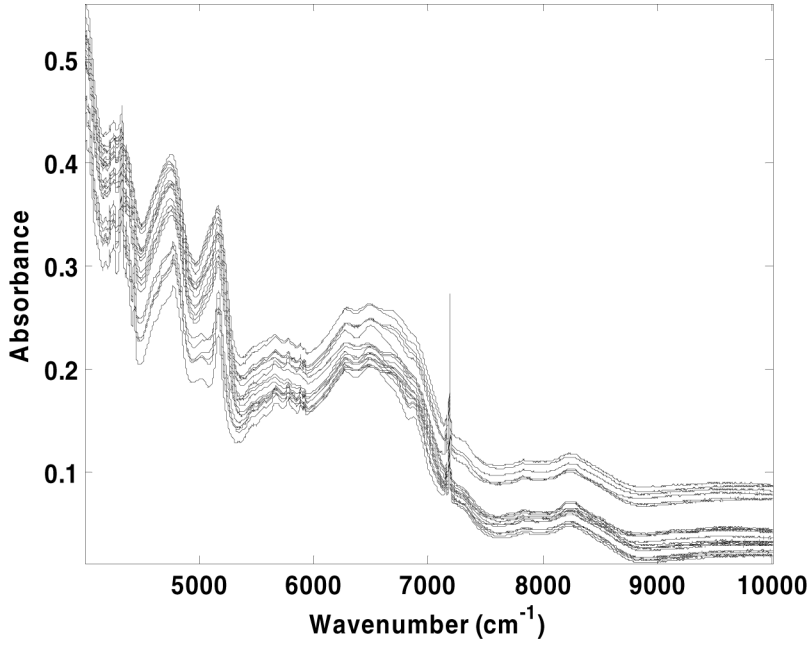


**Figure 2.** Doped samples NIR raw spectra.

by diluting the macerated tablets in a mixture of the formulation excipients. For each sample type, two sets were produced: one for calibration and one for validation. The samples were measured in reflectance mode with a FTNIR instrument (FTLA2000, ABB Bomem, Québec, Canada) equipped with an Indium–Gallium–Arsenide (InGaAs) detector, operating between 4000 $cm^{-1}$ and 10,000 $cm^{-1}$, with a resolution of 2 $cm^{-1}$, and 64 scans. Figures 1 and 2 show the raw spectra of the synthetic and doped samples, respectively.

Partial least squares (PLS1) with leave-one-out cross-validation was used to correlate the NIR spectra with the concentration of neomycin sulfate. The amount of neomycin sulfate present in the samples was confirmed by an HPLC method, with a pre-column derivatisation of the neomycin sulfate with phenylisocyanate, using UV detection at 240 nm. The results obtained with NIR were discussed in terms of figures of merit and compared with the HPLC method.