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Near-infrared spectroscopy as a tool to improve quality[1]

W. Plugge, C. van der Vlies*

Gist-Brocades B.V., Industrial Pharmaceutical Products Division, P.O. Box 1, 2600 MA Delft, The Netherlands

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Abstract

Near-infrared spectroscopy, together with a polar qualification system, is an ideal technique to distinguish between identical pharmaceutical substances differing only slightly in chemical or physical characteristics. Among the attractive applications of this powerful combination are the more precise definition and selection of starting materials used in the manufacturing of medicinal products and its use in blending validation.

Keywords: Lactose; NIR spectroscopy; Pharmaceutical substances; Polar qualification system; Quality point; Validataion

1. Introduction

The vast majority of pharmaceutical ingredients are powders used "as is" or blended, directly or after a simple or complex process. All these powders interact with their environment by way of their surfaces and it is therefore important to be able to measure surface characteristics relevant to their suitability.

Recently, Buckton [1] published a comprehensive review on the effects of processes such as milling, micronization, compaction, spray drying, lyophilization and blending on the surfaces of powder particles. The effects of stress induced during these processes—either chemical or physical in nature—may result in a dramatic change in behaviour but may be very difficult to analyse with traditional methods.

Impurities present on the particle surfaces, e.g. from residual mother liquor, may be low in terms of percentage but they may change in a significant way the behaviour of the powder in the manufacturing process or in the final medicinal product. Therefore, a methodology is needed to detect such deficiencies at the surfaces or in the upper layers of powders. Spectroscopic reflectance techniques of sufficient sensitivity and precision could be of help to discriminate between apparently identical materials from different sources or to detect inconsistencies between batches from the same source. Near-infrared (NIR) spectroscopy is such a technique. It has found its way into pharmaceutical control laboratories in recent years for raw material identification, water analysis and other pharmaceutical analyses [2–5].

In earlier papers, the effect of physical parameters on the reflectance NIR spectrum was often considered a disadvantage but for the modern pharmaceutical analyst this is a blessing in dis-

* Corresponding author.

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guise. Recently Dreassi et al. [6] demonstrated that the technique, applied in the reflectance mode, is well able to discriminate between materials identical according to the compendial monographs, but different in physical aspects. Establishing differences in particle size and powder densities and the presence of polymorphs and distinguishing a levo form from its racemic mixture can be done precisely and rapidly by comparing NIR spectra.

Although the actual recording of spectra by an NIR spectrometer is fast and simple, the mathematics as applied by Dreassi and others in the literature is of considerable complexity. This complexity has caused difficulty with the acceptance of NIR spectroscopy by official bodies for the final control procedure of starting materials and medicinal products. However, the good news is that recently the Ph. Eur. Commission, by approving a general monograph on NIR spectroscopy for identification of pharmaceutical substances, recognized this technique as an official alternative for the identification tests of the monographs. Unfortunately, the USP does not seem to follow this initiative.

One of the most frequently applied mathematical treatments in qualitative NIR analysis is PCA (principal components analysis). PCA reduces the vast amount of information collected in a spectrum into a small data set containing the most relevant information. Although popular among chemometricians, PCA is of a complexity exceeding the comprehension of most pharmaceutical analysts and licensing authorities.

The objective of this paper is to demonstrate with two examples the enormous potential of NIR spectroscopy in combination with PQS (polar qualification system), a relatively simple but very effective mathematical treatment of spectroscopic data that can be successfully used to improve and control the quality of medicinal products [7,8].

2. Equipment and materials

The instruments used in these laboratories are Model 5000 NIR spectrometers from NIR Systems Inc., (Silver Spring, MD) equipped with a rotating drawer to accomodate the powder sample cell. The software to operate the spectrometer is IQ² (1.11) as supplied by NIR Systems. The specific software to run PQS has been written by Plugge in PASCAL 7.0. PQS itself and its mathematical description has been described in detail recently by the authors [8]. All samples have been generously supplied by various companies, were of standard commercial quality and were measured as powders without further treatment.

3. PQS

In PQS, spectra are not presented in a linear way—absorbance plotted against wavelength—but in a polar coordinate system in which the absorbance values are the radii and the angle α is a function of the wavelength. In this paper all spectra are transformed first into their second derivatives and the absolute values of the transformed absorbances are used to obtain the polar spectra. The reason for this transformation is the fact that a polar spectrum has a well-defined center of gravity that may represent a specific quality parameter. In such a polar spectrum, the

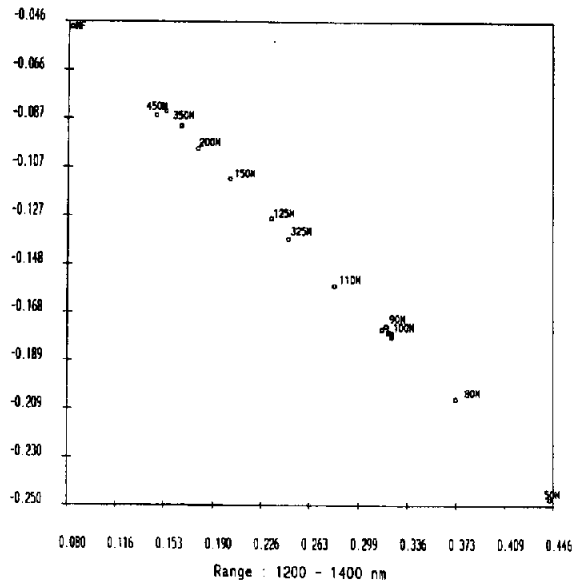

Fig. 1. Quality points (centres of gravity) of the polar spectra of the second derivative NIR spectra of 12 different grades of lactose monohydrate (range 1200–1400 nm).

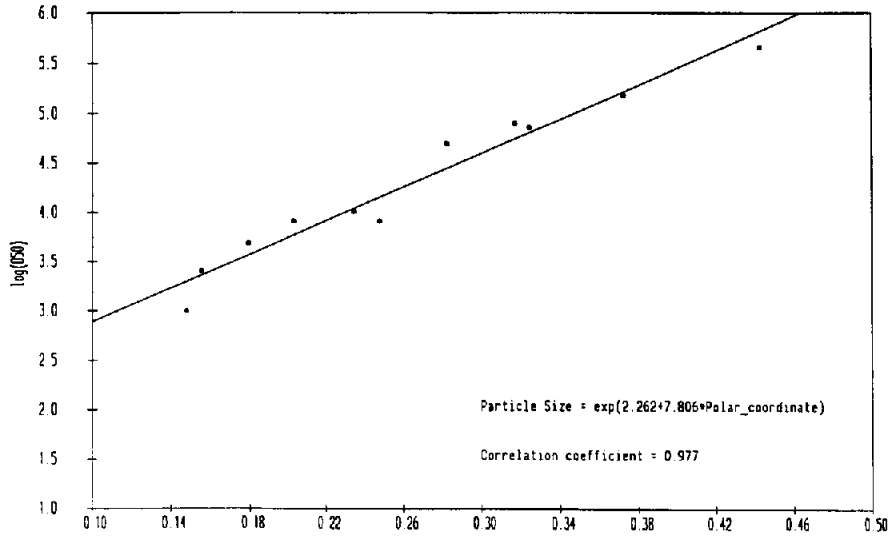

Fig. 2. Correlation between the polar coordinates of the quality points and the logarithms of the assigned d_{50} values of 11 grades of lactose monohydrate (range 1200–1400 nm).

noise is randomly distributed around the circle of the polar coordinate system. Therefore, noise has very little effect on the centre of gravity itself and noticeable differences between spectra must be due to effects other than pure variation in the spectrum.

By applying this simple mathematical treatment, the wealth of information present in an NIR spectrum is in PQS reduced to one or more centres of gravity or quality points, each lying in a two-dimensional plane and defined by two coordinates and a wavelength range.

4. Applications

4.1. Variation in lactose

The effects of the particle size distribution and other physical characteristics of lactose monohydrate on its performance in pharmaceutical manufacturing is well known but little understood. Because of the expected effect of particle size on reflectance the influcene of the grade of lactose on its quality point has been investigated. In Fig. 1 the quality points of 12 lactose samples of different grades ranging from 50M (average $d_{50} = 290$ μm) to MF (microfine, average $d_{50} \approx 5$ μm) are shown for the range 1200–1400 nm. These samples all came from the same manufacturer with the exception of the microfine sample. The cluster of quality points in Fig. 1, obtained by recording independently ten aliquots from the 100M sample, demonstrates the precision of this parameter. Fig. 1 also shows the accuracy of this technique. The quality points relating to the 90M and 325M samples are found between the points relating to 100M and 125M which seems odd at first sight. However, when looking at the average d_{50} values assigned to these grades by the manufacturer, the positions found for these two grades are correct.

If the x or the y coordinates of the quality points in Fig. 1 are plotted against the logarithm of the average d_{50} values as supplied by the manufacturer, a nice linear correlation (correlation coefficient 0.977) is found between the quality points and the assigned particle sizes (Fig. 2). However, in some cases there is deviation from the model, most probably due to large variation in the d_{50} values assigned to these grades by the manufacturer and to the fact that d_{50} values do not distinguish between samples with different distribution patterns.

At this point, it should be emphasized that the coarse grades (50M, 80M, 100M, 90M, 110M, 325M and 125M) are sieved fractions of crys-

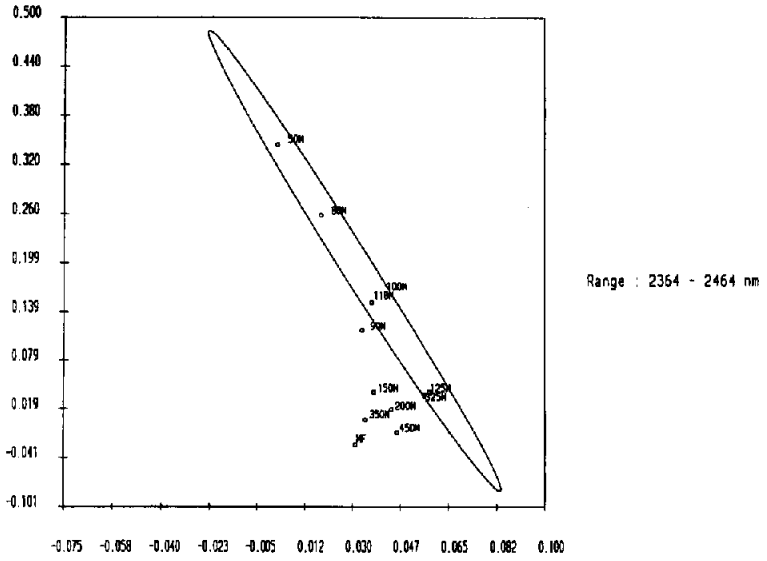

Fig. 3. Quality points of the polar spectra of the second derivative NIR spectra of 12 different grades of lactose monohydrate, obtained by sieving and milling (range 2364–2464 nm).

talline monohydrate whereas the finer grades 150M, 200M, 350M and 450M are obtained by milling. To find out whether this difference in processing has a measurable effect on the NIR reflectance of the samples, the whole NIR region has been scanned in steps of 100 nm. In Fig. 3 the quality points of the sieved grades and their 99% confidence ellipse show that for the region 2364–2464 nm there is still a linear relation with particle size. The quality points due to the grades obtained by milling clearly belong to a different set. This suggests that by applying PQS and careful selection of a specific wavelength range, it is also possible to distinguish between sieved and milled lactose from this manufacturer.

In Fig. 4 the quality points of 30 samples of lactose marketed as "crystalline", "anhydrous" and "powder" are collected together with their

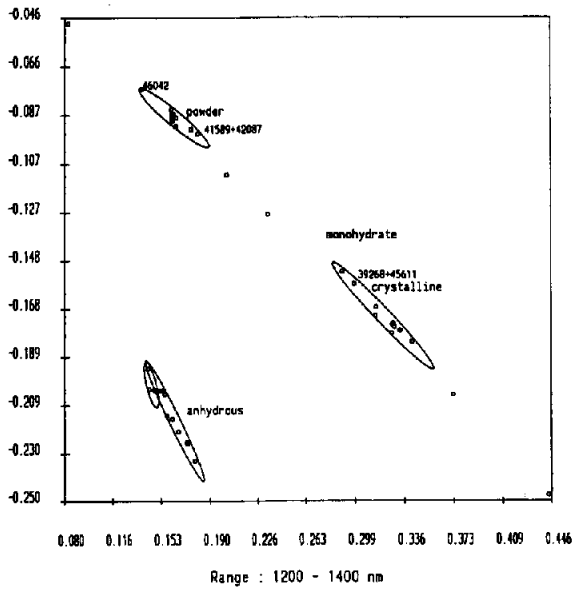

Fig. 4. Quality points of 30 samples of crystalline, anhydrous and powdered lactose monohydrate from different sources (range 1200–1400 nm).

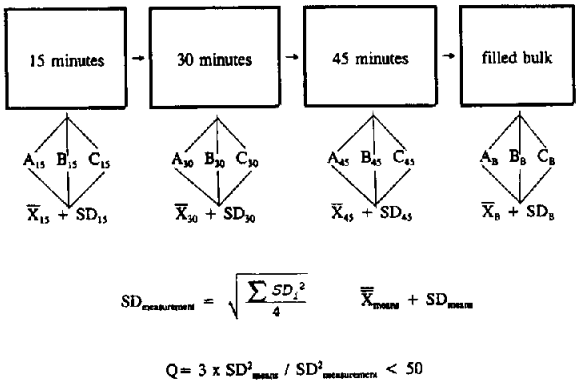

Fig. 5. Experimental design of the preliminary validation study of a three-component blending operation.

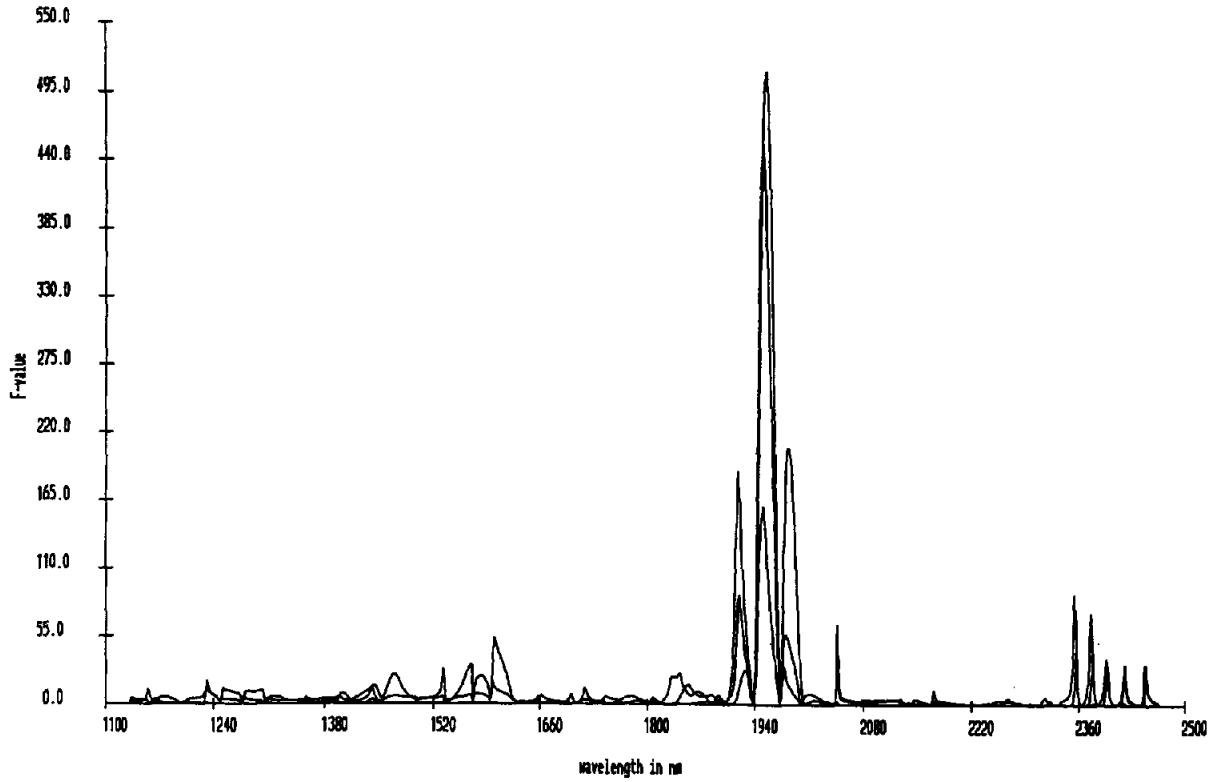

Fig. 6. *F*-spectra of three blends (1, 2 and 3), indicating inhomogeneity between the samples of each blend.

99% confidence ellipses. The 50M, 80M, 125M, 150M and MF samples are added for reference. These 30 samples were all supplied by one pharmaceutical company, with the exception of four anhydrous samples, the quality points of which are enclosed by the very small ellipse. No additional information about the supplier(s), particle sizes or final process steps had been received initially.

The variation between the samples labelled as "crystalline" is larger than might be expected for one quality from one source. Inquiry showed that the batches 39268 and 45611 came from a second supplier. The position of their quality points (Fig. 4) suggests a finer particle size but they passed the rather uncritical test on receipt. The variation found for the other "crystalline" samples, although from one manufacturer, at least suggests that several grades are involved.

The samples labelled as lactose "powder" came from three different sources. The points labelled as 41589 and 42087 in Fig. 4 came from a second supplier and were found to be outside the specification (too coarse) when tested by the intended user. The one sample from the third supplier (labelled as 46042) is clearly the finest of all "powder" samples and did meet the criterion of the sieve analysis.

The quality points for the "anhydrous" samples were all found to be on a straight line about parallel with the line found for the monohydrates. Their variation suggested significant differences in particle size between the samples which could be confirmed by microscopic analysis. For the wavelength range 1200–1400 nm, the direction of these two lines is apparently the direction in which the centre of gravity moves when the particle size changes. However, the direction orthogonal to the particle size line is very probably the direction along which the centre of gravity moves when the

content changes from almost 0% for the anhydrous sample to about 5% for the monohydrate. Unfortunately, the number of samples available did not allow a firm conclusion to be reached on this point. Nevertheless, if in PQS a quality point deviates significantly from that expected, the direction of the deviation simply indicates which part of the spectrum is responsible for that deviation. In the case of PCA a thorough knowledge of the principles of the method is needed to interpret deviating results. Moreover, a substantial number of spectra are needed to obtain reliable estimates for the principal components.

The above results clearly show that by using NIR spectroscopy and PQS it is simple to verify whether a sample of a newly-arrived batch of lactose monohydrate has the correct identity, whether the particle size is consistent with previously received material and, probably, whether it has the correct water content. This, together with the ability to distinguish between batches processed in different ways, may be of great importance to improve and maintain the quality of the final dosage form the batch is intended for.

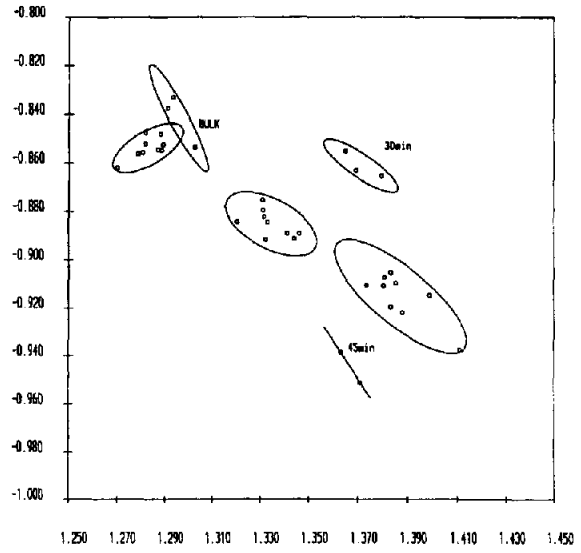

Fig. 7. Quality points of the 12 individual spectra of each of the three blends, identifying the samples responsible for the inhomogeneity (range 1850–2050 nm).

4.2. Blending validation

The requirement that blends of pharmaceutical ingredients have to be homogeneous is self-evident but how to demonstrate that homogeneity is achieved has been the subject of much debate. In any case, the analytical effort for such a validation study is quite extensive if traditional methods like HPLC have to be used. Because of the simplicity of the NIR technique and the sensitivity of the NIR spectrum for small deviations in chemical composition, it was attractive to investigate whether NIR spectroscopy could be of help in a blend validation study.

The blend investigated contained three sterile components in weight ratio of 88:10:2. A preliminary study was carried out to establish the required blending time to reach homogeneity. In this case, samples can be taken aseptically but from only one point of the blender. Samples are taken after 15, 30 and 45 min blending and from a final bulk container after filling (Fig. 5).

To allow measurement of the sample in triplicate, 25 g samples are taken from the blender at each point in time and from the filled bulk container. Each individual sample is homogenized in the laboratory before splitting into three aliquots to obtain three independent NIR spectra. Because of this further homogenization, the variation within the samples as obtained from these spectra will be rather small. Since this variation is used as a reference to detect possible differences between samples, a better sensitivity is obtained.

Although normally not considered to be good laboratory practice, in this case homogenization of the 25 g sample can be justified. The weight of the maximum unit dose for this product is 1 g. The 25 g sample can be considered as the sum of 25 independent unit doses each with an allowed variation of 10% (90–110%). By blending the 25 units to an homogeneous quantity, the allowed variation in that larger homogenized sample is reduced from 10%

$$\pm\frac{10}{\sqrt{25}} = \pm 2\%$$

or an allowed variation of 98–102% in the homogeneized sample. Knowing from previous experi-

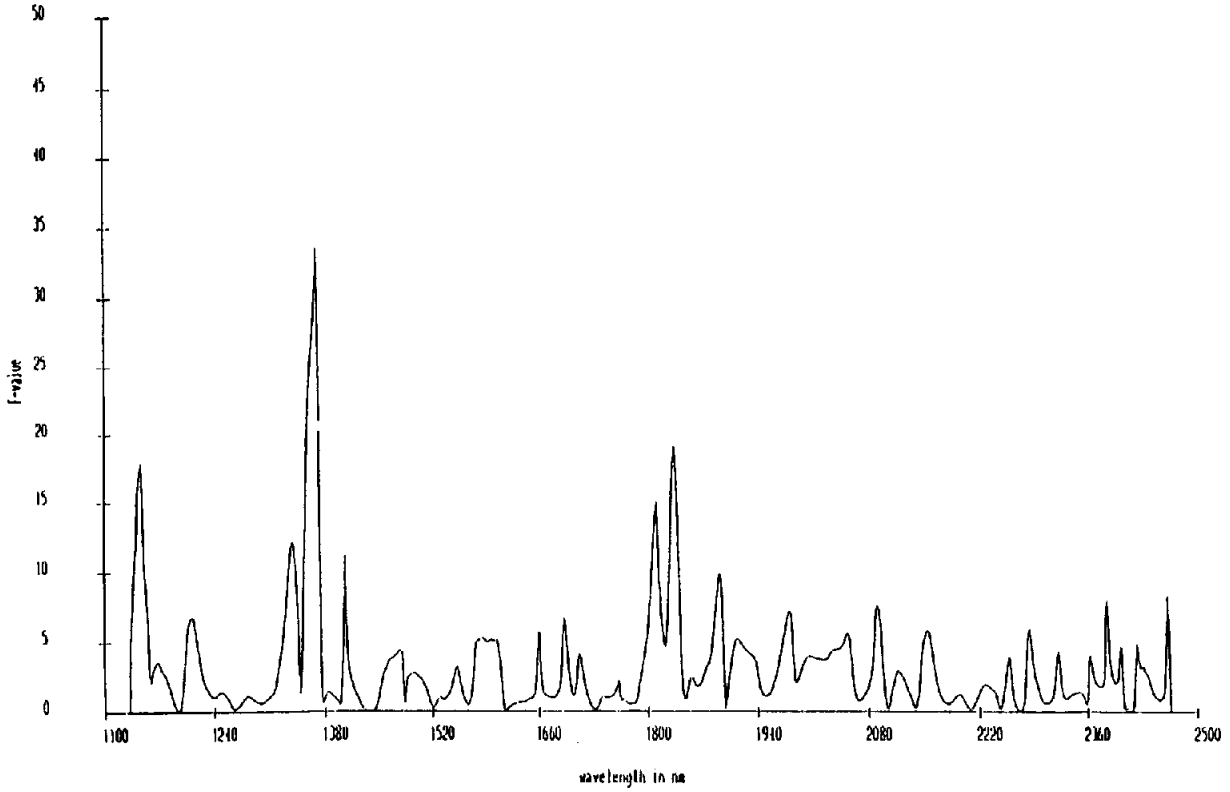

Fig. 8. *F* spectrum of the two-component mixture after milling the coarsest component.

ments that the variation in NIR measurements is about 0.5%, a variation exceeding 2% should be easy to detect.

The three spectra obtained for each point in time (A_{15}, B_{15}, C_{15}, etc. for $i = 30$, 45 min and the bulk) are averaged to obtain the average spectrum X_i along with the standard deviations SD_i for each wavelength. Assuming that the sample is perfectly homogeneous at the analyzed sample size level, this standard deviation (SD_i) is a measure of the variation due to differences between sample cells, the fillings and the actual measurements. Because the experiment was carried out at four points in time, the pooled standard deviation for a measurement is

$$SD_{measurement} = \sqrt{\frac{\sum SD_i^2}{4}}$$

Next, the standard deviation for each wavelength of the four X_i spectra is calculated. This standard deviation (SD_{means}) is a measure of the variation between the samples taken at different times from the blender. This whole preliminary exercise has been carried out on three different blends. In the ideal situation of homogeneity at all points in time, the ratio at each wavelength of

$$\frac{3SD_{means}^2}{SD_{measurement}^2}$$

will be close to 1[2]. Taking into account the very small variation in the NIR measurements (small *denominator*) and accepting a variation in the homogenized samples of about 0.5%, a ratio of 50 can be accepted as the maximum for a homoge-

[2] Because the three spectra from one point in time are averaged, the variance is a factor of three lower, hence the factor in the numerator.

neous sample, according to the tabulated values for an F distribution.

For the three experiments carried out as described, the F values are calculated and plotted as F spectra (Fig. 6). From this Figure it is obvious that the variation between the four samples of blends 2 and 3 is unacceptably large ($F > 400$). In the case of blend 1, the F value was much lower but still above 50. From these F spectra, however, it is not possible to conclude which of the samples cause these large variations in the range 1850–2050 nm. Using PQS and selecting this wavelength range, the quality points calculated for the 12 spectra of each blend very clearly indicated which sample was responsible for the large variation (Fig. 7). The only possible conclusion on the basis of this preliminary experiment was that with this mixture it was not possible to obtain a uniform blend at any time.

An analytical investigation revealed that the two minor components could not be blended to a homogeneous pre-mix, probably due to a small but incompatible difference in particle size. After milling the smallest component, an homogeneous pre-mix could be obtained as was demonstrated by the F-spectrum (Fig. 8). Whether a really homogeneous three-component blend can be obtained has still to be proven.

5. Conclusions

A modern NIR spectrometer connected to a powerful computer and sophisticated software can be an excellent tool for the pharmaceutical analyst.

NIR spectroscopy in combination with PQS is an extremely useful technique to establish chemical and physical product characteristics. Quality points at well chosen wavelength ranges are sensitive parameters to monitor the consistency of starting materials, intermediates and final products.

PQS is based on simple mathematics, compared with more traditionally used methods such as PCA, partial-least squares, Mahalanobis distances and others. This will ease the acceptance of NIR analysis as a final control technique by licensing authorities and manufacturers.

With PQS it is possible to separate certain parameters, which influence each other in the original spectrum, in such a way that they can be estimated independently.

NIR coupled with analysis of variance is a powerful tool to demonstrate homogeneity. In the case of non-homogeneity, PQS can help to detect the deviating samples.

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