Available online at www.sciencedirect.com
SCIENCE DIRECT
Journal of Pharmaceutical and Biomedical Analysis 41 (2006) 89–98
JOURNAL OF PHARMACEUTICAL AND BIOMEDICAL ANALYSIS
www.elsevier.com/locate/jpba

Transfer of NIR calibrations for pharmaceutical formulations between different instruments

Eva-Lotta Bergman [a,*], Henric Brage [b], Mats Josefson [a], Olof Svensson [a], Anders Sparén [a]

[a] *AstraZeneca R&D Mölndal, SE-431 83 Mölndal, Sweden*
[b] *Södra Cell FoU, SE-430 24 Väröbacka, Sweden*

Received 20 September 2004; received in revised form 19 October 2005; accepted 19 October 2005
Available online 6 January 2006

Abstract

In order to evaluate how well existing techniques for transferring NIR calibrations perform for solid pharmaceutical formulations, a study on four assays of active ingredients was undertaken. The study included two configurations of dispersive NIR instruments and one Fourier transform (FT) instrument. Three methods for calibration transfer: slope/bias correction, local centring and piecewise direct standardisation (PDS), were tested and evaluated.

Our conclusions are that the calibration transfer methods tested can perform equally well. It was shown that it is possible to transfer calibrations between instruments of different configurations or even of different types, without loosing the prediction ability of the calibration. To achieve a good calibration transfer, a larger variation in the content of the active ingredient in the samples and more samples are needed for the slope and bias correction method compared to the local centring method. For PDS to be a successful calibration transfer method, an optimisation of the number of transfer samples and how they are selected together with various factors specific for this method is needed.

Local centring is the preferred transfer method as its performance is excellent yet it is simple to perform, no optimisation is needed, only a few transfer samples are required and the transfer samples do not have to vary in their content of the active ingredient.

Keywords: Calibration transfer; Local centring; Slope and bias correction; Piecewise direct standardisation; Dispersive instrument; Fourier transform instrument

1. Introduction

Near infrared (NIR) spectroscopy has become a commonly used analytical technique in many industrial applications due to its rapidness and the fact that it is non-destructive to the samples. NIR calibrations developed on one NIR instrument, usually referred to as the master instrument, may perform less well on another instrument, usually referred to as the slave instrument, because of differences in the hardware. These differences can be due to differences in, e.g. gratings, spectral resolution, different or changed light sources, fibre optics and other changes due to instrumental wear with time. A number of non-instrumental factors might also cause the spectra to change significantly, e.g. change of measuring cell for the sample presentation, experimental conditions, change of raw material manufacturer or a slight change in the formulation of the product under investigation. Some of these changes can, or should, be included in the calibration from the beginning, but unexpected changes may always occur. If the experimental or hardware conditions are changed it can be a serious obstacle to the use of NIR spectrometry for calibrations. As calibration models are in most cases based on a large number of samples that require considerable time and cost for collection, preparation and measurement, it is important to easily and rapidly be able to transfer the calibration without re-measuring too many samples. Ideally, there should be small enough differences between the instruments, so that this could be done in a seamless way.

Many methods for NIR calibration transfer have been suggested and there are a number of excellent reviews on this topic [1–4]. Calibrations can be adjusted by using a simple univariate slope/bias correction of the predicted values [2,3] or by using multivariate correction of the spectra using, e.g. direct standard-

* Corresponding author. Tel.: +46 31 7762806; fax: +46 31 7763768.
E-mail address: eva-lotta.bergman@astrazeneca.com (E.-L. Bergman).

0731-7085/$ – see front matter © 2005 Elsevier B.V. All rights reserved.
doi:10.1016/j.jpba.2005.10.042

isation (DS), piecewise direct standardisation (PDS) [5] or the patented method developed by Shenk and Westerhaus [6].

Slope/bias correction of the predicted values has previously been compared with the more complicated piecewise direct standardisation method based on a multivariate correction of the spectra [7,8]. If the differences are global, then slope/bias correction works as well as PDS. Local centring has previously been compared to orthogonal signal correction (OSC [9]), PDS [10] and curve fitting methods [11] as a transfer method. The results showed that local centring performed very well compared to the other methods. Recently, a study showed that slope/bias correction and local centring, among other methods, worked well as transferring methods for quantitative NIR calibrations [12].

The aim of this study was to compare the calibration transfer techniques slope/bias correction, local centring and PDS for a representative set of working calibrations. The three techniques were applied to four NIR PLS-models predicting the content of active ingredient in four pharmaceutical formulations of coarse composite powders. The calibrations were all developed on dispersive NIR instruments. They were then transferred to other dispersive instruments of the same or of a different configuration. One of the calibrations was also transferred to a Fourier transform instrument. Between 65 and 178 samples were used in the training sets.

2. Materials and methods

2.1. NIR instruments and software

The spectra were acquired on five instruments (Table 1). Four of them were of the dispersive type (model 6500, FOSS NIRSystems, Inc., Silver Spring, MD, USA), two of which were equipped with a fibre optic probe and two of which were equipped with a rapid content analyser (RCA) module. The NIR instruments equipped with a probe are denoted NIR_Probe1 and NIR_Probe2, respectively. The NIR instruments equipped with an RCA module are denoted NIR_RCA1 and NIR_RCA2, respectively. Disposable polyethylene flasks (50 ml, Kebo Lab, Lund, Sweden) having a bottle neck size in which the fibre optic probes fit were used as measuring cells for the probe set-up. The same sample was measured in triplicate and in between every measurement the flask was turned upside down. Glass beakers (50 ml, Duran, Mainz, Germany) were used as measuring cells for the RCA modules. The same sample was measured in duplicate and in between the measurements the sample was poured from the first to a second beaker. The instruments were equipped with PbS and Si detectors and a tungsten-halogen filament lamp was used as the source of radiation. The spectra were collected using the software Vision 2.21 or 2.51 (FOSS NIRSystems, Inc.). The spectra were acquired in the wavelength range 400–2500 nm. The bandwidth was 9–10 nm. The digital resolution was 2 nm and the number of scans per spectra was set to 32.

The fifth instrument was a Fourier transform (FT) instrument (model MB 160 PH, ABB Bomem Inc., Que., Canada) equipped with a PowderSampl*ir* unit. This instrument is denoted NIR_FT. Disposable scintillation vials (Kimble Glass Inc., Vineland, USA) were used as measuring cells. The instrument was equipped with InAs detectors and a quartz-halogen lamp was used as the source of radiation. The spectra were collected using the software *Airs Professional 2.1* (ABB Bomem Inc.), in the wave number range 12,000–400 cm^{-1} (833–2500 nm). The resolution was 16 cm^{-1} and the number of scans per spectra was set to 128. As the spectra collected on the FT-instrument are expressed in wave numbers (cm^{-1}) while the spectra collected on the dispersive instruments are expressed in wavelengths (nm), the spectra collected on the FT-instrument had to be transformed into wavelengths (nm). This was done in *Matlab 6.0* (The Mathworks, Natick, MA, USA). Because of different resolution along the spectrum, new intensity values were calculated by interpolating with respect to the template scale (nm).

All raw spectra were imported into *Simca 8.1* (Umetrics AB, Umeå, Sweden). The first and second derivatives of the spectra were calculated using an in-house software written in Visual Basic. All calibration transfer calculations were carried out in *Matlab 6.0*. The *PLS_Toolbox 2.1* (Eigenvector Research, Lake Manson, WA, USA) was used for PDS.

2.2. NIR calibration models

The four assay calibration models transferred in this study are listed in Table 2. The first calibration model that needed to be

Table 1
Instruments used in the study

Label	Manufacturer	Type	Configuration	Detectors
NIR_RCA1	FOSS	Dispersive	RCA	Si/PbS[a]
NIR_RCA2	FOSS	Dispersive	RCA	Si/PbS[a]
NIR_Probe1	FOSS	Dispersive	Probe	Si/PbS[a]
NIR_Probe2	FOSS	Dispersive	Probe	Si/PbS[a]
NIR_FT	ABB Bomem	Fourier transform	Powder sampl*ir*	InAs[b]

[a] The Si detectors are used in the wavelength range 400–1100 nm and the PbS detectors in 1100–2500 nm. Only wavelengths in the range 1100–2500 nm were used in the calibration models.

[b] The wavelength range for the InAs detectors is 833–2500 nm.

Table 2
Calibration models, transfer methods tested and master and slave instruments for each model

Calibration model	Transfer method	Master instrument	Slave instrument(s)
P1	Slope/bias	NIR_Probe1	NIR_RCA1
	Local centring		NIR_RCA2 NIR_Probe2
R1A	Slope/bias Local centring	NIR_RCA1	NIR_RCA2
R1B	Slope/bias Local centring PDS	NIR_RCA1	NIR_RCA2
R1C	Slope/bias Local centring PDS[a]	NIR_RCA1	NIR_RCA2 NIR_FT

[a] PDS was only tested when calibration R1C was transferred to NIR_FT.

transferred was developed on one of the dispersive probe instruments, NIR_Probe1. This calibration was designated P1. The aim was to transfer calibration P1 to the other dispersive probe instrument (NIR_Probe2) and to the two dispersive RCA-instruments (NIR_RCA1 and NIR_RCA2). In the calibration set used to create the PLS-model for calibration P1, 178 samples measured in triplicate were included. Second derivative spectra calculated according to the Savitzky–Golay algorithm with a filter length of 25 points and fitted with a second-degree polynomial were used in the PLS-model. The *X*-variables were mean centred and scaled to unit variance (1/S.D.) while the *Y*-variable, the content of the active ingredient, was only mean centred. Sixteen PLS-components were used in the model and the wavelength range 1124–2176 nm was included. To transfer this PLS-model from instrument NIR_Probe1 to instruments NIR_Probe2, NIR_RCA1 and NIR_RCA2, 37 samples were measured on all instruments. Of these samples, 2–20 samples were used for calibration transfer and the remaining 17 samples were used as an independent test set.

The other three calibration models that were transferred were all developed on the dispersive RCA-instrument denoted NIR_RCA1. The calibration models were named calibration R1A, R1B and R1C, respectively. The aim was to transfer all three calibrations to the second dispersive RCA-instrument (NIR_RCA2). Calibration R1C was also transferred to the Fourier transform instrument (NIR_FT). In the calibration sets used to create the PLS-models for calibrations R1A–C, 65–84 samples measured in duplicate were included. First derivative spectra, according to the Savitzky–Golay algorithm with a filter length of 11 points and fitted with a second-degree polynomial, were used in these three calibration models. The wavelength range 1100–2500 nm was used as *X*-variables for all three models. In calibrations R1A and R1B, the *X*-variables were scaled to unit variance (1/S.D.) while the *Y*-variable, the content of the active ingredient, was mean centred. In calibration R1C, both the *X*-variables and the *Y*-variable were mean centred. Six PLS-components were used for calibrations R1A and R1C while seven PLS-components were used for calibration R1B. To transfer these three calibrations from instrument NIR_RCA1 to instruments NIR_RCA2 and NIR_FT (only calibration R1C), 26–35 samples were measured on all instruments. Of these samples, 2–20 were used for calibration transfer the remaining 8, 10 or 15 samples were used as independent test sets.

The reference method used was liquid chromatography in all cases.

2.3. Calibration transfer techniques

Slope/bias correction and local centring were applied to all four NIR calibration models for the active ingredient and all slave instruments, while PDS was applied to two of the calibration models, R1B and R1C, transferred from NIR_RCA1 to NIR_RCA2 and from NIR_RCA1 to NIR_FT, respectively.

2.3.1. Slope/bias correction and local centring

Slope/bias correction operates only on *Y*-values and spectra are therefore never corrected. Orthogonal least squares was used to compute the slope and bias correction factors as suggested by Bouveresse et al. [7] such that:

$$Y_{\text{corr}}^{(s)} = \text{bias} + \text{slope} \times Y^{(s)}$$

Local centring means in this case that the spectra are centred by using an average spectrum for each instrument, i.e. spectra from instrument NIR_RCA1 are centred by using a mean spectrum from spectra collected using instrument NIR_RCA1 and spectra from instrument NIR_RCA2 are centred by using a mean spectrum from spectra collected using instrument NIR_RCA2 [10].

To transfer the calibrations using slope/bias correction or local centring, 18 or 20 of the 26–37 samples measured on both the master and the slave instruments were selected. The 18–20 samples were then sorted with regard to their content of the active ingredient. The number of samples selected for slope and bias correction and local centring was varied between 2 and 18–20. The less samples used for the correction, the less the samples differed in the content of the active ingredient. Hence, when using only two samples to transfer the calibrations these two were the ones in the middle of the concentration range and they differed nothing or almost nothing in the content of the active ingredient (Fig. 1). The transfer samples were selected so that the variation in the amount of the active ingredient was as large and as even as possible. This way of selecting the transfer samples was chosen to investigate how important the variation in the content of the active ingredient is for the performance of the various calibration transfer techniques.

2.3.2. Piecewise direct standardisation

In the piecewise direct standardisation technique, spectral intensities at a certain wavelength on the master instrument are related to a spectral window containing the intensities at the same wavelength and a few neighbouring wavelengths on the slave instrument. Multivariate regression models are built between all wavelengths on the master instrument and the corresponding moving spectral window on the slave instrument. This gives a number of regression coefficients that are placed in a so-called transformation matrix that can then be used for new predictions of spectra measured on the slave instrument.

The literature gives no guidance for the determination of the required number of transfer samples but gives a couple of suggestions of in what way these samples should be selected. In this study, two ways of selecting the spectra were tested. The original algorithm for selecting samples, prior to PDS, proposed in the literature [5,13] was used. It is based on selecting samples with a high leverage, i.e. samples with a large influence on the calibration model, and the algorithm can be found in *PLS_Toolbox 2.1* [14]. The other way of selecting the spectra was the algorithm proposed by Kennard and Stone [13,15]. This algorithm effectively spreads the selected samples over the experimental domain.

The effect of different numbers of transfer spectra, different pre-processing of spectra, different numbers of channels in the moving spectral window and different values of the tolerance used in forming the local regression models used in the PDS, was examined for the two different ways of selecting samples.

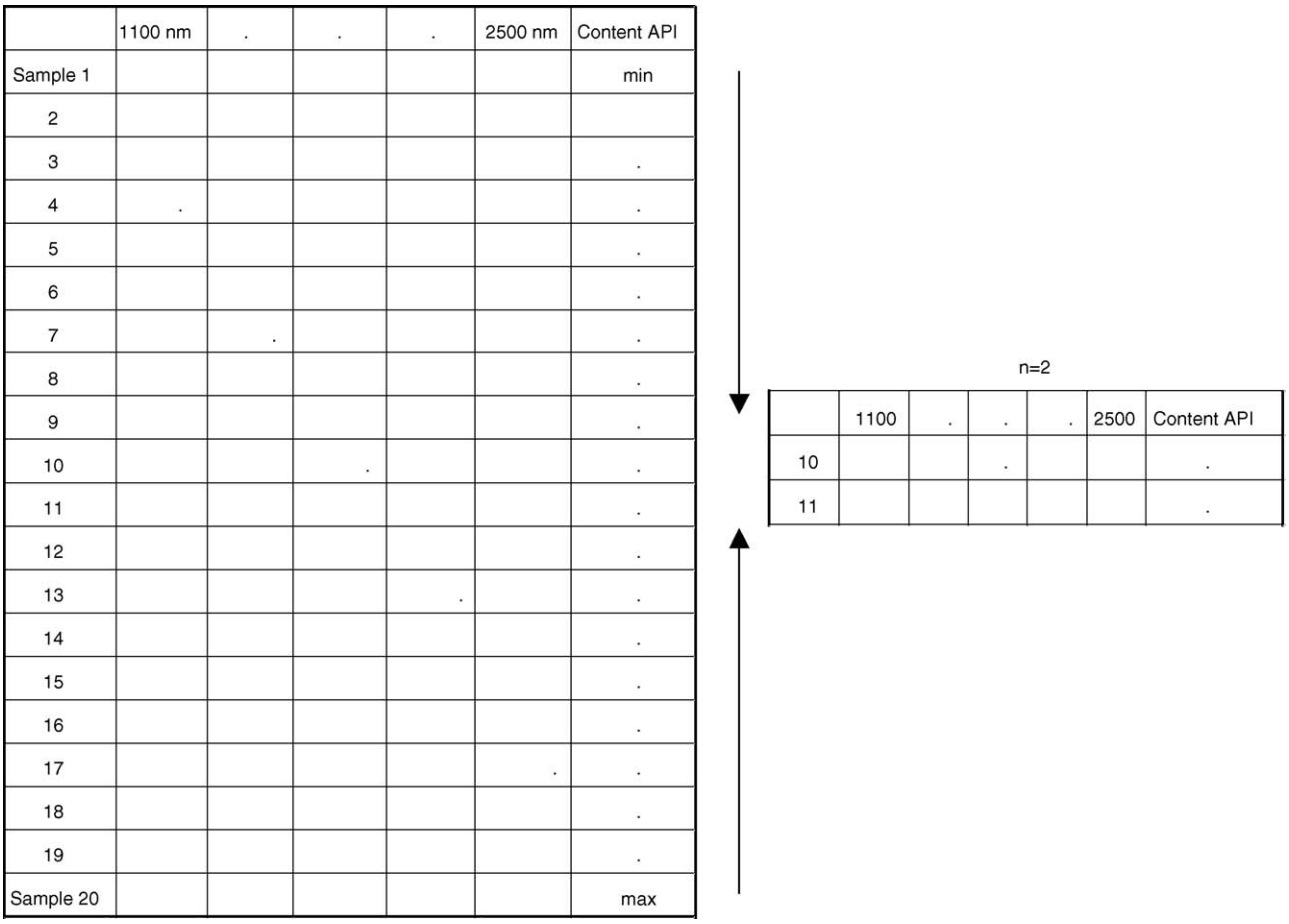


Fig. 1. Selection of samples for the calibration transfer. Twenty samples were selected as transferring samples. They were sorted with respect to their content of the active ingredient predicted using the master instrument. Between 2 and 20 samples were used for the transfer. The fewer samples that were used for the calibration transfer, the less they differed in their content of active ingredient.

The tolerance is equal to the minimum relative size of singular values to include in each model, and therefore the number of components used in each model. The lower the tolerance is set, the more components are used in the local regression models. The selected number of spectra was varied between 2 and 10, and the effect of pre-processing the spectra with first derivative (filter length 11 points, second-degree polynomial) was tested. The number of channels in the moving spectral window was varied between 3 and 9 and the tolerance used in forming the local PDS models was varied between 0.01 and 1.0.

3. Evaluation of results

The results of the transfer methods were evaluated by studying the values of root mean square error of prediction (RMSEP) and mean bias for the corrected predictions using the slave instruments versus the reference method (LC) for all measured samples ($n = 26$–37) and for an independent test set ($n = 8$–17). The Y residuals between the NIR predictions and the values achieved using the reference method were also studied for each sample. The calibration-transfer method resulting in the lowest values of RMSEP and bias and the lowest maximum absolute Y residual was considered to be the best one. The aim was to achieve as low values of RMSEP, bias and maximum absolute Y residual for the slave instruments as for the master instrument. However, the highest acceptable RMSEP was set to 2%, the highest acceptable bias was set to 0.9% and the highest maximum absolute Y residual was set to 4%, as related to the nominal content of the active ingredient in the various formulations. At these values of RMSEP and maximum Y absolute residual, the investigated methods were found to be fit for purpose.

4. Results

4.1. Spectroscopic characterization

Two examples of how different a spectrum from the same sample is depending on which instrument it is measured with is shown in Figs. 2 and 3. Spectra from the same sample in calibration P1, measured on different instruments, are shown in Fig. 2. The differences between the spectra are not due to differences in the wavelength scale, as we have previously shown [12]. Compared to spectra from the instruments with RCA-units, the instruments equipped with a probe have more noise above 2300 nm in the spectra as can be seen in Fig. 2. In the region below 2200 nm, which was used in the calibration, there are not only offset differ-

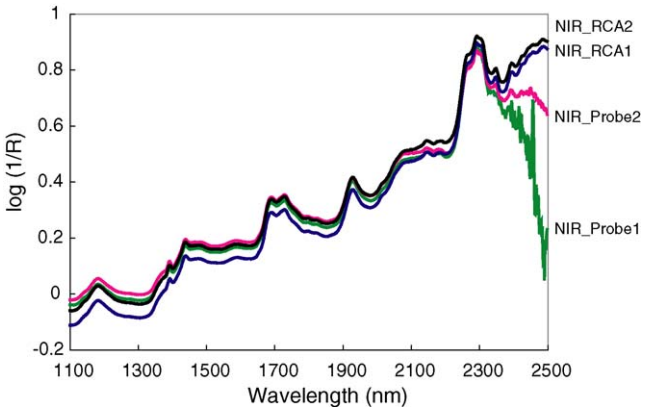


Fig. 2. Spectra from the same sample measured with four different instruments.

ences. The differences in the region between 2070 and 2200 nm are mainly due to the decreasing signal-to-noise ratio when using optical fibres in the upper NIR-wavelength range. The difference in this spectral region is more obvious comparing the RCA and fibre probe instruments than comparing the two probe instruments with each other. The main difference between the two probe instruments is in the 2070–2200 nm region and can be assigned to the different fibre probes.

Fig. 3 shows spectra from the same sample, included in calibration R1C, but measured with an RCA and FT-instrument, respectively. As in Fig. 2, the difference between the spectra is not related to the wavelength scale and is not only a difference in offset. The sharper peak at 1400 nm in the spectrum achieved using the Bomem FT-instrument as compared to the spectrum achieved using the FOSS dispersive instrument (Fig. 3), is due to the Bomem instrument's resolution (3 nm at 1400 nm). The bandwidth for the FOSS instrument is 9–10 nm. Due to the FT-instruments higher resolution an additional peak can be seen at 1680 nm. In the spectral region from 2100 to 2500 nm, there seems to be a difference in the detector response, i.e. the dynamics of the detector. The reason for this is that the RCA-instrument uses a PbS-detector while the FT-instrument uses an InAs-detector.

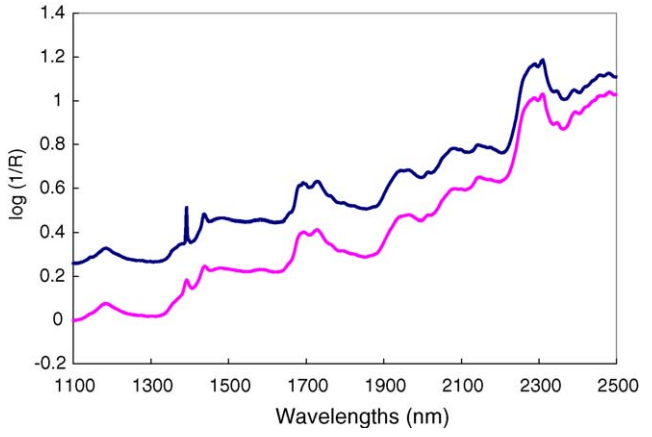


Fig. 3. Spectra from the same sample measured with two instruments, no offset correction is applied in the figure. The Bomem FT-instrument (NIR_FT) yielded the blue upper spectrum and the dispersive FOSS RCA-instrument (RCA1) yielded the pink lower spectrum.

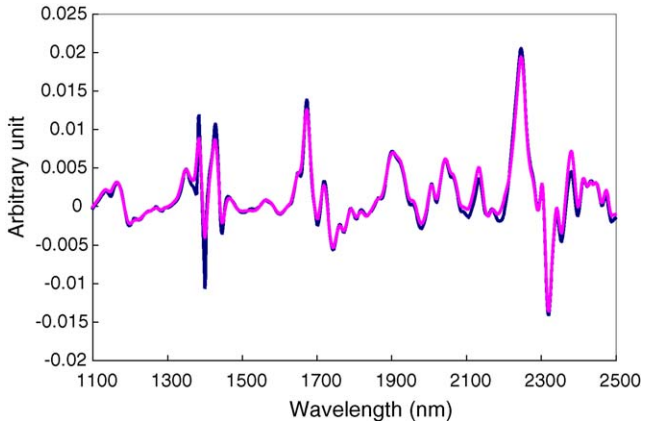


Fig. 4. The spectra shown in Fig. 3 pre-processed with a first derivative. The blue line is a pre-processed spectrum from the Bomem FT-instrument (NIR_FT) and the pink spectrum is a pre-processed spectrum from the dispersive FOSS RCA-instrument (RCA1).

The differences between spectra are less after pre-processing of the spectra with first derivatives (Fig. 4), but the differences discussed above are still present, and will cause significant deviations in the predictions if they are not compensated for with some kind of calibration transfer technique.

4.2. Transfer of calibration P1 from NIR_Probe1 to NIR_Probe2, NIR_RCA1 and NIR_RCA2

To transfer calibration P1, 37 samples were measured on all instruments. In Fig. 5, the correlations between the predicted content of the active ingredient in all samples ($n = 37$) measured using instrument NIR_Probe1, NIR_Probe2, NIR_RCA1 and NIR_RCA2, respectively, versus the reference method are shown. The values of R (correlation coefficient), RMSEP, bias, slope (the slope for the linear fit) and the intercept are shown for the correlations in the figures. The values of RMSEP and bias for the predictions achieved using the master instrument (NIR_Probe1) versus the reference method were very much the same as was achieved using the slave instruments NIR_Probe2 and NIR_RCA1. Also the maximum absolute Y residuals were about the same for the predictions achieved using the three instruments (Table 3). All measures (RMSEP, bias and absolute residual) were well below the values set as acceptable. Both slope/bias correction and local centring using 2–20 samples were tested to transfer calibration P1 to NIR_Probe2 and NIR_RCA1 anyway. In this case, calibration transfer correction did not improve the predictions and we can conclude that no calibration transfer is needed to transfer calibration P1 from instrument NIR_Probe1 to NIR_Probe2 and NIR_RCA1 (Table 3).

A calibration transfer was, however, needed to transfer calibration P1 to instrument NIR_RCA2 (Fig. 5). The results for slope/bias correction and local centring are shown in Fig. 6. Note that no result is shown for slope/bias correction using only two samples, as these two samples did not differ in API. The values of RMSEP, bias and maximum absolute Y residuals are shown in Table 3.

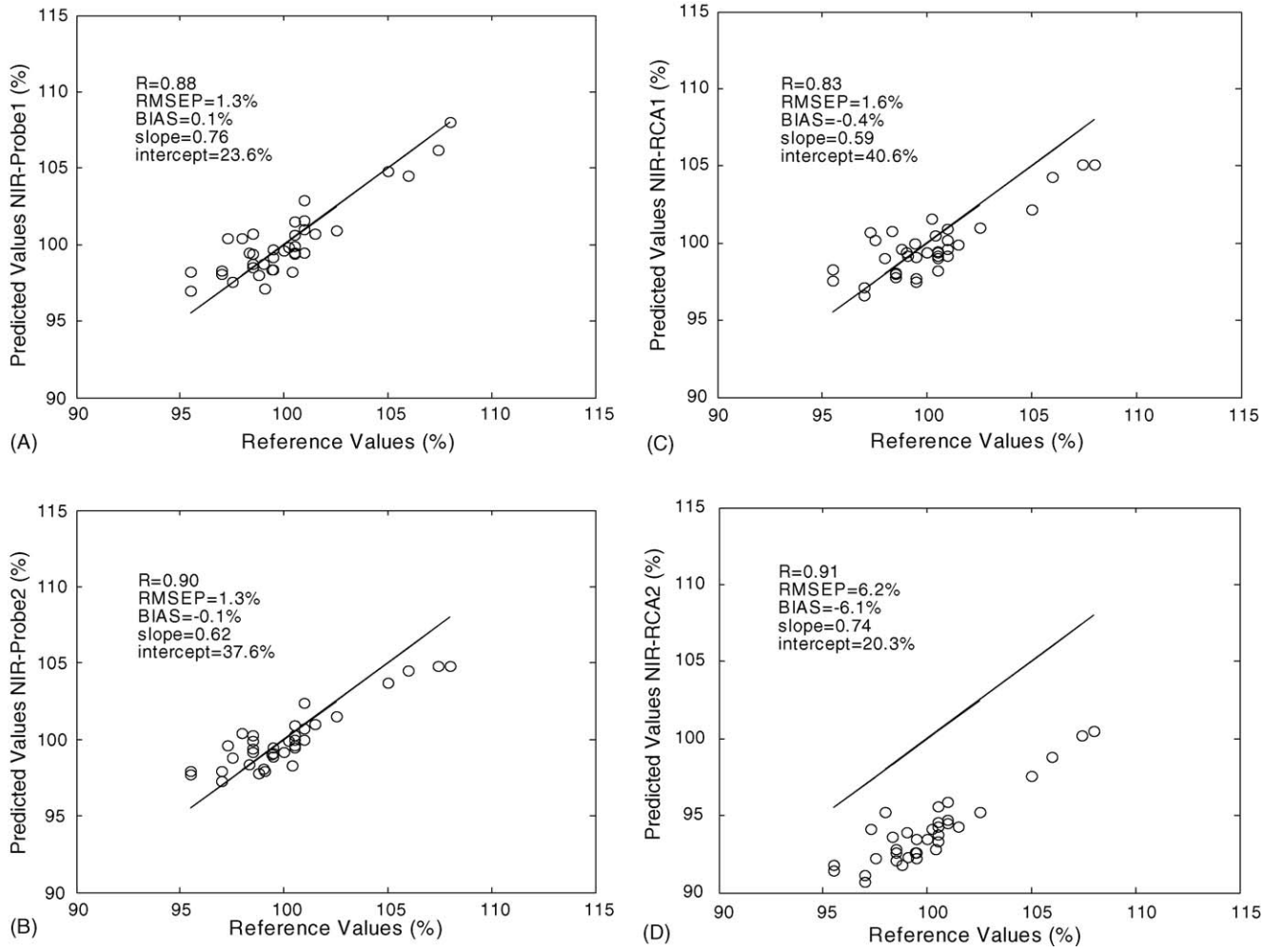


Fig. 5. Correlation between predictions using calibration P1 of the content of the active ingredient in all samples ($n = 37$) when measured using the four NIR instruments (A) NIR_Probe1, (B) NIR_Probe2, (C) NIR_RCA1 and (D) NIR_RCA2 vs. the reference values. All values are expressed as percentage of the nominal content.

4.3. Transfer of calibration R1A, B and C from NIR_RCA1 to NIR_RCA2 using slope and bias correction and local centring

To transfer calibration R1A, B and C, 30–35 samples were measured on the master instrument (NIR_RCA1) and on the slave instrument (NIR_RCA2). The values of RMSEP and bias for the predictions achieved using the slave instrument (NIR_RCA2) were much higher than for the master instrument (NIR_RCA1) and a calibration transfer was thus needed for all three calibrations

The effect of slope/bias correction using 2–20 samples for the transfer is shown in Fig. 7. The effect of local centring using the same 2–20 samples is shown in Fig. 8.

4.4. Transfer of calibration R1B from NIR_RCA1 to NIR_RCA2 using PDS

For PDS, the results showed that using first derivate preprocessed spectra both in the PDS and in the selection of transfer samples seems to remove more instrumental differences than using unprocessed spectra. When the selection of spectra is based on high leverage, the number of spectra should be $n = 4$–6 and the number of channels in the spectral window somewhere between 3 and 9. The tolerance should be set to 0.01–0.1. In the other case, when the spectra selection is based on the Kennard and Stone algorithm, the number of spectra should be $n = 6$–10 with the same number of channels (3–9). The tolerance could in this case be smaller and set to 0.00001–0.1. Hence, the different variables should be varied to optimise the calibration transfer for different numbers of selected spectra. In Fig. 9, the predictions of PDS corrected spectra from instrument NIR_RCA2 are plotted versus the reference values. In Fig. 9A, the transfer was optimised for sample selection based on high leverage, and in Fig. 9B for sample selection based on the Kennard and Stone algorithm.

4.5. Transfer of calibration R1C from NIR_RCA1 to NIR_FT

To transfer calibration R1C from the dispersive master instrument (NIR_RCA1) to the Fourier transform instrument NIR_FT, 26 samples were measured on both instruments. The values of

Table 3
Predictions in terms of RMSEP, bias and maximum absolute residual from calibration P1 applied to spectra measured on the master instrument (NIR_Probe1), instrument NIR_Probe2, NIR_RCA1 or NIR_RCA2 vs. the reference method

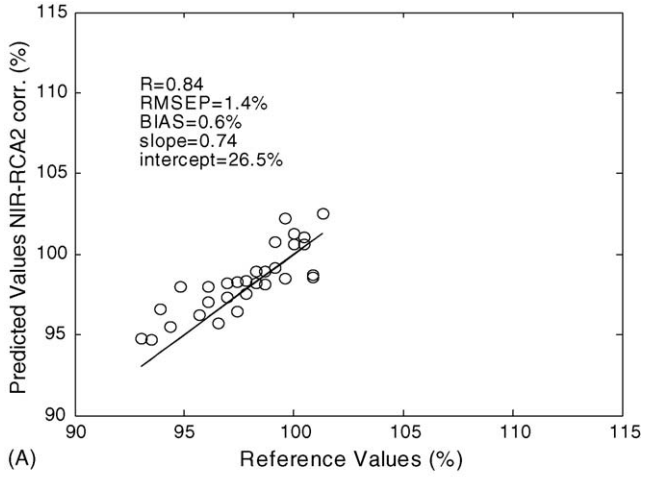
Transfer method	NIR_Probe1			NIR_Probe2			NIR_RCA1			NIR_RCA2		
	RMSEP (%)	Bias (%)	Max residual (%)	RMSEP (%)	Bias (%)	Max residual (%)	RMSEP (%)	Bias (%)	Max residual (%)	RMSEP (%)	Bias (%)	Max residual (%)
No correction	1.3 (n = 37)	0.1 (n = 37)	3.1 (n = 37)	1.3 (n = 37)	−0.1 (n = 37)	3.2 (n = 37)	1.6 (n = 37)	−0.4 (n = 37)	3.4 (n = 37)	6.2 (n = 37)	−6.1 (n = 37)	7.6 (n = 37)
	1.4 (n = 17)	−0.2 (n = 17)		1.3 (n = 17)	−0.4 (n = 17)		1.5 (n = 17)	−0.4 (n = 17)		6.4 (n = 17)	−6.3 (n = 17)	
Slope/bias (n = 20)	n.a.	n.a.	n.a.	1.2 (n = 17)	−0.3 (n = 17)	–	1.8 (n = 17)	0.3 (n = 17)	–	1.1 (n = 37)	0.1 (n = 37)	3.5 (n = 37)
										1.0 (n = 17)	−0.1 (n = 17)	
Local centring (n = 4)	n.a.	n.a.	n.a.	–	–	–	1.6 (n = 37)	0.0 (n = 37)	3.8 (n = 37)	1.2 (n = 37)	0.1 (n = 37)	3.3 (n = 37)
										1.0 (n = 17)	−0.1 (n = 17)	

Values of RMSEP and bias are shown for all samples (n = 37) and for the independent test set (n = 17, samples not among the 20 selected to be used as transferring samples). The number of selected samples used for the best slope and bias correction or local centring is shown.

Fig. 6. Values of RMSEP for NIR predictions achieved using calibration P1 applied to spectra from instrument NIR_RCA2 vs. reference values for an independent test set (n = 17) after slope and bias correction (line with circles) or local centring (line with squares) using transfer samples with different variations in the content of the active ingredient. The less the transfer samples varied in content, the fewer samples were used for the transfer. The straight dotted line shows the highest acceptable RMSEP (2%) and the dashed straight line shows the RMSEP (1.3%) achieved using the master instrument (NIR_Probe1).

Fig. 7. Values of RMSEP for NIR predictions achieved using calibration R1A (circles), R1B (squares) or R1C (triangles) applied to spectra from instrument NIR_RCA2 vs. reference values for the independent test sets (n = 10–15) after slope and bias correction using transfer samples with different variations in the content of the active ingredient. The less the transfer samples vary in content the fewer samples have been used for the transfer. The straight dotted line shows the highest acceptable RMSEP (2%).

Fig. 8. Values of RMSEP for NIR predictions achieved using calibration R1A (circles), R1B (squares) or R1C (triangles) applied to spectra from instrument NIR_RCA2 vs. reference values for the independent test sets (n = 10–15) after local centring using transfer samples with different variations in the content of the active ingredient. The less the transfers samples vary in content the fewer samples have been used for the transfer. The straight line shows the highest acceptable RMSEP (2%).



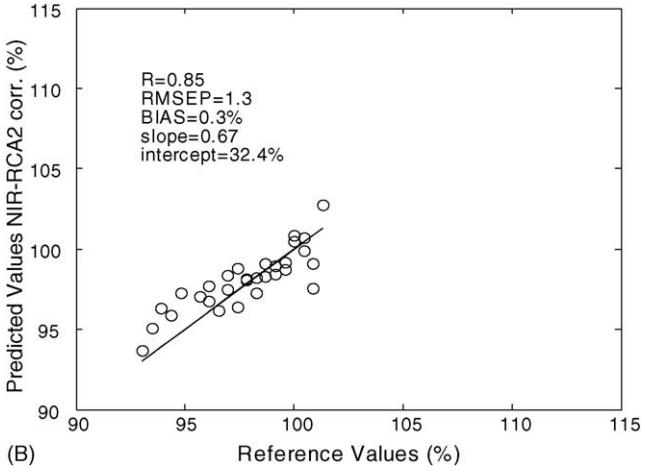


Fig. 9. Prediction of PDS corrected spectra for 30 samples measured using NIR_RCA2 plotted vs. the reference values (LC). In (A), spectra were corrected using $n = 6$ samples, three channels were used in the transform window and the tolerance was set to 0.01. The selection of spectra was based on high leverage. In (B), the selection of spectra was based on KS instead and spectra are corrected using $n = 8$ samples, nine channels are used in the transform window and the tolerance is set to 0.001.

RMSEP and bias for the predictions achieved using the slave instrument (NIR_FT) were much higher than for the master instrument (NIR_RCA1), thus a calibration transfer was needed.

Slope/bias correction, local centring and PDS were tested to transfer calibration R1C to instrument NIR_FT. The results of slope/bias correction and local centring using 2–20 samples are shown in Fig. 10. The average absorption differences, before and after local centring, between spectra for the 26 samples measured on instrument NIR_RCA1 and NIR_FT are shown in Fig. 11. Before the local centring, there were large differences between the spectra collected on the two instruments. These differences were almost removed after local centring. The values of RMSEP, bias and maximum absolute *Y* residual for the master instrument and for the slave instrument after the best slope/bias correction and local centring are shown in Table 4.

The PDS algorithm parameters were optimised differently for different selection methods and numbers of transfer spectra to get the best transfer result between the two instruments. For

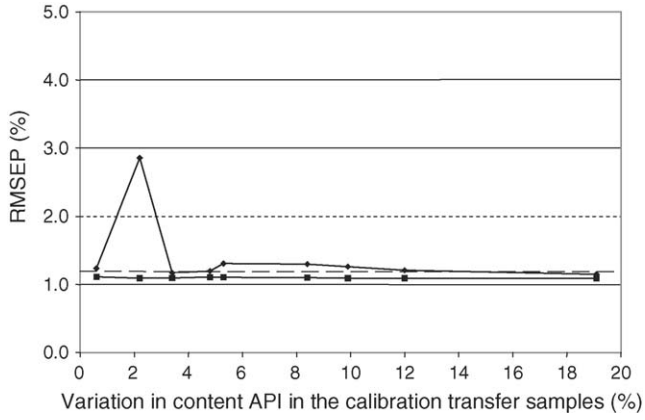


Fig. 10. Values of RMSEP for NIR predictions achieved using calibration R1C applied to spectra from instrument NIR_FT vs. reference values for an independent test set ($n = 8$) after slope and bias correction (line with rhombus) or local centring (line with squares) using transfer samples with different variations in the content of the active ingredient. The less the transfers samples vary in content, the fewer samples have been used for the transfer. The straight rugged line shows the highest acceptable RMSEP (2%) and the dashed line shows the RMSEP (1.2%) achieved using the master instrument (NIR_RCA1).

the PDS transfers, the results showed that using first derivate pre-processed spectra, both in the PDS and in the selection of transfer samples, allows the tolerance to be smaller. It could also be seen that the number of channels in the transform window often should be set to 3 or 5. When selecting samples based on the Kennard and Stone algorithm, the calibration transfer was more successful than using the original algorithm based on high leverage.

In Fig. 12A and B, the values of RMSEP for predictions using calibration R1C applied on PDS corrected spectra from instrument NIR_FT versus the reference values are shown. In Fig. 12C, the average absorbance difference between spectra measured on instrument NIR_RCA1 and NIR_FT can be seen. The figure shows, before and after correction with PDS, when six transfer

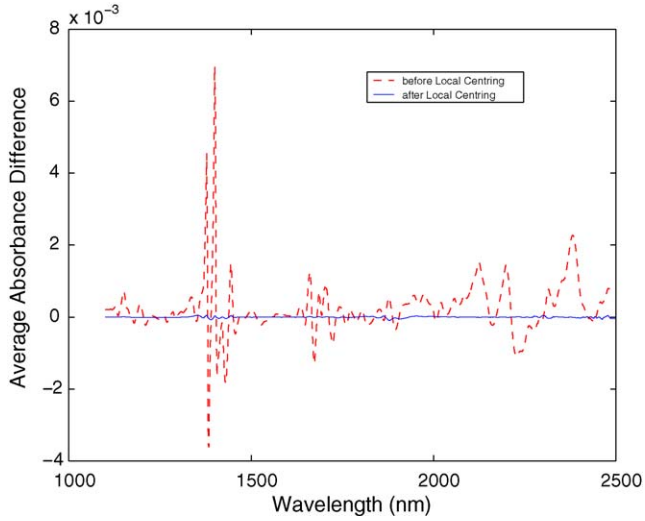


Fig. 11. The average absorbance difference for the 26 samples spectra measured on the master instrument (NIR_RCA1) and instrument NIR_FT. The figure shows before and after local centring using four samples.

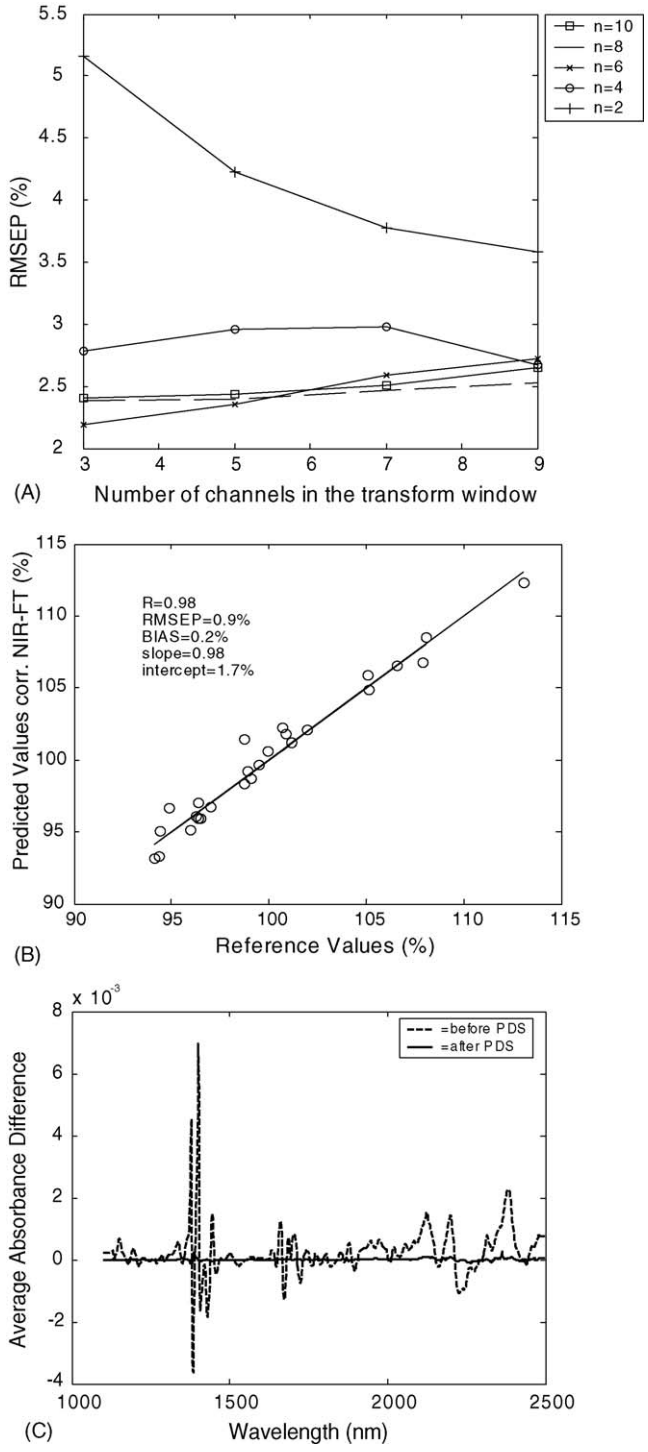


Fig. 12. Predictions achieved using calibration R1C applied on PDS corrected spectra for 26 samples measured using instrument NIR_FT plotted vs. the reference values (LC). In (A), spectra are corrected using $n = 2$–10 spectra, 3–9 channels are used in the transform window and the tolerance was set to 0.001. The selection of spectra was based on the Kennard and Stone (KS) algorithm. In (B), spectra are corrected using $n = 6$ spectra, three channels are used in the transform window and the tolerance was set to 0.001. The selection of spectra was based on the KS algorithm. In (C), the average absorbance difference between spectra measured on the master instrument (NIR_RCA1) and instrument NIR_FT can be seen. The figure shows before and after correction with PDS when six transfer spectra selected using the KS algorithm are used. The tolerance is set to 0.001 and the number of channels in the transform window is three.

samples selected using the Kennard and Stone algorithm was used. The tolerance was set to 0.001 and the number of channels in the transform window was three. Before the PDS correction, there were large differences between the spectra collected on the two instruments. These differences were almost removed after PDS correction. The values of RMSEP, bias and maximum absolute *Y* residual for the master instrument and for the slave instrument after the PDS correction are shown in Table 4.

5. Discussion

All three transfer methods (slope and bias correction, local centring and PDS) worked well for transferring of the present NIR calibrations for solid, pharmaceutical formulations. The methods worked equally well regardless of small or large differences between spectra from the master instrument as compared to spectra from slave instruments due to differences in hardware and sample presentation. With slope and bias correction, samples that varied at least 2.5% in the content of the active ingredient were needed to achieve acceptable values of RMSEP (Figs. 6 and 7). The best results were, however, achieved when the samples used for the correction varied 5–8% in the content of the active ingredient. For the slope and bias correction it is favourable to use several samples, at least 10. Local centring does not need samples that vary in the content of the active ingredient to give acceptable values of RMSEP (Figs. 6 and 8). The values of RMSEP achieved after local centring for all four calibrations (P1, R1A, R1B and R1C) were slightly lower or at the same level as those achieved using the master instrument, except for calibration R1C. For R1C, we could not get as low RMSEP as for the master instrument. But the values of RMSEP, bias and maximum absolute *Y* residuals were still well below the values set to be acceptable. No significant difference could be seen ($p > 0.05$) [16] between the predictions achieved using the master instrument and the ones achieved using the slave instrument after the best local centring.

Optimisation is needed for calibration transfers using PDS. The number of channels in the transform window, the number of transfer spectra, different pre-processing of spectra, different tolerance in the PDS for different ways of selecting samples, all these parameters must be optimised. No significant difference could be shown between the predictions after PDS correction compared to slope/bias correction and local centring ($p > 0.05$). When comparing Figs. 11 and 12C, it seemed that the average absorbance difference between spectra measured on the master instrument and on the slave instrument NIR_FT was slightly smaller after local centring than after PDS. But PDS would probably have been the best calibration transfer technique if there had been differences in wavelength scale between the instruments.

Local centring is the preferred transfer method considering the simplicity of the method compared to PDS. Local centring is also preferred to slope/bias correction as a lower number of transfer samples is needed and their variation in the content of the active ingredient can be lower than what is needed to achieve a good calibration transfer using slope/bias correction. Another advantage of local centring as compared to slope/bias is that the measures we use to detect outliers (distance to model

Table 4
Predictions in terms of RMSEP, bias and maximum absolute residual when calibration R1C is applied to spectra measured on the master (NIR_RCA1) and slave instruments (NIR_FT) vs. the reference method

Transfer method	NIR_RCA1					NIR_FT				
	RMSEP (%)		Bias (%)		Max residual (%)	RMSEP (%)		Bias (%)		Max residual (%)
	$n=26$	$n=8$	$n=26$	$n=8$	$n=26$	$n=26$	$n=8$	$n=26$	$n=8$	$n=26$
No correction	1.0	1.2	0.0	0.4	3.9	8.5	–	8.4	–	10.3
Slope/bias ($n=16$)	n.a.	n.a.	n.a.	n.a.	n.a.	1.2	1.7	0.0	0.5	2.9
Local centring ($n=4$)	n.a.	n.a.	n.a.	n.a.	n.a.	1.1	1.6	0.0	0.6	3.0
PDS[a]	n.a.	n.a.	n.a.	n.a.	n.a.	0.9	1.3	0.2	−0.5	2.7

Values of RMSEP and bias are shown for all samples ($n=26$) and for the independent test set ($n=8$, samples not among the 18 selected to be used as transferring samples). The number of selected samples, or for PDS individual spectra, used for the best slope and bias correction, the best local centring and the best PDS transfer are shown.

[a] Spectra are corrected using $n=6$ spectra, three channels are used in the transform window and the tolerance was set to 0.001. The selection of spectra was based on the Kennard and Stone algorithm.

(DmodX) [17] and Hotelling's T^2 [18]) are put on the same scale. Measuring samples on a different NIR instrument than was used to develop the PLS-model will in most cases yield higher values of DModX and Hotelling's T^2 but there are also occasions where the values will be lower.

6. Conclusions

Slope/bias correction, local centring or PDS can be used to transfer the four studied NIR assays, for solid pharmaceutical formulations, from the dispersive master instrument to another dispersive instrument of the same or of a different configuration or even to a Fourier transform instrument. We consider local centring to be the calibration transfer method of choice since it works without variation of the active content, needs no optimisation of parameters as in PDS, allows the use of similar outlier limits for the transferred calibration, and it is simple to perform either as a correction before the analysis or built into the software used. It is a great advantage that no variation in API for calibration transfer samples is needed as samples that vary in API are hard to get in a normal production environment.

Acknowledgment

Magnus Fransson is thankfully acknowledged for preparing high-resolution graphic files.

References

[1] O.E. de Noord, Chemom. Intell. Lab. Syst. 25 (1994) 85–97.
[2] E. Bouveresse, D.L. Massart, Vibr. Spectrosc. 11 (1996) 3–15.
[3] T. Fearn, J. Near Infrared Spectrosc. 9 (2001) 229–244.
[4] R.N. Feudale, N.A. Woody, H. Tan, et al., Chemom. Intell. Lab. Syst. 64 (2002) 181–192.
[5] Y. Wang, D.J. Veltkamp, B.R. Kowalski, Anal. Chem. 63 (1991) 2750–2756.
[6] J.S. Shenk, M.O. Westerhaus, US Pat No. 4,866,644 (1989).
[7] E. Bouveresse, C. Hartmann, D.L. Massart, I.R. Last, K.A. Prebble, Anal. Chem. 68 (1996) 982–990.
[8] B. Walczak, E. Bouveresse, D.L. Massart, Chemom. Intell. Lab. Syst. 36 (1997) 41–51.
[9] S. Wold, H. Antti, F. Lindgren, J. Öhman, Chemom. Intell. Lab. Syst. 44 (1998) 175–185.
[10] J. Sjöblom, O. Svensson, M. Josefson, H. Kullberg, S. Wold, Chemom. Intell. Lab. Syst. 44 (1998) 229–244.
[11] S. Saranwong, S. Kawano, J. Near Infrared Spectrosc. 12 (2004) 359–365.
[12] H. Leion, S. Folestad, M. Josefson, A. Sparén, J. Pharm. Biomed. Anal. 37 (2005) 47–55.
[13] E. Bouveresse, D.L. Massart, Chemom. Intell. Lab. Syst. 32 (1996) 201–213.
[14] B.M. Wise, N.B. Gallagher, PLS_Toolbox v 2.1, Eigenvector Research, 2000.
[15] R.W. Kennard, L.A. Stone, Technometrics 11 (1969) 137–149.
[16] J.C. Miller, J.N. Miller, Statistics and Chemometrics for Analytical Chemistry, fourth ed., Pearson Education Limited, Harlow, 2000.
[17] User Guide and Tutorial, Simca 8.0, Umetrics, Inc., 1999.
[18] H. Hotelling, Ann. Math. Stat. 2 (1931) 360–378.