Insights into information contained in multiplicative scatter correction parameters and the potential for estimating particle size from these parameters

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ABSTRACT

Empirical preprocessing methods such as multiplicative scatter correction (MSC) and extended multiplicative scatter correction (EMSC) are widely used to remove light scattering effects from spectra of samples containing particulate species. When these methods are used, the parameters that are applied for correcting the spectra are normally discarded. If the scatter correction method is effective, these parameters should contain information regarding the particulate species since it is this component which contributes to the light scattering effects. This study had two objectives. The first objective was to examine the nature and extent of information contained in scatter correction parameters. The second objective is to examine whether this information can be effectively extracted by proposing a method to obtain particularly, the mean particle diameter from the scatter correction parameters. The approach used for this investigation is to examine the scatter correction parameters in terms of the information regarding particle size and particle concentration by using a dataset in which particle size and particle concentration vary significantly. It was found that the MSC parameters contained significant information regarding particle size and concentration. A two-step method to obtain simultaneously the particle concentration and particle diameter was proposed and tested using a 2-component and 4-component data set. It was found that the approach which uses the MSC parameters gave a better estimate of the particle diameter compared to using Partial Least Squares (PLS) regression for the 2-component data. For the 4 component data it was found that PLS regression gave better results but further examination indicated this was due to chance correlations of the particle diameter with the two of the absorbing species in the mixture.

1. Introduction

Multivariate calibration methods such as Partial Least Squares (PLS) regression have been widely used to build calibration models for predicting the concentrations of chemical components from near-infrared (NIR) spectra. When samples containing particles are encountered, multiple light scattering effects introduce nonlinearities leading to degradation in model performance. Several empirical preprocessing methods such as multiplicative scatter correction (MSC), standard normal variate (SNV), extended multiplicative scatter correction (EMSC), orthogonal signal correction (OSC), and optical path length estimation and correction (OPLEC) have been used to mitigate light scattering effects.[1-6] When dealing with particulate systems, it is generally assumed that the information removed from the measured spectra by the application of these empirical methods is essentially the manifestation of the underlying physics of light scattering without significant loss of chemical information, thus improving the performance of the multivariate regression models in estimating chemical information from the corrected spectra.

When these methods are used, the parameters that are applied for correcting the spectra are normally discarded since they are supposed to contain only physical information. If the scatter correction method is effective, the scatter correction parameters would be expected to contain information regarding the particulate species since it is this component which contributes to the light scattering effects. If this information can be extracted then it could provide valuable extra information (particle size) in addition to estimates of concentrations which are obtained from the calibration models built on the scatter-corrected spectra.

Several studies can be found in the literature where scatter correction techniques are applied and compared in terms of the improvement in performance of models built using the corrected spectra. However, the performances of the empirical methods appear to be dependent on the system studied with no single empirical scatter correction method consistently outperforming others across a number of different types of datasets. Among the empirical methods, the more recently developed OPLEC method has been promising,[6, 7] though it has not yet been applied widely enough to conclude that the method is indeed consistently superior to other available methods. A study based on simulations using a rigorous light propagation model indicated that most of the common scatter correction methods led to similar model performances.[8] In addition, this study also indicated that the effectiveness of a particular scatter correction technique was also dependent on measurement configuration. To-date however, to our knowledge, there have been no in-depth studies that have examined the information contained in the scatter correction parameters themselves. Such a study will be useful for understanding the nature and characteristics of information contained in the parameters of a particular scatter correction method. This could help in identifying situations where they perform the best and could potentially help in modifying the methods to produce more effective scatter correction techniques.

The implicit assumption when applying scatter correction methods is that light scattering effects manifesting as an additive or multiplicative or more complex (e.g. wavelength dependent) effects in the measured spectra are removed. However, there are other non-chemical effects which can lead to similar manifestations in the spectra as the assumed effect of light scattering (e.g. instrument drift). In other words, the corrections are not necessarily specific to scattering. Hence the terms Multiplicative Signal Correction and Extended Multiplicative Signal correction can sometimes be found in the literature where “signal” is used instead of “scatter” to denote that the techniques are more general in terms of the non-chemical information removed by them.[5] Similarly, the SNV method is clearly a general method which has also been used to correct light scattering effects.

In any dataset consisting of spectroscopic measurements of particulate systems, we can expect the non-chemical variations to be a combination of effects with the light scattering effects usually being the most dominant. There are four possibilities why one scatter correction technique might work better than others: (1) The method removes the most amount of variation due to light scattering compared to others; (2) The method removes the most amount of variation due to all non-chemical effects present in the measurements; (3) The method linearizes the measurements most effectively compared to other methods, (4) The method removes the least amount of relevant chemical information; and (5) The method is the most effective in terms of a combination of the previous four aspects. Therefore the most effective "scatter correction" method will differ from one system to another depending on the dominant type of non-chemical variations in the measurements that form the datasets.

This study had two objectives. The first objective was to examine the nature and extent of information contained in scatter correction parameters. The second objective is to examine whether this information can be effectively extracted by proposing a method to obtain particularly the particle size from the scatter correction parameters. The approach used for this investigation is to examine the scatter correction parameters in terms of the information regarding particle size and particle concentration by using a dataset in which particle size and particle concentration vary significantly and where the values of these parameters have been accurately measured. Since particle concentration and size are the two sample parameters that affect the extent of light scattering by a sample, it follows that any effective correction step will contain information regarding these two sample parameters. Following this logic, if the scatter correction step is effective, then it should be possible to extract information regarding particle size and/or particle concentrations from the scatter correction parameters. This is investigated through an approach for building models to obtain particle size information using the scatter correction parameters. The investigation into the effectiveness of the scatter correction approach to specifically provide information regarding particle size was carried out using two models systems namely, a two component and a four component system both containing polystyrene latex particles as the scattering species.

2. Materials and Methods

2.1 Experimental dataset

The two datasets used in this study were obtained from previously published works.[9, 10] A brief description of the datasets is given here. Both datasets contain measurements taken using a Cary 5000 spectrometer equipped with an external diffuse reflectance accessory and 1 mm sample thickness was chosen. The first dataset is a polystyrene-water system that consists of a total of 35 samples with 5 particle diameters (d_p = 100, 200, 300, 430 and 500 nm) and 7 particle concentrations (y = 0.1, 0.5, 0.9, 1.23, 1.6, 1.95 and 2.3 in wt. %) for each particle size.[9] Spectra were collected using 0.4 sec as integrating time for a wavelength range of λ =1550 – 1850 nm with 4 nm interval, resulting in 75 discrete wavelengths per spectrum. The raw spectra were smoothed using Savitsky-Golay filter with window width of 9 and polynomial order of 3 to remove noise in the measurements.

The second dataset is a 4-components system that consists of water (H_2O), deuterium oxide (D_2O), ethanol (C_2H_5OH), and polystyrene particles.[10] The concentration of each component was varied so that the correlation between concentration of polystyrene particles and other components in the sample is negligible. In this dataset there are samples containing the same particle diameter and particle concentration while concentrations for other components vary. 5 particle diameters (d_p = 100, 200, 300, 430 and 500 nm) and 5 concentrations (y = 1, 2, 3, 4, and 5 in wt. %) were employed to form this dataset of 45 samples. Spectra were collected in the range of λ =1500 – 1880 nm with 2 nm intervals and 10 sec as the integrating time. The same smoothing conditions applied to the first dataset were also employed for this dataset before subjecting to scatter correction methods. Both datasets contained measurements from three different measurement configurations namely, total reflectance (Rd), total transmittance (Td) and collimated transmittance (Tc).

2.3 Estimation of particle size from MSC parameters

The first step in this approach is to establish the relationship between the MSC parameters and particle size (diameter) using the calibration dataset. In other words we develop models for expressing the additive (a) and multiplicative (b) term of MSC parameters as a function of particle diameter (d_p) and particle concentration (y). As will be seen in the next section, the MSC parameters are dependent on both particle diameter and concentration. Given these “direct” relations, we can then write inverse relations i.e. particle diameter as a function of particle concentration and MSC parameters a or b or both. This relationship can then be used to estimate the diameter of particles in a sample *i* given the concentration of particles and the MSC parameters a_i and b_i for that sample. Usually the actual particle concentration of a sample is also unknown. Therefore it has to be estimated. This can be done in the usual manner of building a calibration model for the concentration using PLS regression. Then in the inverse expression, the estimated particle concentration ($\hat{y}$) is used. The methodology is summarized by the flowchart shown in Fig. 1.

The methodology consists of two stages, the calibration model building stage (Stage 1 shown in black) where the models for estimating d_p and y are developed using the calibration dataset, and the prediction stage (Stage 2 shown in blue) to estimate particle diameter $\hat{d}_p$ and particle concentration $\hat{y}$ from spectra of unknown sample conditions. for a two component system is considered. In Stage 1, measured spectra ($\boldsymbol{x_{meas}}$) from a set of calibration samples of known y and d_p is subjected to an empirical scatter correction method such as MSC. The MSC equation is given by:

$$\boldsymbol{x}_{meas} = a + b\boldsymbol{x}_{ref} + \boldsymbol{e} \qquad (1)$$

where $\boldsymbol{x_{meas}}$ is the spectrum measured from the sample, and $\boldsymbol{x_{ref}}$ is a reference spectrum. The values of parameters a and b are estimated using ordinary least-squares regression of $\boldsymbol{x_{meas}}$ onto $\boldsymbol{x_{ref}}$. The error term, $\boldsymbol{e}$, contains the chemical information of the sample since it is the portion that is not explained by the physical variations (changes in baseline/slope). Note that the letters in bold indicate vectors. Once a and b are estimated, Eq. (1) can be rearranged as follows:

$$\boldsymbol{x}_{corr} = (\boldsymbol{x}_{meas} - a)/b = \boldsymbol{x}_{ref} + \boldsymbol{e}/b \qquad (2)$$

where $\boldsymbol{x_{corr}}$ is the spectrum corrected using MSC and should be as similar to $\boldsymbol{x_{ref}}$ as possible (in a least squares sense). This means that the difference between $\boldsymbol{x_{corr}}$ and $\boldsymbol{x_{ref}}$, i.e. $\boldsymbol{e}/b$, can be considered to be independent of the scattering effect. In this work, the reference spectrum for this example was taken to be the average spectrum of the whole calibration dataset.

Based on the functional forms identified through the analysis of the relationships between the MSC parameters and the particle diameter (d_p) and concentration (y) are obtained. For the two-component dataset, the expressions were (discussed in §3.1):

$$a = \xi_a + \left(\alpha_1 y + \alpha_2 y^2 + \alpha_3 y^3\right)\left(1 + \beta_1 d_p + \beta_2 d_p^2\right) \tag{3}$$

$$b = \xi_b + \kappa_1 y\left(1 + \eta_1 d_p + \eta_2 d_p^2\right) \tag{4}$$

where coefficients (α_i, β_j and η_k) were determined based on the best fit of y and d_p to the MSC parameters a and b. It is worth noting that the expressions may not be unique therefore care has to be taken to ensure that the coefficients used in the functional forms are significant.

Eqs. (3) and (4) can then be re-arranged so that d_p can be expressed as:

$$d_p = f(a,y) = \frac{1}{2\beta_2}\left\{-\beta_1 \pm \left[\beta_1^2 - 4\beta_2\left(1 - \frac{a - \xi_a}{\alpha_1 y + \alpha_2 y^2 + \alpha_3 y^3}\right)\right]^{1/2}\right\} \tag{5}$$

$$d_p = g(b,y) = \frac{1}{2\eta_2}\left\{-\eta_1 \pm \left[\eta_1^2 - 4\eta_2\left(1 - \frac{b - \xi_b}{\kappa_1 y}\right)\right]^{1/2}\right\} \tag{6}$$

It is also possible to obtain an expression for d_p that includes both parameters a, b and the measured and corrected spectrum, $\boldsymbol{x}_{meas}$ and $\boldsymbol{x}_{corr}$. The expression simultaneously makes use of particle size information contained in these parameters as well as that remaining in the corrected spectrum, thereby providing the possibility of better estimation of d_p owing to the augmented information contained in such an expression. In order to do this, we start with the re-arranging Eq.(2):

$$\boldsymbol{x}_{meas} = a + b\boldsymbol{x}_{corr} \tag{7}$$

Substituitng Eqs. (3) and (4) into Eq. (7), and carrying out algebraic manipulations an expression for d_p as a function of a, b, $\boldsymbol{x}_{corr}$, $\boldsymbol{x}_{meas}$ and y can be obtained. Maple version 13 (Waterloo Maple Inc.) was employed to solve for d_p to obtain the following expression:

$$d_p = h(a,\, b,\, y,\, \mathbf{x}_{meas},\, \mathbf{x}_{\mathbf{corr}})$$

$$= \frac{-1}{2y\left(\alpha_3\beta_2 y^2 + \alpha_2\beta_2 y + \alpha_1\beta_2 + \kappa_1\eta_2\mathbf{x}_{\mathbf{corr}}\right)} \left\{ \begin{array}{l} \left(\alpha_3\beta_1 y^3 + \alpha_2\beta_1 y^2 + \alpha_1\beta_1 y + \kappa_1\eta_1 y\mathbf{x}_{\mathbf{corr}}\right) \\ \pm \left[\begin{array}{l} \left(\alpha_3\beta_1 y^3 + \alpha_2\beta_1 y^2 + \alpha_1\beta_1 y + \kappa_1\eta_1 y\mathbf{x}_{\mathbf{corr}}\right)^2 \\ -4y\left(\alpha_3\beta_2 y^2 + \alpha_2\beta_2 y + \alpha_1\beta_2 + \kappa_1\eta_2\mathbf{x}_{\mathbf{corr}}\right) \\ \left(\begin{array}{l} \alpha_3 y^3 + \alpha_2 y^2 + \alpha_1 y + \xi_a + \xi_b\mathbf{x}_{\mathbf{corr}} - \mathbf{x}_{\mathbf{meas}} \\ +\kappa_1 y\mathbf{x}_{\mathbf{corr}} \end{array} \right) \end{array} \right]^{1/2} \end{array} \right\} \quad (8)$$

Note that $\boldsymbol{x}_{corr}$ and $\boldsymbol{x}_{meas}$ are scalars when writing d_p in this form indicating that the measured and corrected absorbance in the equation are for a particular wavelength. Therefore we obtain a solution for d_p at each wavelength. As a result d_p estimated by this equation is obtained by averaging over all the wavelengths.

For the 4-component data, following the same procedure leads to the following equations.

$$a = \xi_a + \alpha_1 y + \alpha_2 y^2 + \beta_1 d_p + \beta_2 d_p^2 + \gamma_1 y \cdot d_p + \gamma_2 y \cdot d_p^2 + \gamma_3 y^2 \cdot d_p + \gamma_4 y^2 \cdot d_p^2 \quad (9)$$

$$b = \xi_b + \kappa_1 y + \kappa_2 y^2 + \eta_1 d_p + \eta_2 d_p^2 + \varsigma_1 y \cdot d_p + \varsigma_2 y \cdot d_p^2 + \varsigma_3 y^2 \cdot d_p + \varsigma_4 y^2 \cdot d_p^2 \quad (10)$$

$$d_p = \frac{-b \pm \sqrt{b^2 - 4a \cdot c}}{2a} \quad (11)$$

where $a = \beta_2 + \gamma_2 \cdot y + \gamma_4 \cdot y^2 + x_{corr} \cdot \left(\eta_2 + \varsigma_2 \cdot y + \varsigma_4 \cdot y^2\right)$

$$b = \beta_1 + \gamma_1 \cdot y + \gamma_3 \cdot y^2 + x_{corr} \cdot \left(\eta_1 + \varsigma_1 \cdot y + \varsigma_3 \cdot y^2\right)$$

$$c = \xi_a + \alpha_1 \cdot y + \alpha_2 \cdot y^2 - x_{meas} + x_{corr} \cdot \left(\xi_b + \kappa_1 \cdot y + \kappa_2 \cdot y^2\right)$$

In Stage 2, the spectrum of a sample whose particle size and concentration have to be estimated is subjected to the scatter correction method using the same reference spectrum ($\boldsymbol{x}_{ref}$) that corrects the calibration set. The corrected spectrum is then subjected to the PLS calibration model built in Stage 1 to obtain an estimate of the particle concentration $\hat{y}$. This value of $\hat{y}$ is then used along with one of the three inverse expressions mentioned above to get $\hat{d}_p$. Thus estimates for both particle diameter and concentration are obtained from the spectrum.

It should be noted that while the methodology is described for the case where MSC is used as scatter correction method, it can be easily applied to any other scatter correction technique provided the scatter correction parameters obtained from a technique have extractable information regarding the particle diameter.

3. Results and Discussion

An initial analysis was carried out using data from each of the measurement configurations, namely total transmittance (Td), total reflectance (Rd) and collimated transmittance (Tc). MSC, and two versions of EMSC namely EMSCL and EMSCW [8, 11] were applied to the datasets and the scatter correction parameters were examined. In this paper, only the results from data taken with the measurement configuration for which the scatter correction parameters exhibit a clear relationship with particle parameters (particle size and concentration) are shown in order to keep the discussion clear and concise. For the 2-component system MSC parameters obtained from the Td spectra and for the 4-component system MSC parameters obtained from the Rd spectra exhibited the clearest relationship with respect to d_p and y. The differences in performance of scatter correction methods in relation to measurement configuration was seen in an earlier simulation study[8] and observations made in this study using experimental data is consistent with that study. Therefore, when applying the method described in this paper for extracting particle size information, the choice of measurement configuration is an important factor.

Initial analysis showed that while EMSC could provide better scatter correction from the point of view of better performing calibration models for particle concentration, for the datasets considered here, the parameters obtained by applying EMSC did not show clear relationship with either d_p or y, indicating that any information on these properties that may be embedded in the parameters are not easily (if at all) extractable. Therefore MSC which showed clear dependence on particle diameter and concentration is used in the discussions below. It should be noted that it is possible to use EMSC for the step where a calibration model is built to predict the particle concentration $\hat{d}_p$ in order to get better estimates of $\hat{y}$ while using the MSC parameters to obtain the particle diameter information. For sake of simplicity, in this paper we chose MSC for correcting the spectra which is used to build the PLS model for $\hat{y}$ as well as for estimating $\hat{d}_p$ from the MSC parameters.

3.1 Analysis of scatter correction parameters in two- and four-component systems

For the first dataset (polystyrene-water), MSC was applied to the Td spectra after smoothing, and the MSC parameters, *a* and *b*, were plotted against d_p and y to investigate the information contained in the parameters. Fig. 2 shows that both parameters vary systematically with the scattering related sample conditions i.e. d_p and y. Figs. 2(a1) and (b1) show the variations in *a* and *b* with variations in particle diameter at fixed concentrations. Figs. 2(a2) and (b2) show the variations in *a* and *b* with variations in particle concentration at fixed particle diameters. It is clear that the MSC parameters are impacted by both particle concentration and diameter. The variation of both *a* and *b* with particle diameter was found to be well explained by a second order polynomial fit for each concentration. This can be seen from the solid curves in Figs. 2(a1) and (b1) which are obtained by regression. The effect of particle concentration on the MSC parameter *a* at fixed particle diameter required a third order polynomial which is indicated by the solid curves in Fig. 2(a2) while *b* was found to be well described by a linear fit which is shown by the solid lines in Fig. 2(b2). This analysis suggested the use of equations of the form given by Eqs. (3) and (4). The coefficients in these equations were estimated using least squares regression. The values and 95% confidence intervals of the coefficients in Eqs. (3) and (4) are given in Table 1. The confidence intervals indicate that all the coefficients are significant.Similar analysis was carried out with Rd spectra of the 4-component system .. Figs. 3(a1) and (b1) show the variations in *a* and *b* with variations in particle diameter at fixed concentrations. Figs. 3(a2) and (b2) show the variations in *a* and *b* with variations in particle concentration at fixed particle diameters. In this case, second order polynomial curves best described the variations of both *a* and *b* with particle diameter at fixed particle concentrations as well as with particle concentration at fixed particle diameters. The solid curves in the subplots of Fig. 3 are the best fit curves obtained by regression in each case. It is observed that, compared to the 2-component system, the MSC parameters for the 4-component system exhibit larger uncertainty in terms of their variations with d_p and y. This leads to higher error in fitting the 4-component samples as can be clearly observed by examining the fitted curves in Fig. 3. This analysis indicates that MSC parameters appear to contain extractable information regarding the scatter-related sample characteristics namely particle size and concentration.

The variations in the MSC parameters at each particle diameter and concentration seen in Fig. 3 suggest that the scatter correction parameters are influenced by one or more factors in addition to particle diameter and concentration. One plausible explanation is that the changes in concentrations of other components in the mixture will result in a change in the refractive index of the suspending medium. This will affect the intensity of light in two ways. It will affect the reflectance/transmittance at the glass boundaries of the cuvette and thus the overall intensity collected by the detector.[12] Also, a change in refractive index of a sample affects the magnitude of light scattered by the particles since light scattering by particles is fundamentally due to the refractive index contrast between the particles and the suspending (liquid) medium.

A simulated dataset consisting of spectra simulated for the same conditions as the samples in the experimental dataset was used to check the above hypothesis. Simulations were based on the Radiative Transfer Theory (RTT) which has been widely used in medical diagnostics and atmospheric sciences to accurately model the propagation of light through turbid media and known to provide good agreement with experimental data [13]. Details of the simulation are given in the supporting information. The absorption and scattering coefficients were calculated by using Mie Theory which accurately models scattering by spherical particles. The bulk absorption coefficients $\boldsymbol{\mu_a}$ and the bulk scattering coefficients $\boldsymbol{\mu_s}$ obtained using Mie theory are shown in Figures 4(c) and (d), respectively. The effect of change in the refractive index of the mixture due to the change in sample composition is observed from the slight difference between two adjacent $\boldsymbol{\mu_s}$ curves in Fig. 4(d). This small difference in the bulk scattering coefficient leads to differences in the spectra of samples which contain the same particle diameter and concentration but different composition of the liquid species in the mixture.

In Fig. 5, the relationship between MSC parameters used to correct the simulated Rd spectra (Rd_sim) with concentration and diameter show very similar patterns as observed in Fig. 3 which was obtained from the experimental dataset. The same uncertainty in MSC parameters for samples with the same particle conditions is also observed from the simulated dataset. It should be noted that in the simulations, no instrumental drift or other physical changes that induce variations in the spectra were included. The similarity in the uncertainties in the MSC parameters therefore implies that the baseline/slope change in the spectra of samples with the same sample conditions is due to the difference in refractive index of the samples due to differences in the concentrations of the liquid species which is captured by the MSC method. This conclusion can be made because in the simulations, the refractive index of the suspending medium comprising the liquid species in the mixture is the only physical property that is varying when particle diameter and particle concentrations are fixed. This analysis indicates that the scatter correction parameters are affected not just by particle size and concentration but also to a small extent by the refractive index of the medium. In other words, these parameters are a function of particle diameter, particle concentration and the refractive index of the mixture.

3.2 Extracting particle size information from scatter correction parameters

Given that the particle size information is present in the scatter correction parameters, it would be of interest to know if this information is extractable. Researchers have attempted to obtain particle size information through applying multivariate calibration models such as PLS to the spectra directly or after correction by empirical preprocessing methods.[15-18] It is however unclear, in these studies, whether it is the particle size or concentration that is modeled since the concentration of the particle in these studies are strongly correlated to the particle size. For instance, Rantanen *et al* reported a method for in-line particle diameter monitoring for high shear granulations in which the particle diameter increases during the process.[18] With the chemical contents in the granulator remaining the same, it implies that the particle number density decreases which can then be related to the changes in the particle diameter. Instead of modeling the particle diameter directly, multivariate regression is likely to model the information related to the particle number density, a correlated factor to the particle diameter, especially on the data preprocessed to remove scatter-related information. Since the effect of particle size on spectra is nonlinear and confounding effects arise due to competing absorption and scattering effects on the spectra, it may be more effective to use the scatter correction parameters. This is because the effect of absorption is decoupled and also because of the possibility of obtaining linear (in the sense of the regression parameters) models relating scatter correction parameters to particle sizes.

In this study, we compared the performance of models for estimating the particle diameter $\hat{d}_p$ using (a) PLS model built on spectra without applying scatter correction ($\boldsymbol{x}_{meas}$); (b) PLS model built on spectra after applying scatter correction ($\boldsymbol{x}_{corr}$); and (c) Regression models using MSC parameters and following the methodology described in §2.3. For the approach (c), 3 equations for estimating particle diameter namely, Eqs. (5),(6) and(8) for the 2-component dataset and Eqs. (9)-(11) for the 4 component dataset, were investigated. The two stage approach proposed in §2.3 was tested using cross-validation. The two steps were carried out by using all but one of the samples in stage 1 and applying the resultant model (Stage 2) to the left-out sample. This process is continued till all the samples have been left out from stage 1 once. Table 2 summarizes the performances of the different models for the 2- and 4-component datasets which are discussed in the proceeding sections.

3.2.1 Two-component system

From Table 2 it is seen that using the MSC parameters to estimate $\hat{d}_p$ leads to an appreciable reduction in the estimation errors. Using PLS models built on either $\boldsymbol{x}_{meas}$ or $\boldsymbol{x}_{corr}$ leads to similar performance in terms of RMSECV which is also evident in the RMSECV curves for the two models in Fig. 8(a). All the three equations used to predict $\hat{d}_p$ using MSC parameters (Eqs. (5), (6), and (8)) lead to appreciable reduction in the error compared to the PLS models. Eqs. (5) and (6) which use MSC parameters *a* and *b* respectively give more or less similar performance with around 55% reduction in error. Eq. (8) which combines the information contained in *a* and *b* provides the best performance with around 70% reduction in error. The predicted versus the actual diameters for the two PLS models and the model using Eq. (8) are given in the Supporting Information (Figxx). As mentioned previously the use of Eqs. (5), (6), and (8) for obtaining $\hat{d}_p$ requires the concentration of the particles to be estimated, and this was provided using PLS model built on the spectra for this purpose. Table 2 summarizes the performance of PLS models built on un-corrected $\boldsymbol{x}_{meas}$ and the scatter-corrected $\boldsymbol{x}_{corr}$ spectra to predict particle concentration. As expected the estimation error in concentration is lower when $\boldsymbol{x}_{corr}$ are used. If the scatter correction method is effective in selectively removing the underlying scattering and other non-chemical effect, then it should lead to a better PLS model for predicting particle concentration. . Therefore when using the three equations (Eqs. (5), (6) and (8)), the concentrations of particles estimated from the corrected spectra were provided as input.

3.2.2 Four-component system

In the case of 4-component system, the results were different from that observed in the 2-component dataset. From Table2, the lowest error in predicting particle diameter is obtained using a PLS model built on the spectra without scatter correction ($\boldsymbol{x}_{meas}$). The PLS model built on $\boldsymbol{x}_{corr}$ leads to more than 100% increase in the error. .. The best model for predicting the particle diameter using the MSC parameters was given by Eq. (11) which combines information in *a*, *b*, and $\boldsymbol{x}_{corr}$. Unlike the 2-component system, the error in this case is more than 100% higher compared to the PLS model using

$\boldsymbol{x}_{meas}$. The reason for this was investigated first by examining the performance of the PLS model to predict particle concentration which is an input for Eq. (11). From Table 2, it is seen that RMSECV for the estimated concentration is much higher compared to the 2-component dataset. Both $\boldsymbol{x}_{meas}$ and $\boldsymbol{x}_{corr}$ give similar levels of error in the estimated concentration though the model built on $\boldsymbol{x}_{corr}$ requires fewer numbers of latent variables. If the large error in estimated diameter $\hat{d}_p$ is due to the error contributed by $\hat{y}$, then by replacing $\hat{y}$ by the actual concentration y should result in significant improvement and lead to similar performances that seen for the 2-component dataset. However, the error in estimated $\hat{d}_p$ did not reduce significantly indicating that the source of this increase in error lies elsewhere.

Further investigation was carried out by examining the concentrations of the different components and their correlation structure. The 4-component dataset was designed to eliminate the concentration correlation between the polystyrene particles and other components of the system. However, in the dataset the particle diameter is weakly correlated to the main constituents of the medium, H_2O and D_2O with a correlation coefficient of about 0.26 with each of these components. This raises the possibility that the PLS model built on $\boldsymbol{x}_{meas}$ for estimating particle diameter will be improved by such a correlation. Examining the scores of the PLS model, it was found that the scores of the first latent variable and to a certain extent the second latent variable are linearly related to d_p, as indicated in Figs. 12(b1) and (b2). Examining the loadings of these two latent variables shown in Figs. 12(a1) and (a2), we see that they appear to be explaining variations that affect the baseline of the spectra i.e. light scattering. Applying MSC and then building a PLS model on $\boldsymbol{x}_{corr}$ would result in the removal of information regarding particle diameter and should lead to models with higher errors in the estimation of particle size. The scores of the first and second latent variables obtained by applying PLS to $\boldsymbol{x}_{corr}$ in Figs. 13(b1) and (b2) shows that the first latent variable no longer possesses a clear relationship with particle diameter. Also the first latent variable now resembles more like the second LV for the un-corrected spectra (Fig. 12(a2)). However, there is no significant pattern in this case with respect to d_p.

It is also interesting to note that the number of latent variables required for the PLS model to predict particle diameter is reduced from 7 when $\boldsymbol{x_{meas}}$ is used to 4 when $\boldsymbol{x_{corr}}$ is used. This explains the increase in the error in the estimated particle diameter when PLS is applied after scatter correction. Despite this removal of particle size information, the model obtained from $\boldsymbol{x_{corr}}$ is still statistically significant and almost of similar level of performance as the models using the scatter correction parameters to estimate particle size. This is probably due to the fact that $\boldsymbol{x_{corr}}$ still has chemical information regarding H_2O and D_2O which are in turn correlated to the particle diameter thus providing the ability to predict particle diameter despite most of the information regarding this parameter has been removed by scatter correction. The MSC parameters on the other hand, do not include the correlation between particle size and the concentrations of H_2O and D_2O, since these parameters are indicative of baseline and slope changes in the spectra while absorptivity changes (and thus information) due to concentration changes in H_2O and D_2O remain in the corrected spectra.

Recalling that the MSC parameters for the 4-component dataset are affected by particle size, concentration and the refractive index of the suspending medium (§3.1), it should be pointed out that the models relating particle diameter to the MSC parameters were developed by neglecting the effect of the refractive index changes. This could also potentially lead to an increase in the error in estimating particle diameter. A further point to be noted is that for the 4-component system, the prediction of particle size by using equations that arise from inverting the expressions relating *a* or *b* (i.e. Eqs. 9) and (10)) led to two positive values for the particle diameter when the quadratic equations are solved. The ambiguity resulting from this meant that the expressions were not practically usable and therefore the results pertaining to these inverted equations are not shown in Table 1. This problem was not encountered when the combined Eq. (11) was used. Since the equations relating the MSC parameters to particle diameter and concentration that are given here are not necessarily unique, it may be possible to develop an alternative regression model to overcome this problem.

4. Conclusions

This study provides an insight into the nature of information contained in the scatter correction parameters. It shows that a scatter correction technique which leads to better calibration models for estimating concentration of chemical species need not necessarily be the best in terms of the scatter correction parameters containing extractable information. It was found that the MSC parameters contained significant information regarding scatter-causing properties namely particle size and concentration. The parameters from EMSC which leads to better performing calibration models compared to MSC do not show a clear relationship with the scatter-causing properties. This may be due to the fact that the information is spread over a larger number of parameters and also the possibility that EMSC might be removing other non-chemical variations that may be presented in the dataset. Further, whether a clear relationship between the MSC parameters and the particle size and concentration was observed depended strongly on the measurement configuration, indicating that the performance of a scatter correction technique will depend on the measurement configuration. This is in line with the observations made in an earlier study based on simulations.[8]

Given that the information regarding particle size is present in the MSC parameters, a method to extract this information was proposed and evaluated using the two-component and four-component datasets. It was found that for the 2-component dataset, the method was effective in extracting this information and the model resulting from this method led to a reduction of about 70% in the error in the estimation of particle size compared to models obtained by applying PLS to the spectra. For the 4-component dataset, the error in using the proposed method was considerably higher. This appears to be due to the increased uncertainty contributed by the changes in the refractive index of the suspending medium which is not included in the model. Also the PLS model built on the spectra led to considerably lower error compared to the proposed method. Analysis indicates that this is due to chance correlations between particle diameter and the concentrations of D_2O and H_2O present in the mixture.

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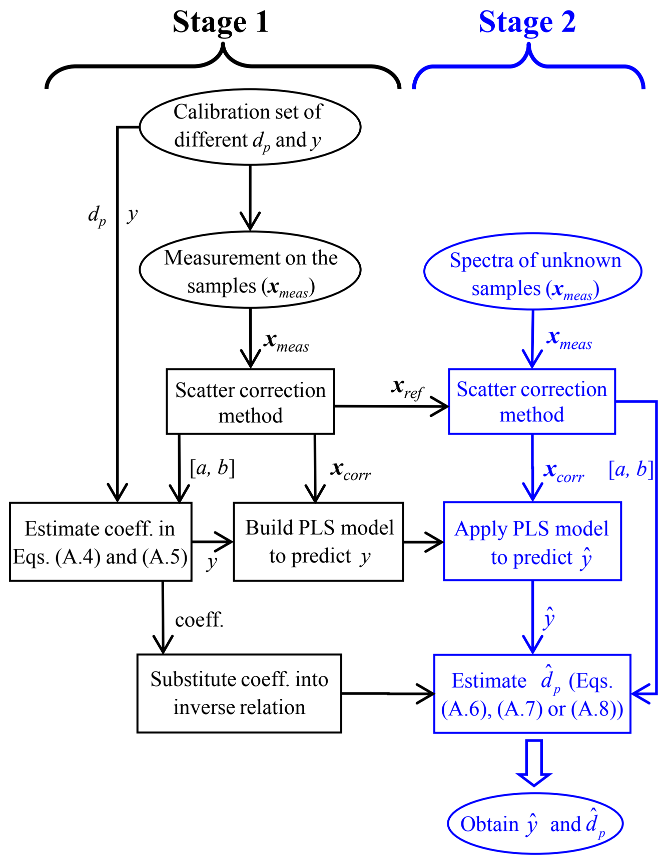
Figure 1



Stage 1
Stage 2
Calibration set of different d_p and y
d_p
y
Measurement on the samples ($\boldsymbol{x}_{meas}$)
Spectra of unknown samples ($\boldsymbol{x}_{meas}$)
$\boldsymbol{x}_{meas}$
$\boldsymbol{x}_{meas}$
Scatter correction method
$\boldsymbol{x}_{ref}$
Scatter correction method
[a, b]
$\boldsymbol{x}_{corr}$
$\boldsymbol{x}_{corr}$
[a, b]
Estimate coeff. in Eqs. (A.4) and (A.5)
y
Build PLS model to predict y
Apply PLS model to predict $\hat{y}$
coeff.
$\hat{y}$
Substitute coeff. into inverse relation
Estimate $\hat{d}_p$ (Eqs. (A.6), (A.7) or (A.8))
Obtain $\hat{y}$ and $\hat{d}_p$

Fig. 1. A flowchart of the methodology used for estimating particle diameter and concentration. The method involves Stage 1 : Calibration model building (steps in black) and Stage 2 : Prediction of particle diameters and concentrations of unknown samples (steps in blue).

Figure 2

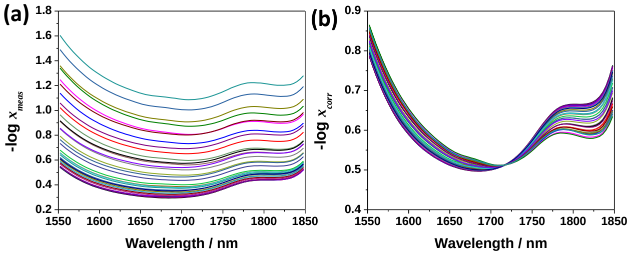


Fig. 2. (a) Measured total transmittance spectra (x_{meas}) of polystyrene-water 2-component dataset. (b) MSC preprocessed spectra (x_{corr}) using the mean of x_{meas} as a reference spectrum.

Figure 3

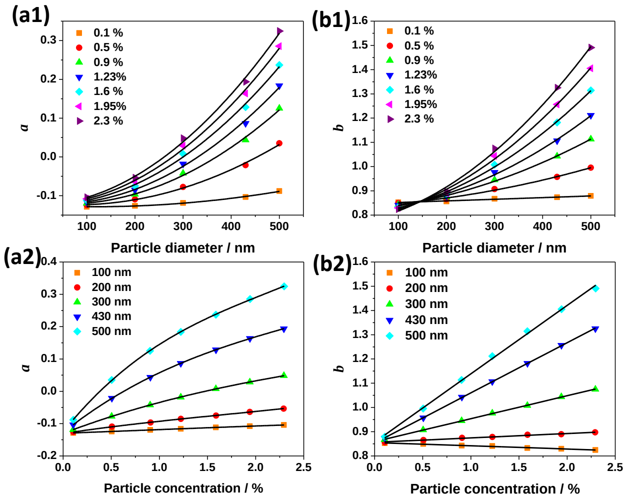


Fig. 3. (a) Changes in MSC parameter *a* in the 2-component system with (a1) particle diameter and (a2) concentrations. (b) Changes in MSC *b* with (b1) particle diameter and (b2) concentrations. Solid curves were generated from the best fit obtained using least squares regression.

Figure 4

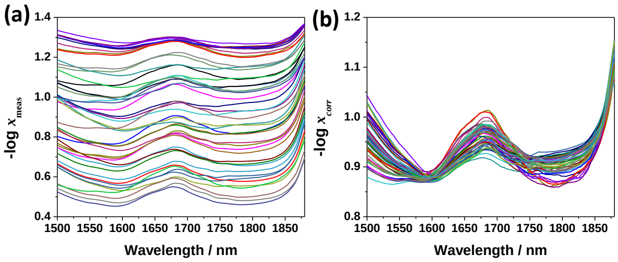


Fig. 4. (a) Measured total reflectance spectra (x_{meas}) of the 4-component dataset. (b) MSC preprocessed spectra (x_{corr}) using the mean of x_{meas} as a reference spectrum.

Figure 5

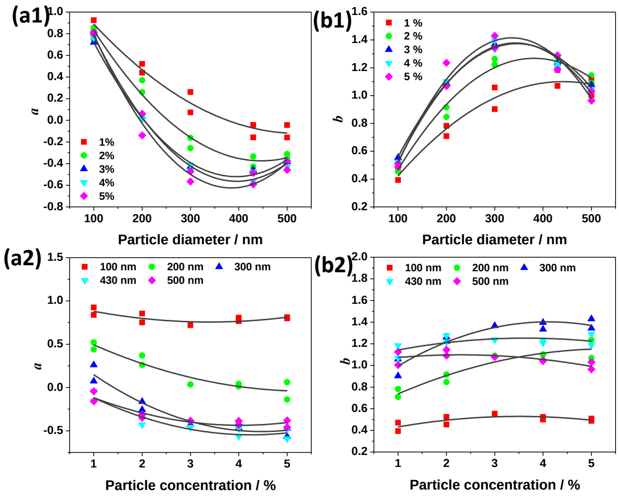


Fig. 5. (a) Changes in MSC parameter *a* in the 4-component system with (a1) particle diameter and (a2) concentrations. (b) Changes in MSC parameter *b* with (b1) particle diameter and (b2) concentration. Solid curves were generated from the best fit obtained by least squares regression.

Figure 6

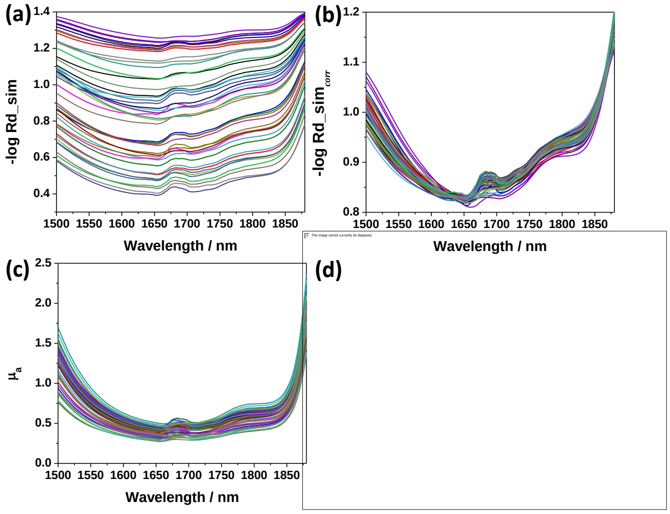


Fig. 6. (a) Simulated total reflectance spectra (Rd_sim) of the 4-component dataset. (b) MSC preprocessed spectra (Rd_sim$_{corr}$) using the mean of Rd_sim as a reference spectrum. The bulk absorption and scattering coefficients used for the simulation are in (c) and (d), respectively.

Figure 7

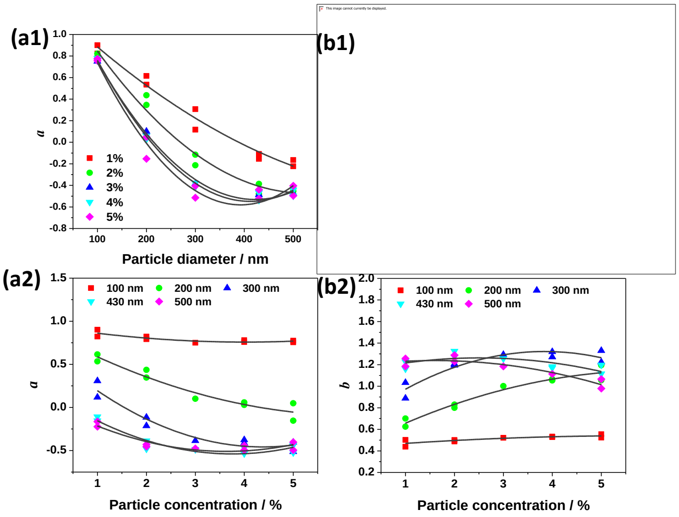


Fig. 7. Results of simulated spectra (Rd_sim) of the 4-component system after MSC preprocessing. (a) Changes in MSC parameters *a* with (a1) particle diameter and (a2) concentrations. (b) Changes in MSC *b* with (b1) particle diameter and (b2) concentration. Solid curves were generated from the best fit obtained by least squares regression.

Figure 8

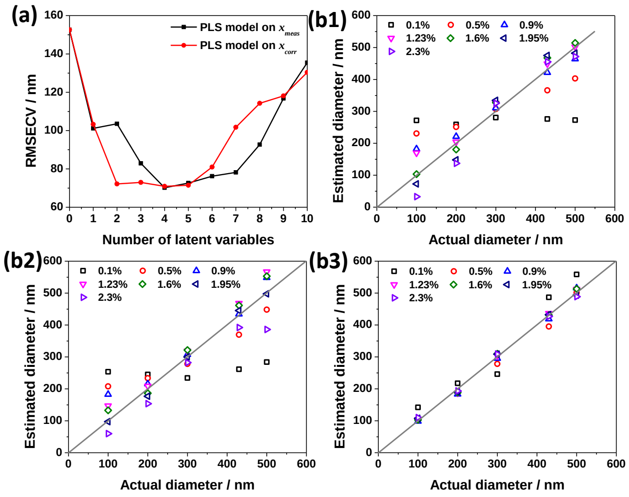


Fig. 8. (a) RMSECV curves of PLS models for estimating particle diameter in 2-component system from x_{meas} and x_{corr}. (b1) and (b2) are the predictions using PLS models built on x_{meas} and x_{corr}, respectively. (b3) is estimated using the inversion Eq. (A.8) in Supplementary Information which combines MSC parameters and x_{corr}.

Figure 9

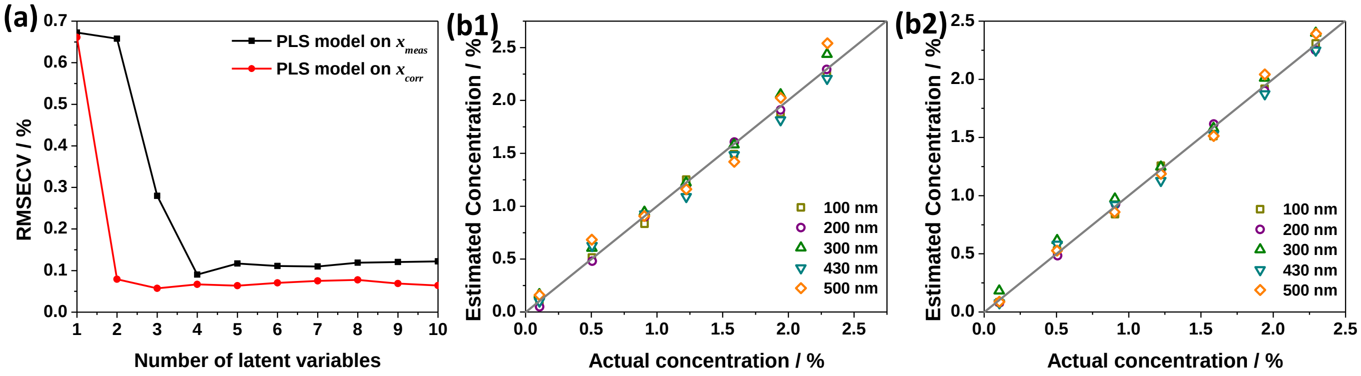


Fig. 9. (a) RMSECV curves of PLS models for estimating particle concentration in the two-component system. (b1) and (b2) show plots of estimated versus actual values of particle concentration in the system for PLS models built on x_{meas} and x_{corr}, respectively.

Figure 10

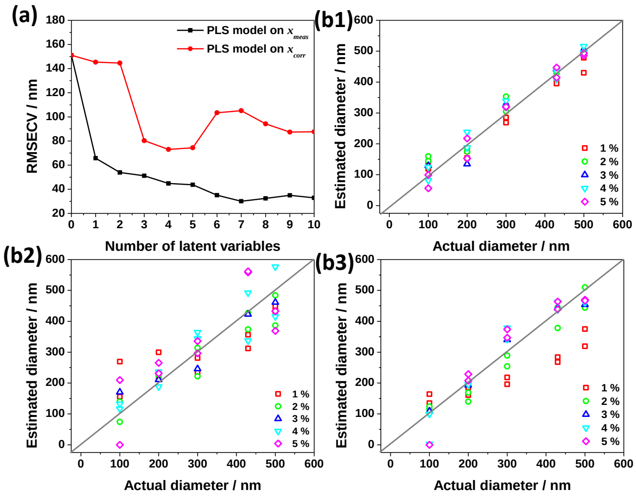


Fig. 10. (a) RMSECV curves of PLS models for estimating particle diameter in 4-component system. (b1) and (b2) are the prediction using PLS models built on x_{meas} and x_{corr}, respectively. (b3) is estimated using inversion Eq. (A.11) in Supplementary Information which combines MSC parameters and x_{corr}.

Figure 11

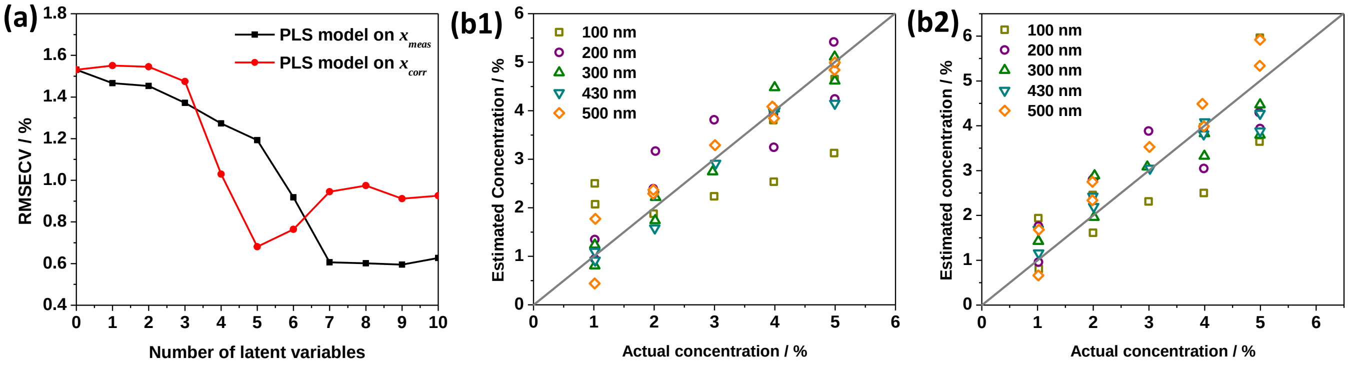


Fig. 11. (a)RMSECV curves of PLS models for estimating particle concentration in the four-component system. (b1) and (b2) show plots of estimated versus actual values of particle concentration in the system for PLS models built on x_{meas} and x_{corr}, respectively.

Figure 12

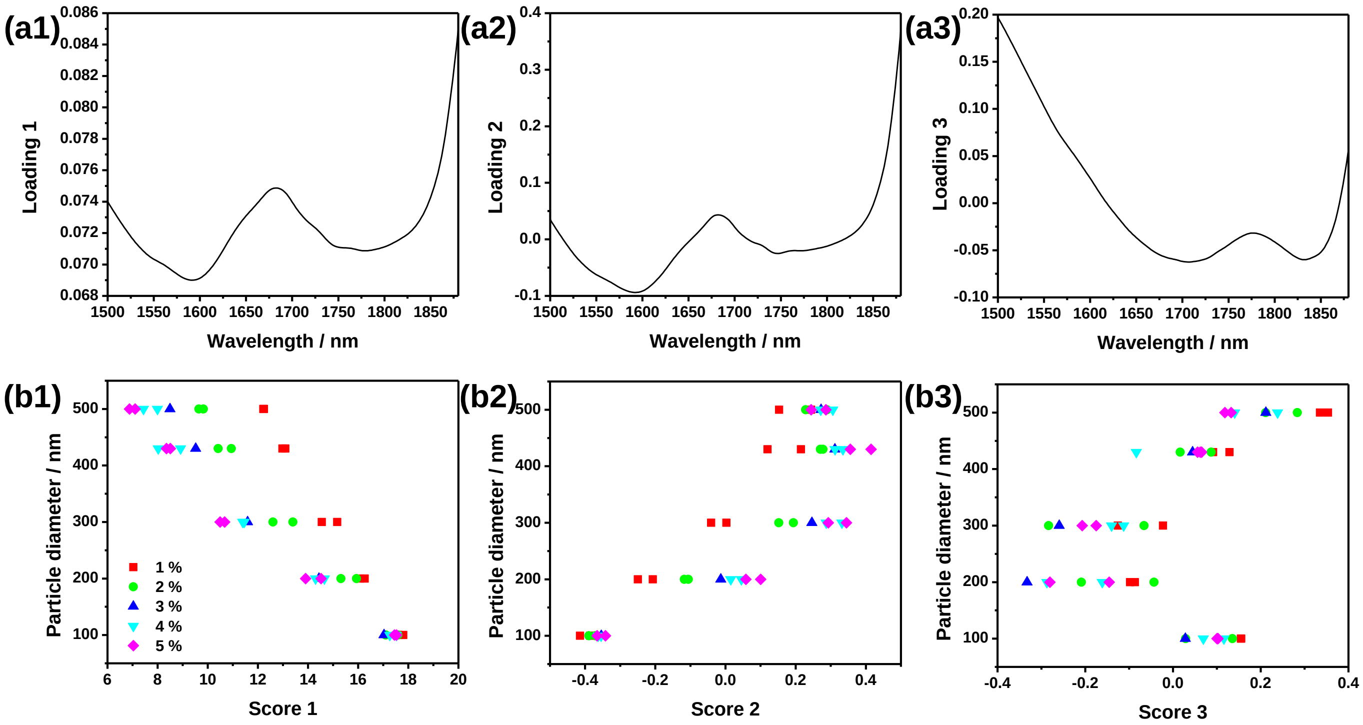


Fig. 12. (a1)-(a3) loading curves and (b1)-(b3) scores of the first 3 loadings of the PLS model built on x_{meas} to estimate particle diameter.

Figure 13

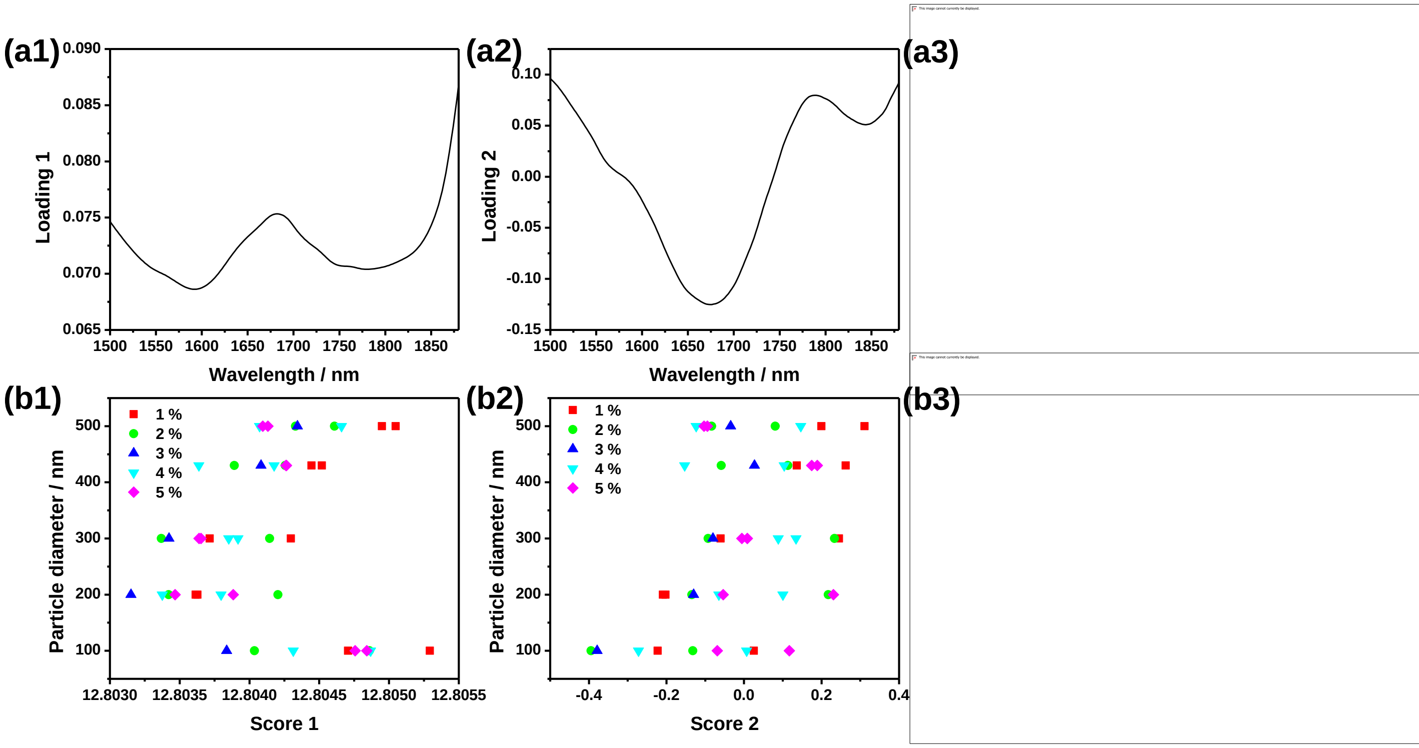


Fig. 13. (a1)-(a3) loading curves and (b1)-(b3) scores of the first 3 loadings of the PLS model built on x_{corr} to estimate particle diameter.