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Predicting percent composition of blends of biodiesel and conventional diesel using gas chromatography–mass spectrometry, comprehensive two-dimensional gas chromatography–mass spectrometry, and partial least squares analysis

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ABSTRACT

The percent composition of blends of biodiesel and conventional diesel from a variety of retail sources were modeled and predicted using partial least squares (PLS) analysis applied to gas chromatography–total-ion-current mass spectrometry (GC–TIC), gas chromatography–mass spectrometry (GC–MS), comprehensive two-dimensional gas chromatography–total-ion-current mass spectrometry (GCxGC–TIC) and comprehensive two-dimensional gas chromatography–mass spectrometry (GCxGC–MS) separations of the blends. In all four cases, the PLS predictions for a test set of chromatograms were plotted versus the actual blend percent composition. The GC–TIC plot produced a best-fit line with slope = 0.773 and *y*-intercept = 2.89, and the average percent error of prediction was 12.0%. The GC–MS plot produced a best-fit line with slope = 0.864 and *y*-intercept = 1.72, and the average percent error of prediction was improved to 6.89%. The GCxGC–TIC plot produced a best-fit line with slope = 0.983 and *y*-intercept = 0.680, and the average percent error was slightly improved to 6.16%. The GCxGC–MS plot produced a best-fit line with slope = 0.980 and *y*-intercept = 0.620, and the average percent error was 6.12%. The GCxGC models performed best presumably due to the multidimensional advantage of higher dimensional instrumentation providing more chemical selectivity. All the PLS models used 3 latent variables. The chemical components that differentiate the blend percent compositions are reported.

1. Introduction

The chemometric technique partial least squares (PLS) analysis was applied to total-ion-current gas chromatography (GC–TIC) separations, gas chromatography–mass spectrometry (GC–MS) separations, comprehensive two-dimensional gas chromatography–mass spectrometry (GCxGC–MS) separations, and GCxGC–TIC separations of biodiesels blended with conventional petroleum-based diesels, hereafter called biodiesel blends. The biodiesel blends varied from 0% to 20% by volume in biodiesel content. The purpose was to use PLS models to predict the percent composition of biodiesel in unknown biodiesel blends, compare the accuracy of predictions between one-dimensional (1D), two-dimensional (2D), and three-dimensional (3D) chromatography models, and identify the chemical species that correlate most with the variety of blend percent compositions using the model loadings.

Since computation time is generally less expensive than instrument time, the theme in this experiment is similar to the theme in high-throughput process analytical chromatography: resolution of all components in a complex sample can indeed be sacrificed for a speedier chromatographic separation if the desired chemical information can still be objectively obtained using automated PLS computations and the multidimensional advantage. The multidimensional advantage theorizes that chemically selective multidimensional instrumentation should increase the chemical information content per unit of instrument time, so it is expected that PLS applied to 3D GCxGC–MS chromatograms will yield a better model than PLS applied to 2D GCxGC–TIC chromatograms, which should yield a better model than PLS applied to 2D GC–MS chromatograms, which in turn should yield a better model than PLS applied to 1D GC–TIC chromatograms because the higher dimensional data contains more chemical information.

Commercial instrument software often does not provide the tools for multidimensional analysis, so it is necessary to export the data out of the commercial instrument software format and import it into a platform such as Matlab which is capable of multidimensional analysis. In this work, the PLS model is first built in Matlab using a training set of chromatograms of biodiesel blends with known percent compositions. PLS works by mathematically

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loading independent variables that have variations in signal intensity across all of the samples which correlate with variations in the given quantitative property, so PLS positively and negatively loads retention times that have signals that positively or negatively co-vary with given blend percent compositions [1–3]. To evaluate the model, independent test set chromatograms are regressed onto the model to yield predicted percent compositions, and the plot of predicted percent composition versus actual percent composition should ideally have a best-fit line with slope equal to 1 and a R^2 value equal to 1. The models will also be evaluated by average percent error, where percent error is defined as the quantity 100% times the absolute value of the actual blend percent composition minus predicted blend percent composition, divided by actual blend percent composition.

We specifically chose to study biodiesel blends because the ability to predict retail biodiesel blend percent composition is important to regulatory agencies, fuel compliance officers, and distributors of transport fuel who may be interested in monitoring authenticity, quality, contamination, adulteration, and accuracy of reported blend percent compositions. Instrumental methods for monitoring biodiesel blends have been reported using Fourier-transform infrared spectroscopy [4–6], near-infrared spectroscopy [7–9], mid-infrared spectroscopy [10], nuclear magnetic resonance spectroscopy [8,11–13], radio carbon analysis [14], electrospray ionization mass spectrometry [15], liquid chromatography [16,17], and GCxGC with flame ionization detection [18,19]. Other reports quantified biodiesel blends through one-dimensional gas chromatography [20] and GC–MS [9,21]. A PLS method was also reported to predict blend percent compositions of light cycle oil (LCO) with diesel fuel using GC–MS with variance of prediction equal to 0.5 for 1–20% LCO blends [22]. Of all of these methods, only the GC methods have the resolution required to attempt to identify individual components that affect the model's accuracy. Herein, we report and compare PLS models built to predict biodiesel blend percent compositions using GC–MS and GCxGC–MS separations. We mimicked references [6,14,15], by choosing to obtain samples where both the biodiesels and conventional diesels originated from different fuel stations in an attempt to address this source of variation in the model. However, calibration transfer is a significant problem in chromatography that we have not fully addressed in this work, and instead we focus on comparing the multidimensional models to the 1D model. The higher dimensional chromatograms should contain more valuable chemical information that is lost during compression into fewer dimensions.

2. Experimental

The PLS algorithm and n-way n-PLS algorithm were from the commercial software PLS Toolbox by Eigenvector Research, Inc (Manson, WA). Both used the same SIMPLS algorithm [1,3]. PLS was applied to the GC–TIC data and n-PLS was applied to the GC–MS, GCxGC–TIC, and GCxGC–MS data. For all of the models, 3 latent variables (LV) were used. To determine the number of latent variables, models for the GC–TIC, GC–MS, and GCxGC–TIC data were constructed using 1 LV to 8 LV and, the number of LV that yielded a minimum root-mean-squared error of cross-validation (RMSECV) was chosen to be 3 LV, as shown in Fig. 1 [1]. The computer crashed due to memory capacity issues during attempts to do leave-one-cross-validation of the GCxGC–MS data, so to be consistent and to avoid over-fitting the model, 3 LV were used for the GCxGC–MS model, as well. Cross-validation models were built, whereby, one-at-a-time, each chromatogram was pulled out of the data set, the model was rebuilt with the remaining chromatograms, and then that left-out chromatogram was submitted to the model to predict that sample's percent composition. This

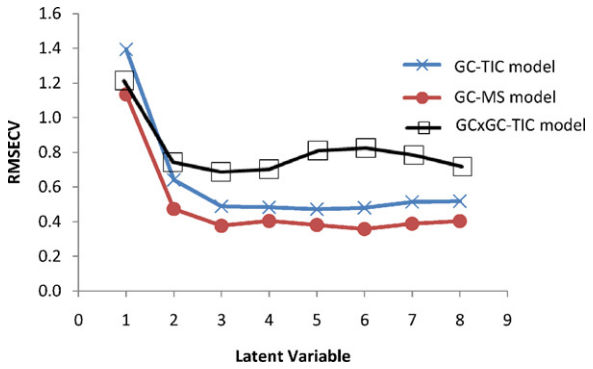

Fig. 1. Plot of RMSECV for each latent variable in the GC–TIC PLS model, the GC–MS n-PLS model, and the GCxGC–TIC n-PLS model.

was repeated for every chromatogram in the data set and yielded a leave-one-out cross-validation plot of predicted percent composition versus actual percent composition, which should ideally have $R^2 = 1.0$ and RMSECV close to 0. These metrics will be reported in Section 3 so that the 1D model can be compared to the multidimensional models to see which yield the most ideal results and ultimately determine which method is "best" for predicting biodiesel blends.

The biodiesel blends and conventional diesels were mixed with each other. The source and composition of each sample is listed in Table 1. The samples were acquired in September 2008, June 2009, July 2009, and August 2009 from Propel stations, Valero, and Union 76. These were mixed in various combinations to yield a range of blend percent compositions between 0% and 20% by volume. Percent composition is defined as the percent, by volume, of biodiesel in the biodiesel and conventional diesel blend. The blend percent compositions reported at the pump are assumed to be accurate.

To acquire GC–MS data, the biodiesel blends were injected neat with no solvent using an Agilent 6890N GC with 5973 quadrupole mass selective detector. The inlet was 250 °C, the injection volume was 1 μL, the split ratio was 120:1 and the flow rate was a con-

Table 1
Sources of biodiesels and conventional diesels used in the training set and test set.

	Source
Samples in training set actual blend percent composition (%v/v)	
0	Crown Hill Pure Diesel
0	Shoreline Pure Diesel
5	Kenmore 20% mixed with Shoreline Pure Diesel
5	West Seattle 20% mixed with Shoreline Pure Diesel
8.75	Kenmore 20% mixed with Shoreline Pure Diesel
8.75	West Seattle 20% mixed with Shoreline Pure Diesel
12.5	Kenmore 20% mixed with Shoreline Pure Diesel
12.5	West Seattle 20% mixed with Shoreline Pure Diesel
12.5	Ballard 99% mixed with Shoreline Pure Diesel
16.25	West Seattle 20% mixed with Shoreline Pure Diesel[a]
16.25	West Seattle 99% mixed with Shoreline Pure Diesel
20	Kenmore 20%
20	West Seattle 20%
Samples in test set actual blend percent composition (%v/v)	
5	Downtown Seattle 5%
5	Bellevue 20% mixed with Shoreline Pure Diesel
8.75	Downtown Seattle 5% mixed with Downtown Seattle 20%
8.75	Bellevue 20% mixed with Shoreline Pure Diesel
12.5	Downtown Seattle 5% mixed with Downtown Seattle 20%
12.5	Bellevue 20% mixed with Shoreline Pure Diesel
16.25	Downtown Seattle 5% mixed with Downtown Seattle 20%
16.25	Bellevue 20% mixed with Shoreline Pure Diesel
20	Downtown Seattle 20%
20	Bellevue 20%

[a] This sample was replaced with Ballard 99% mixed with Crown Hill Pure Diesel for GCxGC–TIC and GCxGC–MS data sets due to sample loss.

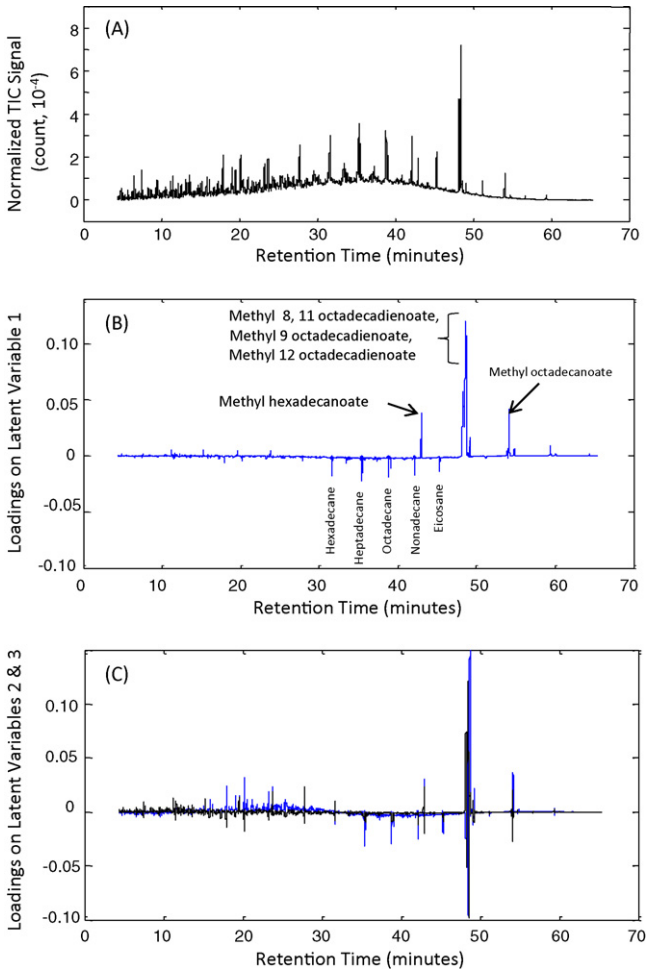

Fig. 2. (A) GC–TIC chromatogram of a 5% biodiesel blend. (B) PLS model loadings on LV 1 of the GC–TIC training set. (C) PLS model loadings on LV 2 (black) and LV 3 (blue) of the GC–TIC training set model are overlaid. The percent variance capture by LV 1, LV 2, and LV 3 was 67.83%, 8.87%, and 12.50%, respectively. (For interpretation of the references to color in this figure legend, the reader is referred to the web version of the article.)

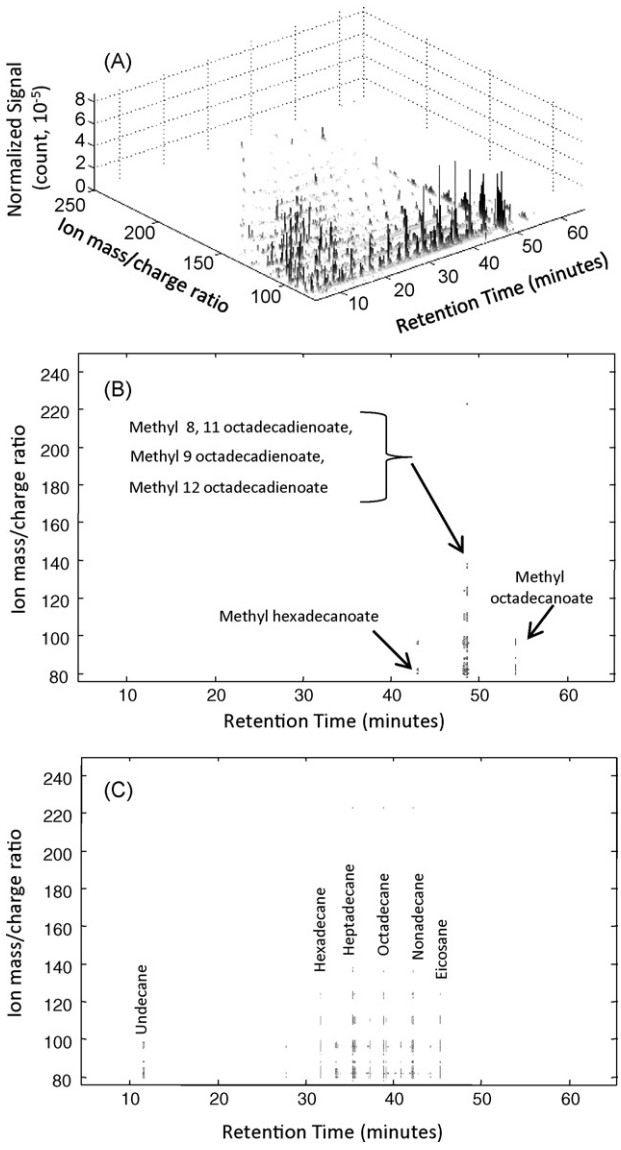

Fig. 3. (A) GC–MS chromatogram of a 5% biodiesel blend. (B) PLS positive loadings on LV 1 of the GC–MS training set. (C) PLS negative loadings on LV 1 of the GC–MS training set.

stant 0.8 mL/min. The oven started at 60 °C, and ramped at 3 °C/min up to 288 °C final temperature, with a 4.4 min solvent delay for the detector. The detector was set to scan ions 76–250 *m*/*z* at 5.6 scans/s with a threshold of 150 ion counts. The column was an Agilent HP-5MS, 30 m × 250 μm × 0.25 μm stationary phase. The ChromaTOF software with the NIST mass spectral matching library was used to identify certain components and the reported identifications agreed among three replicate chromatograms and had matches above 95.

To acquire the GCxGC–MS data, the biodiesel blends were injected neat with no solvent using an Agilent 6890 LECO GCxGC with time-of-light mass spectrometer. The inlet was 275 °C, the injection volume was 1 μL, the split ratio was 200:1 and the flow rate was 1.0 mL/min. The main oven started at 60 °C, held for 0.25 min, and ramped 3 °C/min to 290 °C, with a 6 s delay for the detector. The secondary oven was set at a constant +10 °C offset from the main oven with transfer line temperature to detector set at 305 °C. The second dimension separation time was set at 1.2 s. The detector was set to monitor ions 76-250 *m*/*z* at 100 spectra/s, with detector at 1600 Volts, electron energy at −70 V, and ion source at 300 °C. The first column was a RTX-5MS, 20 m × 250 μm × 0.50 μm. The second column was a RTX-200MS, 1.79 m × 180 μm × 0.20 μm.

To process the data sets that ended up being a maximum of 13 × 120 × 3890 × 105 data points, a 64-bit platform computer with 12 Gb RAM had to be used with Matlab-64, otherwise the computer would run out of memory during PLS calculations. Each chromatogram was normalized to the total sum of its signals.

3. Results and discussion

The 13 biodiesel blends in the training set, and the 10 test set biodiesel blends were all submitted to GC–MS and GCxGC–TOFMS chromatography. Each GC–TIC was obtained by summing each observed mass spectrum in a GC–MS chromatogram into a single scalar value, producing a 1D vector suitable for PLS analysis. A representative GC–TIC chromatogram of a 5% biodiesel blend is shown in Fig. 2A. The large peaks eluting near 50 min are fatty acid methyl esters (FAMEs) that are characteristic of biodiesels [23]. A representative 2D GC–MS chromatogram of a 5% biodiesel blend is shown in Fig. 3A. Parent ions can be seen at high molecular weights for the conventional diesel's alkane components. A representative GCxGC–TIC chromatogram of a 5% biodiesel blend is shown in Fig. 4A. The FAMEs elute at approximately 52 min and are retained longer on the polar second column than many of the

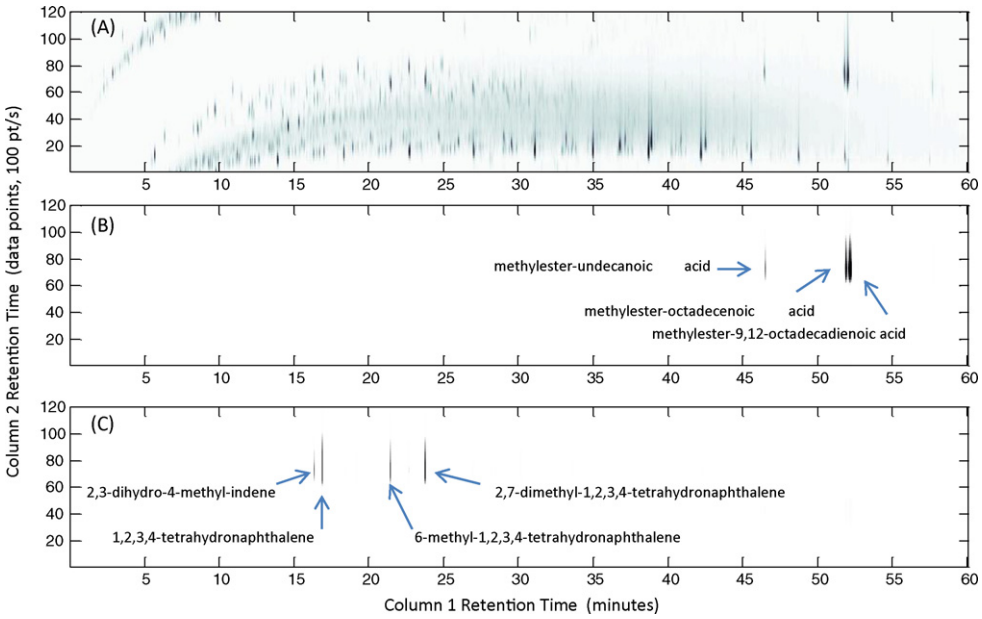

Fig. 4. (A) GCxGC–TIC chromatogram of a biodiesel blend. (B) PLS positive loadings on LV 1 of the GCxGC–TIC training set. (C) PLS negative loadings on LV 1 of the GCxGC–TIC training set.

saturated alkanes. The GCxGC–TIC chromatograms were obtained by summing each observed mass spectrum in a GCxGC–MS chromatogram into a single scalar value, producing a 2D array suitable for PLS analysis. The 3D GCxGC–MS chromatograms were modeled but none are shown in a figure. In the introduction we claimed that chemical class information may be lost with high speed/low resolution 1D chromatography, but it can be recovered by adding a chemically selective dimension to the instrumentation. It should be noted that high resolution 1D chromatography can yield classification results equivalent to 2D low resolution chromatography when the peak capacities are similar, but the peak capacity in our 1D GC–TIC is smaller than the peak capacity in our 2D GCxGC chromatogram. For the GC–TIC, the total separation time was divided by the approximate peak-width-at-base, yielding a theoretical peak capacity of 571 peaks, although a peak-counting algorithm (developed in-house to count local maxima with signals greater than a noise threshold) counted 392 peaks actually present in the 5% blend GC–TIC. For the GCxGC–TIC, the total separation space was divided by the approximate peak-area-at-base, yielding a theoretical peak capacity of 2918 peaks in the same 5% blend, although a peak-counting algorithm written in Matlab counted 795 peaks actually present in the GCxGC–TIC. If sample complexity is determined by the number of peaks present in the chromatogram, then the sample complexity is approximately equal among the 5–20% blends of biodiesels. The 0% blends are missing the 3–10 FAME peaks, so the sample complexity for the pure conventional diesel is slightly less than the biodiesel blends, but ultimately, sample complexity is largely determined by the hundreds of conventional diesel components rather than the biodiesel components [23].

The 13 training set GC–TIC chromatograms were submitted to PLS to model the chemicals that co-varied with the known blend percent compositions. A leave-one-out cross-validation was performed as a way of evaluating the robustness of the model. The PLS cross-validation plot of this 1D data had $R^2 = 0.999$ and RMSECV = 0.488 for 3 LV. Similarly, the 13 training set GC–MS chromatograms were submitted to PLS. The PLS cross-validation plot of this 2D data had $R^2 = 0.997$ and RMSECV = 0.376 for 3 LV.

The 10 test set GC–TIC chromatograms were regressed onto the GC–TIC PLS model and a plot of predicted versus actual blend percent composition for this 1D data had a best-fit line with slope = 0.773, y-intercept = 2.89, and $R^2 = 0.998$, as shown in Fig. 5A. The average percent error was 12.0%. Considering that the expected precision in chromatography with normalization is 1%, then the observed 12.0% error means that uncontrolled sources of sample variation were present, diminishing the reliability of the PLS model for predicting accurate blend percent compositions of unknown samples using 1D GC–TIC [24]. The PLS model yielded more accurate predictions for the 10 test set GC–MS chromatograms. The plot of predicted versus actual blend percent composition for this 2D data had a best-fit line with slope = 0.864, y-intercept = 1.72, and $R^2 = 0.999$, as shown in Fig. 5B. The average percent error was improved to 6.89%.

The 10 GCxGC–TIC test set chromatograms were regressed onto a PLS model that was built using the 13 training set GCxGC–TIC chromatograms and a plot of predicted versus actual blend percent composition for this 2D data had a best-fit line with slope = 0.983, y-intercept = 0.680, and $R^2 = 0.980$, as shown in Fig. 5C. The average percent error was 6.16%. This could be interpreted to mean that the GCxGC–TIC data produces a more robust model that yields more accurate predictions than the GC–TIC data, assuming a two-fold improvement in prediction error is significant.

When the 13 GCxGC–MS chromatograms were combined, a single 4D variable was produced that was too big for the Matlab memory to handle during PLS calculations, so a subset of the m/z were chosen and extracted to make a smaller 4D variable that could be used to build the GCxGC–MS PLS model. The subset of m/z were chosen by summing a 3D chromatogram of a 20% biodiesel blend in both chromatographic dimensions to yield a 1D mass spectrum and any m/z that had a summed signal greater than 1.12×10^8 was chosen, yielding 105 m/z, so each 3D GCxGC–MS chromatogram was reduced from $120 \times 3890 \times 176$ to $120 \times 3890 \times 105$. The 10 GCxGC–MS test set chromatograms were regressed onto a PLS model that was built using the 13 training set GCxGC–MS subset chromatograms and a plot of predicted versus actual blend percent composition for this 3D data had a best-fit line with slope = 0.980, y-intercept = 0.620, and $R^2 = 0.977$, as shown in Fig. 5D. The average percent error was 6.12%. The GCxGC–MS data produces a robust model that yields predictions as accurate as the GCxGC–TIC model. In general, the GCxGC data did contain more valuable chemical information than the GC data, which agrees with the multidimen-

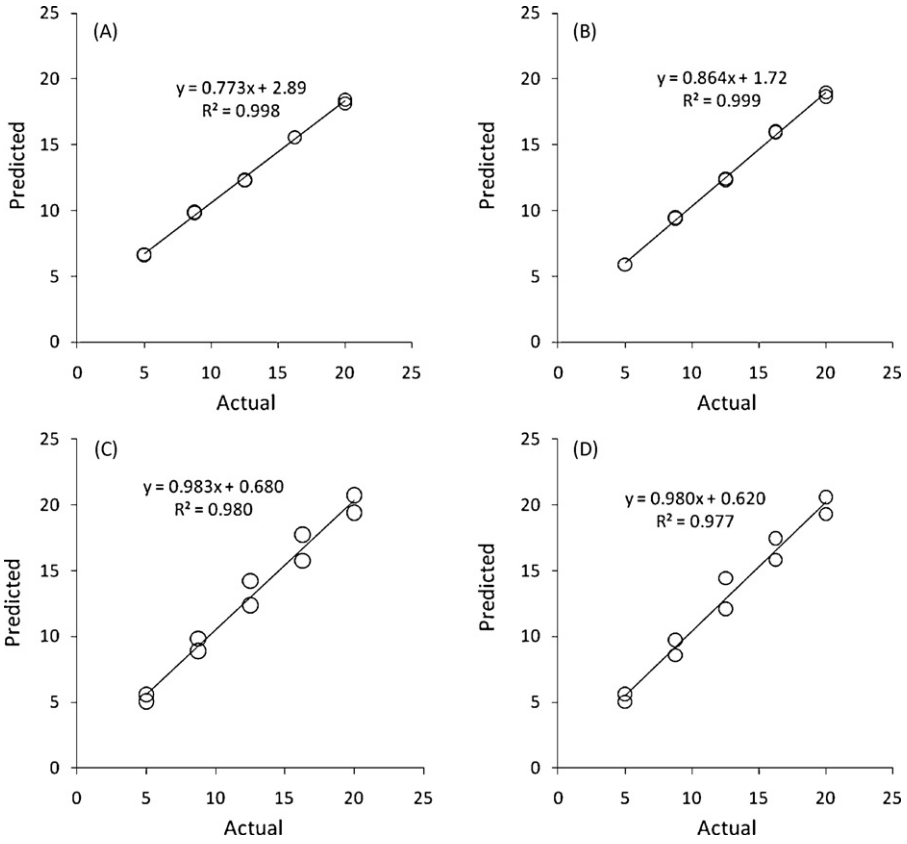

Fig. 5. Predicted blend percent composition (%v/v) is plotted versus actual blend percent composition (%v/v) for the test set using PLS models built with (A) 1D GC–TIC training set data, (B) 2D GC–MS training set data, (C) 2D GCxGC–TIC training set data, and (D) 3D GCxGC–MS training set data.

sional advantage theory. However, the third added dimension of the GCxGC–MS was not necessary because accuracy of the GCxGC–MS model was so similar to the accuracy of the GCxGC–TIC model. The standard deviation of the *y*-values of the best-fit line in Fig. 5D was 0.9, so if variance is defined as the square of the standard deviation, then the variance of prediction was 0.8. This is more than the 0.5 variance reported in reference [22] for 1–20% LCO blends. To understand the chemistry related to why a multidimensional separation produces a different prediction model than a 1D separation, we look at the model loadings in the next section.

The most highly loaded variables (in terms of absolute value) are the chemical signals that were most useful for predicting blend percent compositions. In general, the model loadings revealed that fatty acid methyl esters were most positively loaded and higher molecular weight alkanes or substituted naphthalenes were most negatively loaded. This is expected because a biodiesel is usually composed of a small number of FAMEs, while a conventional diesel is generally composed of many saturated hydrocarbon families, aromatics and naphthalenes. As the given percentage of biodiesel increases, the FAME signals increase, yielding a positive correlation to the percent composition. Also, as the given percentage of biodiesel increases, the signals decrease for the saturated hydrocarbons and aromatics that are characteristic of conventional diesel, yielding a negative correlation to the percent composition. For both GC–TIC and GC–MS models, the top five most positively loaded variables on LV 1, shown in Fig. 2B and Fig. 3B, were the following FAMEs: methylhexadecanoate, methyl-8,11-octadecadienoate, methyl-9-octadecadienoate, methyl-12-octadecadienoate, and methyloctadecanoate. The top five most negatively loaded variables, shown in Fig. 2B and Fig. 3C, were the following saturated straight chain hydrocarbons: hexadecane, heptadecane, octadecane, nonadecane, and eicosane. The GC–MS model had heavily loaded parent ions that were not available in the GC–TIC model, which may explain the improvement in prediction accuracy when comparing the GC–MS test set prediction results (Fig. 5A) to the GC–TIC test set prediction results (Fig. 5B). We do assume we can correlate loadings on LV 1 to chemicals that are important for modeling, however, we should point out that the models are a linear combination of LV 1, LV 2 and LV 3. So if highly loaded peaks in LV 1 are "cancelled out" by peaks in LV 2 or LV 3 that are highly loaded with an opposite sign, then it diminishes the importance of the chemicals we identified as being useful for prediction. The loadings on LV 2 and LV 3 are overlaid and shown in Fig. 2C for the GC–TIC model.

The GCxGC–TIC model most positively loaded the following three FAMEs shown in Fig. 4B: methylester-undecanoic acid, methylester-octadecenoic acid, and methylester-9,12-octadecadienoic acid. The GCxGC–TIC model most negatively loaded the following four aromatic compounds shown in Fig. 4C: 2,3-dihydro-4-methyl-indene, 1,2,3,4-tetrahydronaphthalene, 6-methyl-1,2,3,4-tetrahydronaphthalene, and 2,7-dimethyl-1,2,3,4-tetrahydronaphthalene. The added chemical selectivity provided by the second chromatographic column dimension helped resolve the aromatic compounds from the saturated alkanes, presumably causing the small improvement in prediction accuracy when comparing the predictions of the GC–MS model (Fig. 5B) to the predictions of the GCxGC models (Fig. 5C and D).

4. Conclusion

Monitoring blends of biodiesels and conventional diesels is a research topic currently gaining popularity as responsible stewardship of natural resources becomes increasingly important. We showed that n-way PLS can be used to predict or authenti-

cate biodiesel blend percent compositions with 6.12% error using GCxGC–MS and assuming future samples contain similar features to the original training set.

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References

[1] B.M. Wise, N.B. Gallagher, R. Bro, J.M. Shaver, W. Windig, S.R. Koch, PLS_Toolbox 3.5 for Use with Matlab™ 144–148 (2004) ISBN: 0-97611840-8.
[2] R.G. Brereton, Chemometrics:, Data Analysis for the Laboratory and Chemical Plant, Wiley, New York, 2003, 297.
[3] S. de Jong, Chemometr. Intell. Lab. 18 (1993) 251.
[4] M.F. Pimentel, M.G. Grece, S. Ribeiro, R.S. da Cruz, L. Stragevitch, J.G.A. Pacheco, L.S.G. Teixeira, Microchem. J. 82 (2006) 201.
[5] C.C.O. Flavia, R.R.B. Christian, F.R. Hugo, L.A.F. da Costa, A.Z.S. Paulo, J.C. Rubim, Anal. Chim. Acta 587 (2007) 194.
[6] J.S. Oliveira, R. Montalvao, L. Daher, P.A.Z. Suarez, J.C. Rubim, Talanta 69 (2006) 1278.
[7] R.E. Morris, M.H. Hammond, J.A. Cramer, K.J. Johnson, B.C. Giordano, K.E. Kramer, S.L. Rose-Pehrsson, Energy Fuel 23 (2009) 1610.
[8] G. Knothe, J. Am. Oil Chem. Soc. 78 (2001) 1025.
[9] J.A. Cramer, R.E. Morris, B. Giordano, S.L. Rose-Pehrsson, Energy Fuel 23 (2009) 894.
[10] M.A. Aliske, G.F. Zagonel, B.J. Costa, W. Veiga, C.K. Saul, Fuel 86 (2007) 1461.
[11] B. Diehl, G. Randel, Lipid Technol. 19 (2007) 258.
[12] M.R. Monteiro, A.R.P. Ambrozin, L.M. Liao, A.G. Ferreira, Fuel 88 (2009) 691.
[13] M.R. Monteiro, A.R.P. Ambrozin, M.S. Santos, E.F. Boffo, E.R. Perreira-Filho, L.M. Liao, A.G. Ferreira, Talanta 78 (2009) 660.
[14] C.M. Reddy, J.A. Demello, C.A. Carmichael, E.E. Peacock, L. Xu, J.S. Arey, Environ. Sci. Technol. (2008), doi:10. 1021/es071814.
[15] I. Eide, K. Zahlsen, Energy Fuel 21 (2007) 3702.
[16] T.A. Foglia, K.C. Jones, J.G. Phillips, Chromatographia 62 (2005) 115.
[17] M. Kaminski, E. Gilgenast, A. Przyjazny, G. Romanik, J. Chromatogr. A 1122 (2006) 153.
[18] J.V. Seeley, S.K. Seeley, E.K. Libby, J.D. McCurry, J. Chromatogr. Sci. 45 (2007) 650.
[19] F. Adam, F. Bertoncini, V. Coupard, N. Charon, D. Thiebaut, D. Espinat, M.C. Hennion, J. Chromatogr. A 1186 (2008) 236.
[20] P. Bondioli, L. Della-Bella, Riv. Ital. Sostanze Grasse 80 (2003) 173.
[21] R.C.M. Faria, J.J.C. Rezende, C.M. Rezende, A.C. Pinto, Quim. Nova 30 (2007) 1900.
[22] K.J. Johnson, S.L. Rose-Pehrsson, R.E. Morris, Pet. Sci. Technol. 24 (2006) 1175.
[23] A. Steinbach, A Comprehensive Analysis of Biodiesel, Biodiesel Magazine, November, 2007, http://www.biodieselmagazine.com/.
[24] D.A. Skoog, F.J. Holler, T.A. Nieman, Principles of Instrumental Analysis, 5th ed., Saunders College Publishing, Philadelphia, 1998, p. 696.