Published in final edited form as:

Appl Spectrosc. 2013 March ; 67(3): 283–290. doi:10.1366/11-06541.

Insight into Resolution Enhancement in Generalized Two-Dimensional Correlation Spectroscopy

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Abstract

Generalized two-dimensional correlation spectroscopy (2D COS) can be used to enhance spectral resolution in order to help differentiate highly overlapped spectral bands. Despite the numerous extensive 2D COS investigations, the origin of the 2D spectral resolution enhancement mechanism(s) are not completely understood. In the work here we studied the 2D COS of simulated spectra in order to develop new insights into the dependence of the 2D COS spectral features on the overlapping band separations, their intensities and bandwidths, and their band intensity change rates. We find that the features in the 2D COS maps that derive from overlapping bands are determined by the spectral normalized half-intensities and the total intensity changes of the correlated bands. We identify the conditions required to resolve overlapping bands. In particular, 2D COS peak resolution requires that the normalized half-intensities of a correlating band have amplitudes between the maxima and minima of the normalized half-intensities of the overlapping bands.

Keywords

Two dimensional correlation spectroscopy; resolution enhancement; overlapping bands; normalized half-intensity

INTRODUCTION

Two-dimensional correlation spectroscopy (2D COS) was first introduced by Noda,^{1, 2, 3, 4} as a mathematical technique to investigate the behavior of a system in response to external perturbations (temperature, pH, concentration, time etc.). 2D COS has been successfully applied to analyze NMR, IR, Raman, UV-Vis, fluorescence spectra etc.^{3, 4, 5, 6, 7, 8} 2D COS establishes correlations between spectral intensity variations within a set of spectra by generating two-dimensional maps that are known as the 2D synchronous and asynchronous spectra. Information such as the similarity, direction, and sequential order of correlated spectral intensity variations can be extracted by studying the 2D correlation peak features.^{4, 9} 2D COS also enhances spectral resolution by spreading highly overlapped band features along a second dimension.^{6, 10, 11, 12} Overlapping peaks are thought to be signaled by the appearance of correlation peaks in the 2D synchronous and asynchronous spectra. For these cases, 2D COS is a powerful tool to investigate complex spectral data from polymers, colloids, proteins, peptides, and pharmaceuticals etc.^{8, 13, 14}

Despite the utility of 2D COS resolution enhancement as recognized in 1990s,^{3, 15} and its wide use,^{16, 17, 18} the underlying resolution enhancement mechanism(s) is still not well

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understood. 2D synchronous or asynchronous correlated peaks occur if the intensities of two spectral features change in-phase or out-of-phase with each other.⁴ Although it is generally expected that the number of correlation peaks will be equal to the number of overlapping bands, this result does not always occur. For example, Yu et al. surprisingly found the presence of three correlation peaks in the 2D COS spectra that derived from one narrow band embedded within a single broad band.¹⁹

Previous studies indicate that artifactual, misleading features can be generated in 2D COS spectra that are easily misinterpreted to indicate the presence of overlapping bands. For example, bandwidth changes and band shifting can generate peculiar 2D COS peak features that do not derive from real resolved spectral bands.^{18, 20, 21, 22, 23, 24} It is hard to distinguish single band peak shifts from intensity changes from two overlapped bands, since both spectral changes generate similar 2D spectral patterns.^{24, 25} These results indicate that great care must be paid to correctly interpret any apparent resolution enhancement 2D COS spectral feature.

Much effort has been expended to understand the origin of 2D COS spectral features. Most recently, a diagnostic test was developed by Jung et al to distinguish features resulting from single band shifts from those resulting from overlapped bands.²⁶ Czarnecki's simulations show that multiple overlapping bands with changing relative intensities can give features very similar to 2D COS spectral features that result from frequency shifts of a single band. Thus, there can be confusion discriminating between peak shifts and those features deriving from overlapping bands.²⁷

It is clear that a deeper understanding of the origin of 2D COS spectral features is necessary in order to interpret 2D COS resolution enhancement features correctly. Thus, in the work here, we examine the dependence of 2D COS spectral features on overlapping band separations, band intensities and bandwidths, and the rates of band intensity changes. We identify the conditions required to resolve overlapping bands, and discuss the features in the 2D COS maps that signal overlapping bands.

EXPERIMENTAL SECTION

2D Correlation Analysis

Noda proposed a practical method to compute generalized 2D correlation spectra.^{4, 28} Assuming a set of m spectra collected over perturbation t at equally spaced increments, the 2D synchronous $\Phi(\nu_1, \nu_2)$ and asynchronous $\Psi(\nu_1, \nu_2)$ correlation intensities at ν_1 and ν_2 are calculated:

$$\Phi(\nu_1, \nu_2) = \frac{1}{m-1} \tilde{y}(\nu_1)^T \tilde{y}(\nu_2) \quad (1)$$

$$\Psi(\nu_1, \nu_2) = \frac{1}{m-1} \tilde{y}(\nu_1)^T N \tilde{y}(\nu_2) \quad (2)$$

where $\tilde{y}(\nu_i)$ is the dynamic spectral intensity matrix,

$$\tilde{y}(\nu_i) = \tilde{y}(\nu_i, t) = \begin{bmatrix} \tilde{y}(\nu_i, t_1) \\ \tilde{y}(\nu_i, t_2) \\ \dots \\ \tilde{y}(\nu_i, t_m) \end{bmatrix} \quad (3)$$

$\tilde{y}(\nu_i, t_j)$ is the dynamic spectral signal at ν_i and time t_j defined by the following equation:

$$\tilde{y}(\nu_i, t_j) = y(\nu_i, t_j) - \frac{1}{m} \sum_{k=1}^m y(\nu_i, t_k) \quad (4)$$

where $y(\nu_i, t_j)$ is the measured spectral signal at ν_i and time t_j . N in Eq. (2) is the Hilbert-Noda matrix, given by:

$$N = \frac{1}{\pi} \begin{bmatrix} 0 & 1 & \frac{1}{2} & \frac{1}{3} & \dots \\ -1 & 0 & 1 & \frac{1}{2} & \dots \\ -\frac{1}{2} & -1 & 0 & 1 & \dots \\ -\frac{1}{3} & -\frac{1}{2} & -1 & 0 & \dots \\ \dots & \dots & \dots & \dots & \dots \end{bmatrix} \quad (5)$$

The synchronous spectrum is the inner product of two dynamic spectral vectors measured at two different spectral frequencies, ν_1 and ν_2 . The asynchronous spectrum is the inner product of the dynamic spectral vectors and the orthogonal Hilbert-Noda matrix.

The 2D correlation spectra here were calculated as discussed above. We will consider here only those cross-peaks located below the correlation diagonal and label them as (ν_1, ν_2) , where ν_1, ν_2 refer to the abscissa and ordinate spectral frequencies.

Simulations

The simulated spectra used here were composed of 13 Gaussian bands $I_{i0}(\nu) = A_i e^{-\frac{(\nu - \nu_{ci})^2}{2w_i^2}}$ as shown in Fig. 1A, where w_i determines the full bandwidth at half height ($FWHH = 2(2\ln 2)^{1/2} w_i$), ν_{ci} is the band center frequency, ν is the spectral frequency, and A_i is the maximum amplitude of the i th Gaussian band. The i th band intensity decreases exponentially with time as $f_i(t) = e^{-k_i t}$. In the following discussion, B_i refers to the i th band.

The Gaussian band parameters and their intensity change functions are summarized in Table 1. The first five bands have identical maximum intensities and bandwidths and are separated by 100 cm^{-1} . The other eight bands are paired to form 4 overlapping bands B6/B7, B8/B9, B10/B11 and B12/B13. B6 and B7 are separated by 30 cm^{-1} , while B8 and B9, B10 and B11, B12 and B13 are separated by 8 cm^{-1} . All bands have $w = 14 \text{ cm}^{-1}$, except for B9 and B11 where $w = 11 \text{ cm}^{-1}$, and B13 with $w = 5 \text{ cm}^{-1}$. All bands have $A = 100$, except for B11 where $A = 36$.

The B1 to B5 band intensity change rate constants k are 0.05, 0.085, 0.1, 0.15, and 0.2 sec^{-1} , respectively. For B6, B8, B10, and B12, $k = 0.05 \text{ sec}^{-1}$, and for B7, B9, B11, and B13 $k = 0.2 \text{ sec}^{-1}$.

Fig. 1B shows the simulated spectra:

$$I(\nu, t) = \sum_{i=1}^m f_i(t) I_{i0}(\nu) \quad (6)$$

where $m = 13$ is the number of the Gaussian bands, and $f_i(t)$ is defined as the intensity change coefficient calculated as $f_i(t) = e^{-k_i t}$.

The simulation consists of 21 spectra with spectrum 1 the starting spectrum at $t = 0 \text{ sec}$. The time interval between spectra is 1 sec . The spacing between spectral data points is 2 cm^{-1} .

Spectral Normalization

The normalized spectral intensity $F(\nu, t)$ is calculated:

$$F(\nu, t) = [I(\nu, t) - I(\nu, 0)] / [I(\nu, T) - I(\nu, 0)] \quad (7)$$

where $I(\nu, t)$ is the spectral intensity at t .²⁹ Normalization makes the spectral intensities range from 0 to 1 over the total perturbation time T . The results of spectral signal intensity normalization are shown in Fig. 2. Eq. (7) can be rewritten:

$$I(\nu, t) = K(\nu)F(\nu, t) + b(\nu) \quad (8)$$

where $K(\nu) = I(\nu, T) - I(\nu, 0)$, and $b(\nu) = I(\nu, 0)$. The normalized half-intensity (NHI) is defined as the normalized intensity at $t = T/2$, as shown in Fig. 2B. Note, the 2D COS spectra in the paper are calculated from the original spectra without normalization. The normalized half-intensity parameter was used to explain the 2D COS spectral features.

Recently, as a further interpretation of Noda's sequential rule,⁴ Yu et al proposed the following correlation of the signs of the synchronous and asynchronous cross-peaks to the relative values of the NHI at ν_1 and ν_2 (Fig. 2)²⁹: *if $\Phi(\nu_1, \nu_2)\Psi(\nu_1, \nu_2) > 0$, then the spectral NHI at ν_1 is larger than that at ν_2 ; If $\Phi(\nu_1, \nu_2)\Psi(\nu_1, \nu_2) < 0$, then the spectral NHI at ν_1 is smaller than that at ν_2 .* The relationship was verified for spectra consisting of non-overlapping bands where the *band intensities change monotonically* and where the *normalized intensity time dependencies do not cross* (Fig. 2B).²⁹

The correlation proposed by Yu et al. between the relative NHI values at ν_1 and ν_2 and the signs of the synchronous and asynchronous 2D COS can be understood from Eq. (2). If the curvature of the normalized intensity time dependence at ν_1 is larger than that at ν_2 , the NHI at ν_1 will exceed that at ν_2 .

Multiplication of the normalized intensity spectral matrix of lower curvature at ν_2 by the Hilbert-Noda matrix, followed by multiplication by the higher curvature normalized spectral intensity matrix at ν_1 leads to a positive value $\Psi(\nu_1, \nu_2)$. Conversely, if the curvature of the normalized intensity time dependence at ν_1 is smaller than that at ν_2 , the $\Psi(\nu_1, \nu_2)$ value will be negative. If the normalized intensity time dependencies at ν_1 and ν_2 do not cross, the intensity time dependence curvatures are directly proportional to the NHI values.

RESULTS AND DISCUSSION

2D correlation analysis of simulated spectra

Significant resolution enhancement is provided by 2D COS. Fig. 1B shows that only the overlapping B6/B7 bands can be visually resolved in the one dimensional simulated spectra. Fig. 3 shows the 2D COS spectra generated by correlating the simulated spectra of the non-overlapping bands B1 to B5 with the overlapping bands B6 to B13. The synchronous and asynchronous spectral correlation peak features differ for the different overlapping bands, due to differences in band separations, band intensities, bandwidths, and temporal intensity changes (Fig. 3).

Fig. 3A shows the 2D COS synchronous spectra. The positive signs of the auto-peaks and cross-peaks in the synchronous spectrum indicate that the spectral intensity changes of all bands occur in the same direction. In this case, the 2D COS synchronous spectrum does not provide more information than does the original one-dimensional spectrum; no resolution improvement occurs.

The occurrence of doublet asynchronous cross-peaks with different signs in the (B2, B6/B7), (B3, B6/B7), (B4, B6/B7), (B2, B8/B9), (B3, B8/B9), and (B2, B10/B11) correlation regions in the Fig. 3B 2D asynchronous spectrum clearly resolves these overlapping bands. However, the cross-peak maxima and minima do not always correspond to the center frequencies of the overlapping Gaussian bands. This fact has previously been observed and discussed.^{21, 24}

In the (B1, B6/B7), (B5, B6/B7), (B1, B8/B9), (B4, B8/B9), (B1, B10/B11), and (B3, B10/B11) correlation regions, the 2D asynchronous spectrum shows only single negative or positive cross-peaks (Fig. 3B). The low values of the correlations in these regions indicate that the correlated bands have similar intensity change rates.

In the (B5, B8/B9), (B4, B10/B11), (B5, B10/B11) and (B5, B12/B13) asynchronous correlation regions, single cross-peaks dominate the entire overlapping band frequency regions. The 2D COS asynchronous spectrum does not resolve the underlying overlapping bands.

More complex triplet cross-peaks (positive, negative and positive) are observed in the (B2, B12/B13) and the (B3, B12/B13) asynchronous correlation regions. Triplets of cross-peaks are expected to result from three underlying components instead of the two overlapping Gaussians here. Similar 2D COS results were previously reported by Yu et al.¹⁹ Further complexity is evident in the (B4, B12/B13) correlation region that shows two positive cross-peaks instead of the positive and negative doublet cross-peaks commonly expected.

Modeling of the effect of band overlap on spectral normalized intensity

Yu et al²⁹ indicates that the normalized band intensities play an important role in determining the 2D COS spectral features. Here we examine the mechanism of formation of 2D correlation spectra to develop deeper insight.

Given the intensities of two Gaussian bands at frequency ν and at $t=0$, $I_{1,0}(\nu)$ and $I_{2,0}(\nu)$, an intensity matrix can be defined as:

$$I_0(\nu) = \begin{bmatrix} I_{1,0}(\nu) & I_{2,0}(\nu) \end{bmatrix} \quad (9)$$

An intensity change coefficient matrix is defined as:

$$f = \begin{bmatrix} f_1 \\ f_2 \end{bmatrix} = \begin{bmatrix} f_{1,0} & f_{1,1} & f_{1,2} & \cdots & f_{1,n} \\ f_{2,0} & f_{2,1} & f_{2,2} & \cdots & f_{2,n} \end{bmatrix} \quad (10)$$

where the f_1 and f_2 vectors consist of component $f_{i,j}$, which are the intensity change coefficients of the i th band for the j th time or perturbation step. The spectral intensity of the overlapping bands can be calculated as:

$$I(\nu) = I_0(\nu) f = \begin{bmatrix} I_{1,0}(\nu) & I_{2,0}(\nu) \end{bmatrix} \begin{bmatrix} f_{1,0} & f_{1,1} & f_{1,2} & \cdots & f_{1,n} \\ f_{2,0} & f_{2,1} & f_{2,2} & \cdots & f_{2,n} \end{bmatrix} \quad (11)$$

and rewritten as:

$$I(\nu) = \begin{bmatrix} I_{1,0}(\nu)f_{1,0} + I_{2,0}(\nu)f_{2,0} & I_{1,0}(\nu)f_{1,1} + I_{2,0}(\nu)f_{2,1} & \cdots & I_{1,0}(\nu)f_{1,n} + I_{2,0}(\nu)f_{2,n} \end{bmatrix} \quad (12)$$

For the j th perturbation time step, the normalized intensity at frequency ν :

$$F_j(\nu) = \frac{I_{1,0}(\nu)f_{1,j} + I_{2,0}(\nu)f_{2,j} - I_{1,0}(\nu)f_{1,0} - I_{2,0}(\nu)f_{2,0}}{I_{1,0}(\nu)f_{1,n} + I_{2,0}(\nu)f_{2,n} - I_{1,0}(\nu)f_{1,0} - I_{2,0}(\nu)f_{2,0}} \quad (13)$$

The normalized intensity $F_{i,j}(\nu)$ of a single Gaussian band i at the j th time step would be written:

$$F_{i,j}(\nu) = \frac{I_{i,0}(\nu)f_{i,j} - I_{i,0}(\nu)f_{i,0}}{I_{i,0}(\nu)f_{i,n} - I_{i,0}(\nu)f_{i,0}} = \frac{f_{i,j} - f_{i,0}}{f_{i,n} - f_{i,0}} \quad (14)$$

It can be seen that $F_{i,j}(\nu)$ does not depend on frequency. Eq. (14) can be rewritten as:

$$f_{i,j} = (f_{i,n} - f_{i,0})F_{i,j} + f_{i,0} = \lambda_i F_{i,j} + \delta_i \quad (15)$$

where $\lambda_i f_{i,n} - f_{i,0}$ and $\delta_i = f_{i,0}$. Substituting Eq. (15) into (13), the normalized intensities of overlapping bands at the j th time step can be expressed as follow:

$$F_j(\nu) = \frac{\lambda_1 I_{1,0}(\nu)F_{1,j} + \lambda_2 I_{2,0}(\nu)F_{2,j} - \lambda_1 I_{1,0}(\nu)F_{1,0} - \lambda_2 I_{2,0}(\nu)F_{2,0}}{\lambda_1 I_{1,0}(\nu)F_{1,n} + \lambda_2 I_{2,0}(\nu)F_{2,n} - \lambda_1 I_{1,0}(\nu)F_{1,0} - \lambda_2 I_{2,0}(\nu)F_{2,0}} \quad (16)$$

Since $F_{1,0} = F_{2,0} = 0$ and $F_{1,n} = F_{2,n} = 1$, then $F_j(\nu)$ can be simplified:

$$F_j(\nu) = \frac{\lambda_1 I_{1,0}(\nu)F_{1,j} + \lambda_2 I_{2,0}(\nu)F_{2,j}}{\lambda_1 I_{1,0}(\nu) + \lambda_2 I_{2,0}(\nu)} \quad (17)$$

For Gaussian bands with $I_{i0}(\nu) = A_i \cdot e^{-\frac{(\nu - \nu_{ci})^2}{2w_i^2}}$, $F_j(\nu)$ is

$$F_j(\nu) = \frac{\lambda_1 A_1 e^{-\frac{(\nu - \nu_{c1})^2}{2w_1^2}} F_{1,j} + \lambda_2 A_2 e^{-\frac{(\nu - \nu_{c2})^2}{2w_2^2}} F_{2,j}}{\lambda_1 A_1 e^{-\frac{(\nu - \nu_{c1})^2}{2w_1^2}} + \lambda_2 A_2 e^{-\frac{(\nu - \nu_{c2})^2}{2w_2^2}}} \quad (18)$$

The normalized spectral intensity $F_j(\nu)$ is a function of the Gaussian center frequencies, ν_{ci} , bandwidths, w_i , band amplitudes A_i , and λ_i .

Normalized intensities of simulated spectra

Fig. 4A shows the time and frequency dependence of the normalized simulated spectral intensities, $F_j(\nu)$. The normalized spectral intensities at the different time steps show somewhat similar profiles, except at $t=0$ and $t=20$ sec that are given by horizontal lines of fixed value. The NHI value of $F_{10}(\nu)$ given by $F_{T/2}(\nu) = F_{10}(\nu)$ at $t=10$ sec, is shown in Fig. 4B. $F_{T/2}(\nu)$ for each of the non-overlapping B1 to B5 bands have single values that are independent of frequency, except in the wings where the adjacent band intensities begin to dominate. $F_{T/2}(\nu)$ in the overlapping band regions is frequency dependent.

Fig. 4C compares the time dependence of the normalized spectral intensity, $F_j(\nu)$ of the non-overlapping band B3 to the $F_j(\nu)$ of the overlapping band B12/B13 at frequencies 2162, 2202 and 2210 cm^{-1} . The normalized spectral intensities $F_j(\nu)$ monotonically increase with time. The normalized intensities $F_j(2162 \text{ cm}^{-1})$ at all times are smaller than those of the B3

band. However, as ν increases, the $F_j(\nu)$ time dependence curve shows an increasing curvature as shown in Fig. 4C. The B3 band $F_{B3,j}$ curve crosses that of $F_j(2202 \text{ cm}^{-1})$ (Fig. 4C). The normalized intensity time dependence of the B3 band crosses $F_j(\nu)$ in the B12/B13 overlapping band frequency region, as well as in the other regions that are indicated by the Fig. 4B thick black vertical bars.

Fig. 4D shows the frequency dependence of the NHI, $F_{T/2}(\nu)$ in the frequency region between 1830 and 2250 cm^{-1} . In the overlapping B6/B7 frequency region, for $\nu < 1850 \text{ cm}^{-1}$, $F_{T/2}(\nu)$ has the value associated with NHI of the isolated B6 band $F_{B6,T/2}$. As the frequency increases, the B7 band begins to dominate, and $F_{T/2}(\nu)$ reaches the maximum value associated with isolated single B7 band.

The B8/B9 and B10/B11 doublet bands overlap more than do the B6/B7 bands. Further, bands B9 and B11 are narrower than are bands B8 and B10 (Table 1). The result is that bands B8 and B10 dominate only in the low frequency wings. Thus, the NHI of the overlapping B8/B9 and B10/B11 bands never reach the NHI values of the isolated B9 and B11 bands.

Band B13 is much narrower than is band B12, and their band center frequencies are close (Table 1). In areas 3 and 4, B12 dominates, and $F_{T/2}(\nu)$ has the value of the isolated band B12, $F_{B12,T/2}$. Between area 3 and area 4 $F_{T/2}(\nu)$ shows a narrower peak due to the narrow B13 band contribution.

Interpretation of asynchronous spectral features

Yu and coauthors discussed the relationship between the 2D COS spectra calculated from the original spectra and from the normalized spectra:²⁹

$$\Phi(\nu_1, \nu_2) = K(\nu_1)K(\nu_2)\Phi'(\nu_1, \nu_2) \quad (19)$$

$$\Psi(\nu_1, \nu_2) = K(\nu_1)K(\nu_2)\Psi'(\nu_1, \nu_2) \quad (20)$$

where $K(\nu_i) = I(\nu_i, T) - I(\nu_i, 0)$ is the total spectral intensity change at frequency ν_i ;

$\Phi'(\nu_1, \nu_2) = \frac{1}{m-1} \tilde{F}(\nu_1)^T \tilde{F}(\nu_2)$ and $\Psi'(\nu_1, \nu_2) = \frac{1}{m-1} \tilde{F}(\nu_1)^T N \tilde{F}(\nu_2)$ are the synchronous and asynchronous correlation intensities of the normalized spectra, respectively; m is the number of spectra, $\tilde{F}(\nu_i)$ is the normalized dynamic spectral intensity matrix:

$$\tilde{F}(\nu_i) = \tilde{F}(\nu_i, t) = \begin{bmatrix} \tilde{F}(\nu_i, t_1) \\ \tilde{F}(\nu_i, t_2) \\ \dots \\ \tilde{F}(\nu_i, t_m) \end{bmatrix} \quad (21)$$

$\tilde{F}(\nu_i, t_j)$ is the normalized dynamic spectral intensity at frequency ν_i and time t_j defined by:

$$\tilde{F}(\nu_i, t_j) = F(\nu_i, t_j) - \frac{1}{m} \sum_{k=1}^m F(\nu_i, t_k) \quad (22)$$

Normalization does not affect the relative values of the NHI.²⁹

Eq. (19) and (20) shows that the asynchronous correlation signs and intensities depend on $K(\nu_1)K(\nu_2)$ and $\Psi'(\nu_1, \nu_2)$. If $K(\nu_1)K(\nu_2)$ is positive, the signs of $\Psi'(\nu_1, \nu_2)$ and $\Psi(\nu_1, \nu_2)$, and $\Phi(\nu_1, \nu_2)$ and $\Phi'(\nu_1, \nu_2)$ are identical. Following Yu's hypothesis,²⁹ given the positive values of $\Phi(\nu_1, \nu_2)$ and $\Phi'(\nu_1, \nu_2)$, in our case, if the NHI at ν_1 is larger than that at ν_2 , $\Psi'(\nu_1, \nu_2)$ and $\Psi(\nu_1, \nu_2)$ will be positive, while if the NHI at ν_1 is less than that at ν_2 , $\Psi'(\nu_1, \nu_2)$ and $\Psi(\nu_1, \nu_2)$ will be negative. If the NHI of the two correlated signals are equal, $\Psi'(\nu_1, \nu_2) = 0$. The larger the NHI difference between the two correlated bands, the larger will be $\Psi'(\nu_1, \nu_2)$.⁴

$K(\nu_1)K(\nu_2)$ is the product of the total intensity changes at frequencies ν_1 and ν_2 over the overall perturbation time range. The smaller the intensity changes, the smaller will be the $\Psi(\nu_1, \nu_2)$ correlation intensity. Thus, the correlation intensity maximum occurs at a frequency where $K(\nu_1)K(\nu_2)\Psi'(\nu_1, \nu_2)$ is a maximum. The maximum values of $K(\nu_1)K(\nu_2)$ and $\Psi'(\nu_1, \nu_2)$ occur at frequencies that are not necessarily the Gaussian center frequencies (Fig. 3D). Thus, the maximum value of $K(\nu_1)K(\nu_2)\Psi'(\nu_1, \nu_2)$ will not necessarily occur at Gaussian center frequencies (Fig. 3D).

The 2D asynchronous spectral features can be predicted from the calculated NHI and the $K(\nu_1)K(\nu_2)$ values

For example, in the correlation between the B3 band and the overlapping bands in the frequency regions [p1, p2], [p3, p4] and [p7, p8], $F_{B3,T/2}$ is smaller than $F_{T/2}(\nu)$ (Fig. 5). Thus, given Yu's hypothesis,²⁹ with a positive value of $K(\nu_1)K(\nu_2)$, $\Psi'(\nu_1, \nu_2)$ and $\Psi(\nu_1, \nu_2)$ will be negative, as observed. Asynchronous cross-peaks $\Psi(\nu_1, \nu_2)$ are expected to be positive in frequency regions where $F_{B3,T/2}$ is larger than $F_{T/2}(\nu)$. However, the $K(\nu_1)K(\nu_2)$ values in the four blue shaded areas of Fig. 5 are very small, since $-K(\nu_2)$ are small. Thus, the correlation intensities $\Psi(\nu_1, \nu_2)$ between the B3 band and the overlapping bands in these areas will be very small, as observed.

The asynchronous correlation peak intensities $\Psi(\nu_1, \nu_2)$ also depend on the relative values of $F_{T/2}(\nu)$ of the overlapping bands and $F_{B3,T/2}$. In the frequency region [p5, p6], $F_{B3,T/2}$ is slightly smaller than $F_{T/2}(\nu)$. Thus, the 2D correlation will give vanishingly small cross-peaks. All of these conclusions are verified in the calculated 2D asynchronous spectrum.

At frequencies around points p1 to p8 (Fig. 5), the time dependencies of the normalized intensities cross that of band B3 (black vertical bars in Fig. 4B). This crossing makes the signs of $\Psi(\nu_1, \nu_2)$ not predictable from Yu et al.'s conclusions. As shown in Fig. 4C, the time dependence $F_j(\nu)$ curvature increases as the frequency increases. Thus, the normalized intensity difference between the B3 band and the overlapping bands between 2162 and 2210 cm^{-1} decrease monotonically ($F_{B3,T/2} - F_{T/2}(\nu)$) as the frequency increases. Thus, the 2D asynchronous correlation intensities $\Psi(\nu_1, \nu_2)$ decrease monotonically: from positive to negative in the frequency range between 2162 to 2210 cm^{-1} . At frequencies close to p7=2202 cm^{-1} , the 2D correlation asynchronous intensities will be very small. Similar situations will occur whenever the normalized intensities of correlated bands cross.

Interpretation of the (B3, B12/B13) correlation spectral feature

In the B12/B13 band frequency region, the overlapping B13 band is much narrower than is the B12 band. The separation between these bands is small (Fig. 4D). The overlapping B12/B13 band $F_{T/2}(\nu)$ peak has a much narrower bandwidth than does the difference spectrum between the simulated spectral at $t=0$ and $t=20 \text{ sec}$, $-K(\nu_2)$ (Fig. 5).

Between points O₁ and p7 (Fig. 5), $F_{B3,T/2}$ is larger than that of the B12/B13 band $F_{T/2}(\nu)$. Thus, the asynchronous cross-peak $\Psi(\nu_1, \nu_2)$ will be positive. In contrast, in the frequency region between p7 and p8, $F_{B3,T/2}$ is smaller than $F_{T/2}(\nu)$, forcing the asynchronous cross-

peak $\Psi(\nu_1, \nu_2)$ to be negative. In the frequency region between p8 and O₂, $F_{B3,T/2}$ is again larger than $F_{T/2}(\nu)$. The values of $K_1(\nu_1) K_2(\nu_2)$ are sufficient large to generate a positive cross-peak. Thus, triplet asynchronous cross-peaks are generated in the (B3, B12/B13) correlation region.

Interpretation of the (B4, B12/B13) correlation spectral feature

Fig. 5 shows that in the overlapping B12/B13 band region $F_{B4,T/2}$ is always larger than $F_{T/2}(\nu)$. Thus, positive asynchronous cross-peaks will occur. However, since the difference between $F_{B4,T/2}$ and $F_{T/2}(2210 \text{ cm}^{-1})$ is small, $\Psi'(\nu_1, \nu_2)$ will be small. Thus, the asynchronous correlation intensity $K_1 K_2 \Psi'(\nu_1, \nu_2)$ at 2210 cm^{-1} will be weak. The correlation of band B4 with the overlapping B12/B13 bands generates two positive asynchronous cross-peaks, with a minimum between them at 2210 cm^{-1} .

Overlapping band resolution

It can be seen in Fig. 5 that all of the overlapping bands are clearly resolved in correlations with the B2 and B3 bands. In contrast, the correlation with band B5 does not give clear resolution. Good resolution requires that the NHI of a single correlating band have a value between the maximum and minimum of the NHI of the overlapping correlating bands. These observations suggest a useful 2D COS method to analyze highly overlapping bands. Spectral resolution would be enhanced by correlating complex spectra with simulated single band spectra with variable intensity change rates.

2D correlation analysis of spectra simulated using Lorentzian function

We performed a similar study using Lorentzian functions with the same band parameters. While the contour shape of 2DCOS spectral peaks change because of the differing Lorentzian bandshape, most of the Gaussian band 2D COS features remain. The Lorentzian 2D COS spectral features can be explained identically as for the Gaussian bandshapes using the total intensity changes and normalized half-intensities.

CONCLUSIONS

Our study shows that spectral normalized intensities are important in determining the 2D COS spectral intensities. The signs and number of 2D cross-peaks are related to the normalized half-intensity (NHI) differences and the total intensity changes of the correlated bands. These insights into the origin of the 2D spectral features will help interpret the 2D asynchronous COS spectra. Our results indicate a method to help resolve overlapping bands. This method would correlate the overlapping band regions with a series of discrete bands of different intensity change rates.

Acknowledgments

We gratefully acknowledge Isao Noda for useful discussions. This work is supported by NIH Grant No. 1R01EB009089 (S.A.A.) and NSF Grant No. CHE-1152752 (I.K.L.).

ABBREVIATIONS USED

2D COS	Two-dimensional correlation spectroscopy
NHI	Normalized half-intensity

LIST OF SYMBOLS

A_i	Maximum amplitude of the i th Gaussian band
f	An intensity change coefficient matrix
f_i	Intensity change coefficient of the band B_i
f_{ij}	Intensity change coefficient of the i th band for the j th time step
$F_{i,j}(\nu)$	Normalized intensity of a single Gaussian band i at the j th time step
$F_j(\nu)$	Normalized intensity at j th time step at frequency ν
$F(\tilde{\nu}_i)$	Normalized dynamic spectral matrix
$F_{B_i T/2}$	Normalized half-intensity of B_i band
$F_{T/2}(\nu)$	Normalized half-intensity at frequency ν
$I_0(\nu)$	Intensities matrix of simulated Gaussian bands at frequency ν
$I_{i,0}(\nu)$	Intensities of simulated Gaussian band i at frequency ν
$I(\nu, t)$	Intensity of simulated spectra at frequency ν and at time t
k_i	Intensity change rate constant of band B_i
K_i	Total intensity change of spectra during the perturbation at ν_i
m	Number of spectra collected over perturbation
w_i	Full band width at half height = $2(2\ln 2)^{1/2}w_i$
$y(\nu_i, t_j)$	Measured spectral signal at ν_i and time t_j
$\mathcal{Y}(\nu_i)$	Dynamic spectral intensity matrix
$\mathcal{Y}(\nu_i, t_j)$	Dynamic spectral signal at ν_i and time t_j
λ_i	Total change of intensity change coefficient of band B_i during the perturbation period
ν	Frequency
ν_{ci}	Gaussian center frequency
$\Phi(\nu_1, \nu_2)$	Synchronous correlation intensity
$\Phi'(\nu_1, \nu_2)$	Synchronous correlation intensity of normalized spectra
$\Psi(\nu_1, \nu_2)$	Asynchronous correlation intensity
$\Psi'(\nu_1, \nu_2)$	Asynchronous correlation intensity of normalized spectra

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Figure 1.
(A) 13 Gaussian band spectra. (B) Simulated spectra.

Figure 2.

(A) Time dependence of intensities of bands $I_A(t) = e^{-0.2t}$, $I_B(t) = 0.4e^{-0.01t} + 0.6e^{-0.27t}$ and $I_C(t) = e^{-0.05t}$. (B) Normalized intensity time dependence of these bands. $F_{A,T/2}$, $F_{B,T/2}$ and $F_{C,T/2}$ refer to the NHI of these bands. The black oval shows the region where the normalized intensities of bands A and B cross.

Figure 3.
 (A) Synchronous and (B) Asynchronous spectrum calculated from the simulated spectra of Fig. 1B. The dashed horizontal lines indicate the Gaussian band maxima.

Figure 4.

(A) Frequency dependence of normalized spectral intensities $F_j(v)$ at different time steps. (B) NHI $F_{T/2}(v)$ frequency dependence of the simulated spectra. The five horizontal lines indicate the NHI of bands B1 to B5. The thick black vertical bars indicate spectral regions where the time dependence of $F_{B3,j}$ and $F_j(v)$ cross. (C) Time dependence of B3 band $F_{B3,j}(1570 \text{ cm}^{-1})$, and of $F_j(v)$ at 2162, 2202 and 2210 cm^{-1} . The green arrow indicates the direction of increasing frequency from 2162 to 2210 cm^{-1} . (D) NHI $F_{T/2}(v)$ frequency dependence overlaid on the simulated Gaussian bands. The shaded bars indicate frequency regions where bands do not overlap, where single bands dominate.

Figure 5.

Asynchronous 2D correlation spectrum and the NHI frequency dependence, $F_{T/2}(\nu)$. $F_{T/2}(\nu)$ along left vertical axis (red curve) overlaid with the difference spectrum between the simulated spectra at $t=0$ and $t=20$ sec ($-K(\nu_2) = I(\nu_2, 0) - I(\nu_2, 20)$), blue line) in the overlapping band regions. The four horizontal blue shaded areas indicate regions of small $-K(\nu_2)$ values. O_1 and O_2 label the edges of areas 3 and 4. The black vertical line indicates the value of the NHI of the B3 band, $F_{B3,T/2}$. Points p1 to p8, along the black vertical line indicate frequencies where $F_{B3,T/2} \approx F_{T/2}(\nu)$. p1=1882 cm⁻¹, p2=1952 cm⁻¹, p3=1994 cm⁻¹, p4=2046 cm⁻¹, p5=2112 cm⁻¹, p6=2128 cm⁻¹, p7=2202 cm⁻¹, p8=2216 cm⁻¹. The top horizontal spectrum shows the B1 to B5 band difference spectrum between $t=0$ and $t=20$ sec ($-K(\nu_1) = I(\nu_1, 0) - I(\nu_1, 20)$).

Table 1

Gaussian band parameters and the band intensity change functions.

	$f(t)$	$\nu_c(\text{cm}^{-1})$	A	$w(\text{cm}^{-1})$
B1	$e^{-0.05t}$	1370	100	14
B2	$e^{-0.085t}$	1470	100	14
B3	$e^{-0.1t}$	1570	100	14
B4	$e^{-0.15t}$	1670	100	14
B5	$e^{-0.2t}$	1770	100	14
B6	$e^{-0.05t}$	1870	100	14
B7	$e^{-0.2t}$	1900	100	14
B8	$e^{-0.05t}$	2000	100	14
B9	$e^{-0.2t}$	2008	100	11
B10	$e^{-0.05t}$	2100	100	14
B11	$e^{-0.2t}$	2108	36	11
B12	$e^{-0.05t}$	2200	100	14
B13	$e^{-0.2t}$	2208	100	5