Generalized Implementation of Rapid-Scan Fourier Transform Infrared Spectroscopic Imaging

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We describe a novel, generalized data acquisition sequence to allow rapid-scan Fourier transform infrared (FT-IR) spectroscopic imaging using focal plane array (FPA) detectors. This technique derives its applicability from the reproducible performance of modern FT-IR instrumentation and the availability of FPAs with simultaneous, full array acquisition, or snapshot electronics. Instead of sampling the entire interferogram in one mirror sweep over a predetermined retardation, as in traditional continuous-scanning techniques, the modulated light from the interferometer is recorded over several mirror sweeps. The FPA detector is synchronized for data acquisition after a specified delay with respect to the initiation of the mirror motion to provide a highly under-sampled interferogram. By incorporating appropriate delays in subsequent interferometer mirror scans, the entire interferogram is sampled and reconstructed. The signal-to-noise ratios (SNR) of the resulting interferograms are analyzed and are compared with step-scan spectroscopic imaging data.

Index Headings: **Rapid-scan, FT-IR; Spectral imaging.**

INTRODUCTION

A Fourier transform infrared (FT-IR) interferometer coupled to a focal plane array (FPA) detector represents the next generation of infrared spectroscopy.[1] As with many instrumental innovations, there is a gestation period between commercial availability and the proliferation of reliable, integrated instrumentation. For FT-IR spectroscopic imaging instrumentation, this period is exacerbated by two related factors; first, the cost of a commercial spectral imaging system incorporating expensive FPA detectors is significantly higher than that of an FT-IR spectrometer employing single element detection, and second, data acquisition times for array detectors are greater than that of single-element detectors, thus necessitating the use of expensive step-scan interferometers. These large data acquisition times, coupled with the inherent, relatively poor signal-to-noise (SNR) characteristics of FPA equipped systems, result in only moderate quality data, which are nevertheless acquired orders of magnitude in time faster than single element detectors used in comparable sample mapping experiments.[2] Improvements in spectral imaging performance may be furthered by incorporating faster detectors;[3] however, such advances are rare and economically nonviable for researchers already possessing FPA detectors. Additionally, improvements may be achieved at the interferometer stage using instrumental modifications and novel data acquisition methodologies to reduce data acquisition times and to improve SNR characteristics.[4–7]

While data acquisition modification attempts have been successful, they have focused primarily on focal plane array detectors coupled to step-scan interferometers.[3,4,7] Less expensive, continuous-scan interferometry coupled to detectors possessing fast readout electronics has been suggested as a method for acquiring data faster while reducing instrumentation costs.[8] Such schemes, however, are not generally applicable to all focal plane arrays, specifically those that are not equipped with fast electronics or those whose large numbers of pixels require longer readout and storage times. To the best of our knowledge, continuous-scan FT-IR spectroscopic imaging with either small format[8] or large format detectors[9] has not been implemented in more than a few laboratories. Recently, commercial, rapid-scan imaging instruments have become available using small linear (16×1)[10] or two-dimensional (16×16 and 32×32)[11] arrays. While small detectors allow rapid-scan imaging, the large multichannel detection advantage, such as that for a 256×256 array, is lost. Further, there is a restrictive upper limit to the mirror scanning velocity, which is determined by the FPA characteristics and limits the Fourier frequencies attained in a continuous-scan experiment.[12]

Previous studies reporting the implementation of continuous-scan imaging were, thus, restricted to mirror scanning speeds of 0.0158 cm/s for a 64×64 array[8] and 0.00625 cm/s for a 256×256 array.[9] In this manuscript, we present a generalized implementation of continuous-scan infrared spectroscopy that can be applied to a large number of detector array sizes and interferometer scanning velocity combinations. Furthermore, this implementation allows scanning rates suitable for rapid-scan imaging, that is, for scanning velocities such that the Fourier frequencies are in the audio range.[13] In general, continuous-scan interferometry is said to be rapid-scanning when the moving-mirror velocity $v_m \geq 0.01$ cm/s, and slow-scan if $v_m \leq 0.01$ cm/s. This translates into Fourier

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frequencies of ~20 Hz, the lower audio limit, at 1000 cm^{-1}, the lower wavenumber cutoff of many mid-infrared FPA detectors commercially available. However, to modulate at audible Fourier frequencies throughout the entire mid-infrared with a lower cutoff of ~400 cm^{-1}, the scanning velocity for rapid-scan interferometry must be at least 0.025 cm/s. To this date, this lower limit has only been exceeded by very small format multichannel detectors.[10,11]

THEORY

Quantification of the quality of data measured by FT-IR spectrometers, in general, and FT-IR imaging spectrometers, in particular, can be carried out in terms of the SNR, wavenumber precision, and reproducibility. For an interferometer providing acceptable wavenumber accuracy and reproducibility, the SNR is the primary assessment metric of the quality of data produced by a particular instrumental configuration. High SNR is often a requirement for identifying subtle spectral differences, especially when applying sophisticated analyses to the large volumes of data generated in an imaging experiment. Generally, step-scan imaging spectrometers are employed as their wavenumber accuracy and reproducibility reflects stable interferometer operation and an SNR that can be easily increased by FPA frame and data set averaging.

Step-Scan Interferometry. In step-scan interferometry, the optical retardation achieved by the mirror movement in the interferometer is held constant for a predetermined time period and then rapidly advanced to the subsequent retardation position. In this manner, the interferogram is built point by point by moving the mirror, stabilizing its retardation, and then acquiring data. Optimal step-scan spectroscopic imaging experiments are designed around the characteristics of an FPA.[7] The two determinants of FPA performance relevant to step-scan imaging are the frame rate, or the inverse of the time required to capture one FPA frame, and the time required to first read, and then store the acquired data for each interferometer retardation step, which is termed the readout time. The interferometer mirror settling time is incorporated into the readout time of the array; hence, the smaller of the interferometer settling time or the array readout time plus the FPA integration time represents the fastest that the interferogram can be sampled. The time required to obtain an interferogram data set using step-scan interferometric imaging may be described by Eqs. 1 and 2

$$t = t_s n_s \tag{1}$$

$$t_s = t_d + t_f n_f \tag{2}$$

where t is the total time to acquire one interferogram data set, t_s is the sample time per data point in the interferogram, n_s is the number of interferogram data points, t_f is the reciprocal of the frame rate of the FPA, and n_f is the number of frames coadded. A consequence of utilizing the step-scan method for spectral imaging is that it introduces an additional delay time, $t_d n_s$, into the acquisition. Regardless of this disadvantage, step-scan interferometry is commonly utilized in spectral imaging, since it provides acceptable performance for many applications, for example, diffusion processes in polymeric materials[14] and studying the histopathological changes in heterogeneous tissues.[15–17] For routine spectroscopic examinations, however, step-scan spectroscopic imaging is a far from ideal method of data acquisition.

Rapid-Scan Interferometry. An ideal spectroscopic imaging system should allow the FPA sufficient time to acquire high quality data, while not contributing additional delays to the overall process. Continuous-scan interferometry, in contrast to step-scan systems, does not require a delay time for the stabilization of the moving mirror since the mirror motion is constant. While continuous-scan data acquisition is more efficient, the advantages of frame coaddition, enjoyed in step-scan spectral imaging, are mitigated as mirror positioning errors become contributors to overall noise. Consequently, it has been proposed that only one frame be coadded for every retardation sampling point, which again precludes taking advantage of frame coaddition and requires complete image coaddition to achieve a desired SNR.[8] Hence, compared to step-scan interferometry, the increase in efficiency derived from the elimination of mirror stabilization times is reduced by the loss in efficiency due to the multiple mirror accelerations, decelerations, and directional changes required in continuous-scan interferometry to achieve an acceptable SNR. The primary advantages provided by continuous-scan are a savings in cost, the opportunity to subject the detector to high modulation frequencies, which may improve SNR characteristics, and the speed of data acquisition. As previously noted, however, the readout rates of the FPAs currently available do not allow true rapid-scanning unless a large multichannel detection advantage is sacrificed.[10,11] In particular, conventional continuous-scan data acquisition methods applied to the moderately large format arrays available today (for example, a 256 × 256 MCT) simply do not allow rapid-scan imaging, as the array sampling frequency (frame rate) is not high enough even at the minimum mirror scan rates set to achieve rapid-scan spectroscopy.

Generalized Rapid-Scan Interferometry. To address the FPA induced limitations for rapid-scan data acquisition, we describe a generalized data acquisition scheme that takes advantage of the stable performance of a modern rapid-scan interferometer. This approach can be applied either to conduct rapid-scan imaging while employing a detector of virtually any size with variable readout characteristics or to provide greater interferometric scan rates compatible with smaller arrays. Essentially, this method employs a repeated undersampling of the interferogram for time intervals determined by the focal plane array frame rate. Thus, mirror motion and detection are decoupled such that mirror retardation changes and subsequent array detection are synchronized by an external clock.

To illustrate our approach, the curve shown in Fig. 1a represents a waveform to be sampled in which the open squares indicate the fastest sampling capability of the detector. If the same curve were to be sampled by a slower detector (assume, half as fast as the detector in Fig. 1a), then the sampling frequency would be reduced by half. As a consequence, the detector would only be able to sample every second point compared to the fast detector, as shown by the squares in Fig. 1b. If the same waveform were repeated a second time and the detector were ca-

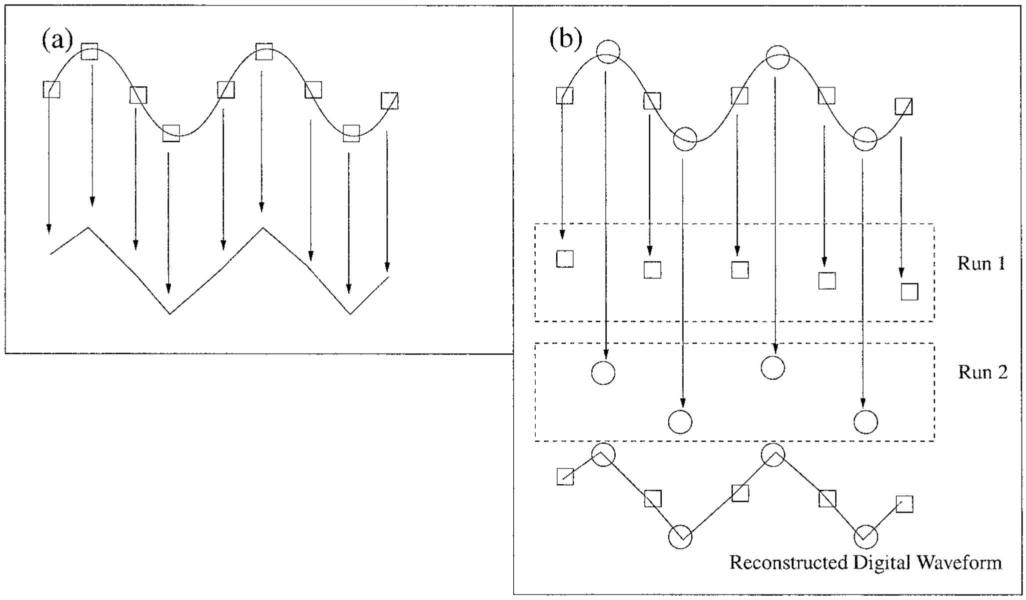

FIG. 1. (**a**) Traditional sampling is when the waveform is sampled completely in one interferometer pass. (**b**) Iterative sampling is when the waveform is undersampled per scan and subsequently assembled into a completely sampled waveform.

pable of being appropriately delayed before it acquired the first data point, a reduced sampling frequency could be employed to access the curve between the points sampled in the previous run, shown as circles in Fig. 1b. Therefore, by employing a slower detector with an appropriate delay mechanism, the invariant waveform can be completely and appropriately sampled, as indicated in Fig. 1b.

For continuous-scan spectroscopy at any speed, the interferometric laser fringe pattern (a sinusoid) is employed to sample the detector signal at every ith zero crossing of the fringe waveform. In repeated mirror sweeps, approximately the same laser fringe pattern is obtained, thus allowing for the waveform to be sampled at predictable intervals. For many large FPA detectors, or FPA detectors with slow readout electronics, the required sampling is not possible in one mirror scan. By employing a multiple pass scheme, as depicted in Fig. 1b, the detector signal at any desired retardation interval may be sampled. For each observation of the laser fringe signal, the undersampling of the detector readout is given by a factor, n_R, defined as $n_R = \Delta/t_f v n_f$, where Δ is the total retardation of the moving mirror for a given resolution. In the limit that the frame rate is sufficiently large, the generalized equation yields $n_R = 1$, as in the traditional rapid-scan approach.

EXPERIMENTAL

A Bruker IFS66/s FT-IR spectrometer (Bruker, MA) coupled to a microscope (IR Scope II, Bruker) equipped with a 256 × 256 element HgCdTe focal plane array (Santa Barbara Focalplane, CA) is employed for all measurements. The interferometer can be operated in either step-scan or continuous-scan modes, for which the retardation dwell time or scanning velocity can be arbitrarily adjusted. Several 15× Cassegrain beam condensing optics were employed to image an area about 750 × 750 μm on a sample. In rapid-scan mode, the acquisition of the signal was initiated with the start of the forward motion of the mirror. The measurement at each retardation point was not locked in to the laser signal, as with conventional rapid-scan operation, since the interferometer operation was found to be suitably stable with respect to the high detector noise levels. For step-scan operation, the time per retardation step was varied to incorporate mirror stabilization, frame, and readout times. An oscilloscope (Tektronics) and function generators (Agilent Technologies, CA) were used to monitor and to trigger data acquisition.

For rapid-scan measurements, the interferometer was set to scan the moving mirror at 0.025 cm/s, which allows for Fourier frequencies in the audible range over the entire mid-infrared spectral region. For the step-scan measurements, the moving mirror was allowed to stabilize for 40 ms before acquisition was initiated. An additional 250 ms/step was allowed to acquire data. The FPA frame rate was set at 114 Hz with an integration time of 0.0290 ms. The interferogram was sampled at every fourth HeNe laser crossing to yield interferograms consisting of 512 points over the region 0–3950 cm^{-1} for a nominal spectral resolution of 16 cm^{-1}. The interferograms were Fourier transformed using triangular apodization to yield single beam spectral profiles. A thin film (~7 μm) polystyrene sample (obtained from ICI, UK) was clamped between two BaF_2 windows and imaged.

A complete sampling at every fourth crossing of the laser of the interferogram or 10 ms time interval is carried out using four iterations of a data acquisition sequence, as depicted in a portion of the interferogram shown in Fig. 2. After all iterations of the mirror scan were completed, the digital waveform corresponding to the interferogram was reconstructed by interleaving the individually acquired sequences into an equivalent curve, as illustrated in Fig. 2. While we have arbitrarily selected a mirror speed to demonstrate this concept by dividing the sampling of the interferogram over four mirror scans,

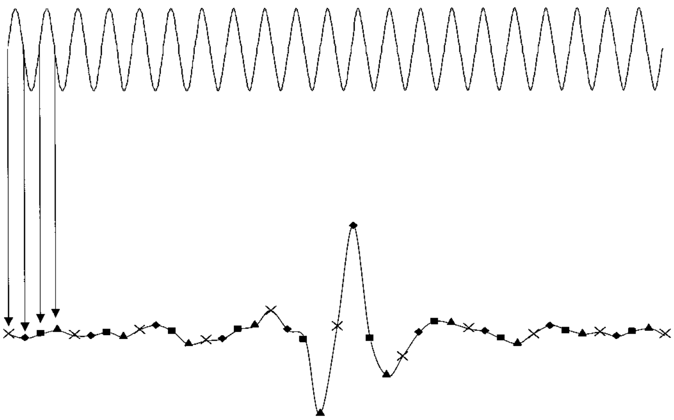

FIG. 2. The illlustration shows the reconstruction of iteratively, undersampled interferograms where points ×, ♦, ■, and ▲ represent sequentially acquired waveforms.

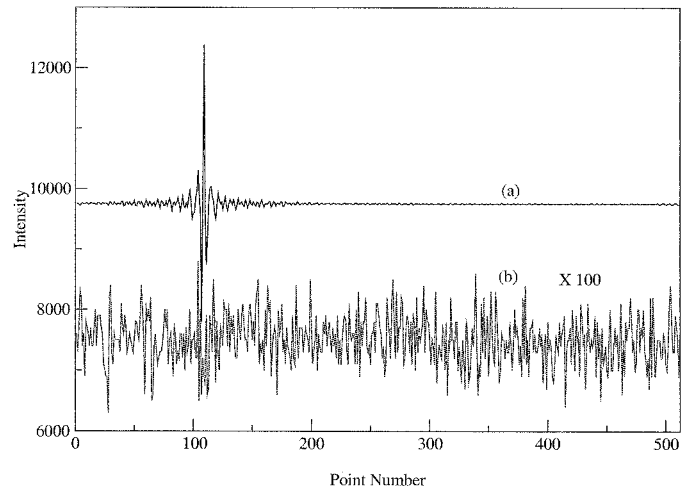

FIG. 3. Plots of (**a**) the assembled interferogram sampled with four passes and (**b**) the subtracted sequential interferograms offset by 7500 intensity units.

an interferogram at other mirror scanning rates can be measured in a straight-forward manner. Since four iterations were employed in the example of an FPA sampling rate of 100 Hz, the second, third, and fourth iterations were delayed by 2.5, 5.0, and 7.5 ms, respectively.

Root mean square (rms) noise and SNR calculations were performed on single beam spectra between the 2000 and 2233 cm^{-1} region. The signal was measured at $\bar{\nu}_o$ = 2110 cm^{-1} with the noise being determined at ±5 resolution elements around $\bar{\nu}_o$. All processing was performed using IDL/ENVI routines written in-house on a PC.

RESULTS AND DISCUSSION

Image data sets acquired by employing the rapid-scanning generalized approach displayed no gross distortions, as the data points corresponded to similar interferograms determined by step-scan interferometry. Specifically, randomly selected interferograms from multiple image data sets (32 in number) did not appreciably differ from corresponding interferograms (from the same pixel) extracted from any other data set. This robust procedure is not prone to producing errors in the data acquisition. Any errors arising, however, from interfacing the interferometer to the FPA would be the same for all pixels. Therefore, instead of considering entire images, it is more instructive to examine an average spectrum from a central portion of the image in which pixel operability is the highest. The random component of the FPA noise is reduced in the average spectrum, and thus, errors in the average spectrum offer a stricter test for interferometric errors. A representative interferogram is shown in Fig. 3a. Average interferograms from the central 32 × 32 pixels of two randomly selected data sets were subtracted to enhance any systematic noise. As shown in Fig. 3b, the difference spectrum indicates that the variations in data sets are limited to random noise.

In most reported imaging experiments, the noise contribution from the detector was considered to be random and dominant. The next largest source of noise in a continuous-scan imaging system has been postulated to arise from the change in mirror position during the sampling of the modulated light over every retardation element.[8,12] Thus, for rapid-scan spectroscopy the total noise combines both FPA noise and noise due to positioning that may occur during sampling of the interferogram, as determined by Eq. 3:

$$SNR_{max} = \frac{4}{\Delta l \bar{\nu}_{max}} \quad (3)$$

where the error in mirror position is given by Δl and the maximum of the spectral range by $\bar{\nu}_{max}$. While there are other sources of noise that occur in rapid scan spectroscopy,[13] they are orders of magnitude smaller than the two described above. For example, additional noise can arise if the mirror velocity is not constant. To assess this aspect, we measured the laser fringe pattern as a function of time and did not find deviations greater than experimental error between successive patterns. Another noise source arises from instrumental drift in, for example, the source, beamsplitter, detector, and in temperature between interferometer passes; this is expected to be negligible in the approximately 10 s required to acquire a completely sampled interferogram.

Clearly, only the variation in mirror position during the integration time is significant for determining positioning errors.[12] As a result, it may be suggested that a low integration time be employed to reduce the positioning error while acquiring data at the highest possible frame rate. Since the signal is directly proportional to the integration time, a reduction in integration time will result in lower noise and lower signal, which may affect the SNR of the acquired data. Total noise is the sum from sources that cause noise to increase proportionally to the integration time (positioning errors) and that contribute noise only as the square root of the integration time (random FPA noise). Consequently, the SNR varies in a complex manner with regard to the integration time employed. By changing the integration time, we did not find any nonlinear degradation in the SNR, which is probably due to the noise of the detector being dominant for a single frame. From this observation one can deduce the largest integration time that is allowed by the dynamic range employed. As an example, when the signal is sampled for a duration of 0.0430 ms, the mirror has traveled a distance of 10.75 nm for a mirror velocity of 0.025 cm/s. The variation in mirror position is much larger than

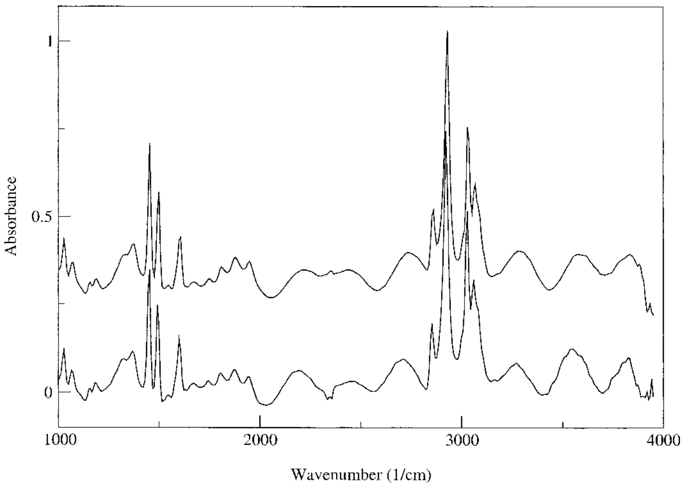

FIG. 4. Spectra of polystyrene: rapid-scan (lower trace) and step-scan (upper trace).

the typical variation from a step-scan interferometer (0.1–0.5 nm). Thus, the error due to variation in mirror positioning is expected to be twenty to one hundred times greater. However, calculating the resulting SNR from Eq. 3, one notes that the predicted limiting SNR for mirror positioning errors is ~750, which is much larger than the SNRs actually achieved. Therefore, it is reasonable to conclude that the net noise is random and the usual practice of coadding image cubes or time-averaging data is expected to be effective.

Figure 4 displays both step- and rapid-scan spectra of polystyrene, spectra which are virtually identical. The accuracy of the wavenumber scale and the accuracy of the spectral response determination of the generalized rapid-scan approach can thus be established. A comparison of spectral features shows no loss of resolution, and an examination of the difference spectrum between the two results only in noise. The reproducibility of the multipass interferometric sampling method is demonstrated by difference spectra obtained from consecutive interferogram measurements. The residual differences between consecutive step-scan and rapid-scan measurements consist only of noise and are of the same levels (data not shown). The residuals show only nonstructured, random noise, suggesting that data acquired using iterative interferometric sampling is at least as reproducible as data from conventional step-scan interferometry.

The total time for the acquisition for a 1024 element interferogram image in the rapid-scan mode is 10.2 s for a four iteration sampling method using a mirror scanning velocity of 0.025 cm/s. This is less than the time for obtaining a comparable image using the fast step-scan mode (~15 s).[3,12] There is a loss in efficiency for step-scan interferometry when the mirror is stepped, since a stabilization period is required at each retardation element. Similarly, for the generalized rapid-scan approach, there is a loss of efficiency due to multiple mirror direction changes involving deceleration to rest and accelerations, as noted above; however, the total delay time due to stepping at each retardation is greater than the turn around time as implemented in this study. We have implemented rapid-scan interferometry with data acquisition only in the forward scan direction. For increased efficiency, data acquisition may also occur when the mirror velocity is reversed, although phase errors that may be introduced would be difficult to correct since a complete interferogram would not have been acquired in either direction. Reversing the mirror at the highest possible speed alleviates the loss in efficiency due to mirror travel in the opposite direction.

CONCLUSION

An iterative data acquisition scheme that allows almost any detector array to be operated in conjunction with a rapid-scan interferometer has been proposed and implemented. This approach has allowed, for the first time, true rapid-scan imaging data to be recorded from a large format array. The primary noise source in this implementation is random noise from the FPA, allowing the usual trade off of rapid-scan spectroscopy to be applied. In comparison to data acquisition using the step-scan methodology for imaging, the rapid-scan approach, despite its inherent inefficiency, was found to be superior in terms of the required time for obtaining a single image cube, thereby increasing the scope for monitoring dynamic phenomena in real-time. The same technique can be employed to achieve higher mirror scanning velocities for small format arrrays. Hence, the application of a generalized method of rapid-scan imaging, as demonstrated in this manuscript, will allow the full advantages of rapid-scan interferometry to be achieved for almost any desired experimental conditions with any detector array.

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