

Characterization of Octadecylsilane Stationary Phases on Commercially Available Silica-Based Packing Materials by Raman Spectroscopy

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Two commercially available solid phase extraction packing materials containing octadecylsilane stationary phases, Isolute C18MF and Isolute C18, were characterized using Raman spectroscopy. Raman spectra of excellent quality can be obtained from such systems and provide direct information about the alkyl chain conformation in the stationary phase. Data from the $\nu(\text{C}-\text{C})$ and $\nu(\text{C}-\text{H})$ spectral regions suggest that the alkyl chains of these stationary phases exist in a highly disordered state which is very similar to that of the neat liquid.

Chemically modified silicas are popular stationary phases for reversed phase liquid chromatography and solid phase extraction (SPE).^{1–3} These stationary phases are typically formed by covalent attachment of alkylchlorosilanes to a silica gel support using mono-, di-, or trichlorosilanes.³ Chromatographic materials made from di- and trichlorosilanes are referred to as “polymeric” stationary phases, whereas those made from monochlorosilanes are known as “monomeric” stationary phases. The properties of these stationary phases play an important role in governing the selectivity and efficiency of separations.^{2–5} To elucidate the retention mechanisms for such separations, an understanding of the stationary phase behavior under various separation conditions is essential. Several experimental techniques, including fluorescence spectroscopy,^{6–12} NMR,^{13–18} IR spectroscopy,^{19–22} and

chromatographic methods,^{23–26} have been used to study these stationary phases. Despite the valuable information provided by these studies, a complete understanding of retention has not been achieved. This deficiency is due to a lack of understanding of the intermolecular interactions between the alkyl moieties of these stationary phases, the solutes, and the mobile phases in the retention process. This information can only be provided by investigating the stationary phase using techniques that are sensitive to alkyl chain conformation.

Raman spectroscopy is a powerful tool for the characterization of conformational changes in alkyl chains.^{27–29} Previously, its application for the study of silica gel-based stationary phase systems has been limited by the very weak Raman scattering of alkylsilane stationary phases and the large background scattering arising from the silica particles. Recent advances in instrumentation, such as the use of charge-coupled device detectors³⁰ and better rejection of background scattering,^{31–33} have greatly improved the sensitivity of this technique, however. In this paper, we present the first demonstration of Raman spectroscopy for the characterization of commercially available, silica-based alkylsilane stationary phases. Two commercial SPE packing materials containing octadecylsilane stationary phases, Isolute C18MF and Isolute C18, are chosen as examples for this demonstration. The two main objectives of this study are as follows. First, we seek to investigate the conformational behavior of the alkyl chains of these stationary phases. Second, we hope to demonstrate the feasibility of using Raman spectroscopy to characterize real chromatographic stationary phases.

EXPERIMENTAL SECTION

Dimethyloctadecylchlorosilane (DOS, >97%) was obtained from United Chemical Technologies, Inc., and octadecyltrichlo-

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Table 1. Manufacturer's Specifications for C18MF and C18TF

	C18MF (monomeric)	C18TF (polymeric)
surface area (m ² /g)	300	561
average pore size (Å)	60	54
carbon loading (C%)	13.9	19.3
surface coverage (μmol/m ²)	1.9	1.6
average particle diameter (μm)	63	62

rosilane (OTS, 95%) was purchased from Aldrich. Both chemicals were used as received. Raman spectra were collected using 100 mW of 514.5 nm radiation from a Coherent Innova 90-5 Ar⁺ laser on a Spex 1877 Triplemate spectrograph as described previously.^{27–29} The detector in these experiments was a Princeton Instruments charge-coupled device (CCD) system based on an RTE-1100-PB CCD of pixel format 1100 × 330, cooled with liquid N₂ to –120 °C. Sealed NMR tubes were used as sample holders for the acquisition of Raman spectra on the chromatographic packing materials. Integration times for each spectrum are provided in the figure captions.

The two commercial SPE stationary phases, Isolute C18MF (C18MF) and Isolute C18 (C18TF), were a gift from International Sorbent Technology. C18MF is a monomeric stationary phase, and C18TF is a polymeric stationary phase. The manufacturer's specifications of these materials, including surface coverage of the stationary phase, are listed in Table 1. Both packing materials have a similar average particle size (~60 μm) and average pore size (~60 Å). Typically, polymeric phases have higher surface coverages of the alkyl moieties than those of the monomeric stationary phase. However, for these particular materials, both phases have similar alkyl chain surface coverage (1.9 μmol/m² for C18MF and 1.6 μmol/m² for C18TF) according to the manufacturer. It should also be noted that results from the Raman spectroscopic characterization (below) indicate that both C18MF and C18TF exhibit similar alkyl chain conformation; therefore, the results of their characterization are discussed together.

RESULTS AND DISCUSSION

Spectral results from two regions, 800–1400 (ν(C–C) region) and 2700–3100 cm^{–1} (ν(C–H) region), are presented here. These regions contain vibrational modes that are sensitive to the conformation of the alkyl chains. Raman spectra of crystalline and liquid dimethyloctadecylchlorosilane and octadecyltrichlorosilane are also provided for comparison. DOS is the precursor for synthesis of the monomeric stationary phase (i.e., C18MF), whereas OTS is the precursor for the polymeric phase (i.e., C18TF).

Figure 1 shows Raman spectra between 800 and 1400 cm^{–1} for (a) neat liquid DOS, (b) neat liquid OTS, (c) crystalline DOS, (d) crystalline OTS, (e) C18MF, and (f) C18TF. The bands of interest in this region are the CH₂ rk_G, CH₃ rk_T, CH₃ rk_G, ν(C–C)_T, ν(C–C)_G, CH₂ twist_G, and CH₂ twist_T vibrational modes (where the subscripts G and T refer to gauche and trans conformers, respectively). Peak frequencies and the band assignments are listed in Table 2. The assignments are based on those previously reported for alkyl chain systems.^{34,35}

The first point to be made about the spectra for C18MF and C18TF in Figure 1 is their very high quality in terms of signal-

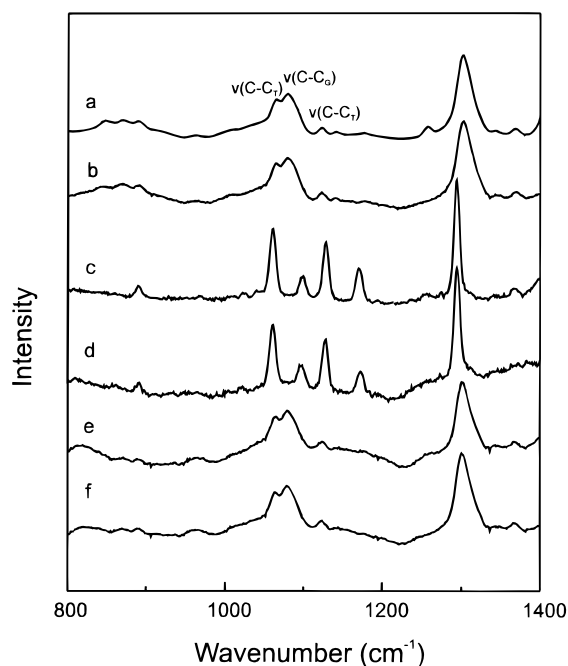


Figure 1. Raman spectra in ν(C–C) region for (a) neat DOS, (b) neat OTS, (c) crystalline DOS, (d) crystalline OTS, (e) C18MF, and (f) C18TF. Integration times: (a) 30 s, (b) 30 s, (c) 10 s, (d) 20 s, (e) 30 min, and (f) 30 min.

to-noise ratio. The observation of spectra of this quality was surprising based on our earlier attempts to study these systems³⁶ and based on past work on chemically modified silica systems (either by adsorption or covalent attachment³⁴). Furthermore, based on the completely transparent nature of silica at this excitation wavelength, we believe that the Raman response originates from octadecylsilane chains bound both to the outer surface of the silica support and within the pores of the silica support. Thus, the total response represents the composite of alkyl chain environments existing for the stationary phase in these systems.

Neat liquid DOS and OTS represent the more disordered state of the alkyl chain due to the existence of a variety of conformations in the liquid state. This disorder is indicated by the large intensity of bands associated with gauche conformers relative to trans conformers in the alkyl chain. Specifically, the large number of gauche conformers is indicated by the relatively large ν(C–C)_G intensity at ~1080 cm^{–1} in the Raman spectra (Figure 1a,b). Further evidence for the disordered nature of the alkyl chains is shown by the breadth and asymmetry of the CH₂ twist_{T,G} at 1302 cm^{–1} and the presence of the CH₂ rk_G mode at ~847 cm^{–1}. In the crystalline state (Figure 1c,d), the alkyl chains adopt a more ordered conformation, as evidenced by the increase in relative intensity of the two ν(C–C)_T bands at ~1061 and 1129 cm^{–1}, while the bands associated with G conformations, such as the ν(C–C)_G, and CH₂ rk_G, disappear from the spectra. In addition, the CH₂ twist_T at ~1294 cm^{–1} also increases in intensity, shifts 10 cm^{–1} to lower frequency, and becomes narrower and more symmetric.

In the Raman spectra of both commercially available packing materials, C18MF (Figure 1e) and C18TF (Figure 1f), a large

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Table 2. Raman Frequencies and Assignments for Different Alkylsilane Environments

neat DOS (cm ⁻¹)	crystalline DOS (cm ⁻¹)	neat OTS (cm ⁻¹)	crystalline OTS (cm ⁻¹)	C18MF ^a (cm ⁻¹)	C18TF ^b (cm ⁻¹)	assignment
2978	2979					$\nu_a(\text{CH}_3)^c$
2965	2962	2963sh ^d		2963	2963	$\nu_a(\text{CH}_3)$
2931sh	2929sh	2930sh	2935sh	2930sh	2933	$\nu_s(\text{CH}_2, \text{FR})$
2901	2902		2907	2899	2892	$\nu_a(\text{CH}_3)$
2886sh	2880	2891	2881	2885sh	sh	$\nu_a(\text{CH}_2)$
2852	2846	2852	2847	2855	2852	$\nu_a(\text{CH}_2)$
1302	1294	1303	1294	1301	1301	$\text{CH}_2 \text{ twist}_{\text{T,G}}^{e,f}$
1177	1170		1172			
1123	1129	1123	1128	1125	1123	$\nu(\text{C}-\text{C})_{\text{T}}$
	1099		1096			$\text{CH}_2 \text{ twist}_{\text{T}}$
1080		1080		1081	1081	$\nu(\text{C}-\text{C})_{\text{G}}$
1065	1061	1065	1061	1064	1064	$\nu(\text{C}-\text{C})_{\text{T}}$
890	889	890	891			$\text{CH}_3 \text{ rk}_{\text{T}}^g$
869		868				$\text{CH}_3 \text{ rk}_{\text{G}}$
847		843				$\text{CH}_2 \text{ rk}_{\text{G}}$

^a C18MF, Isolute C18MF, monomeric octadecylsilane stationary phase. ^b C18TF, Isolute C18, polymeric octadecylsilane stationary phase. ^c ν , stretch. ^d sh, shoulder. ^e T, trans. ^f G, gauche. ^g rk, rock.

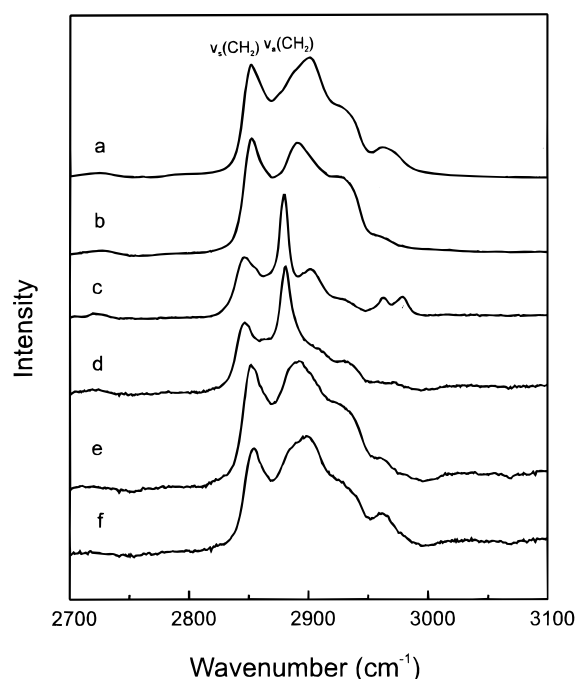


Figure 2. Raman spectra in $\nu(\text{C}-\text{H})$ region for (a) neat DOS, (b) neat OTS, (c) crystalline DOS, (d) crystalline OTS, (e) C18MF, and (f) C18TF. Integration times: (a) 20 s, (b) 20 s, (c) 4 s, (d) 10 s, (e) 4 min, and (f) 4 min.

intensity of the $\nu(\text{C}-\text{C})_{\text{G}}$ band at 1081 cm⁻¹ and a broad asymmetric $\text{CH}_2 \text{ twist}_{\text{T,G}}$ at ~1301 cm⁻¹ are observed. These observations imply a considerable number of gauche conformers within the alkyl chains. The relative intensities and peak frequencies of all bands in these two spectra are essentially identical to those observed in the spectra of the neat alkylsilanes (Figure 1a,b), indicating that the bonded phases have a degree of disorder comparable to that of the liquid state.

Additional information about the conformation of the alkyl chains can be found in the $\nu(\text{C}-\text{H})$ region of the Raman spectra. Figure 2 presents the Raman spectra between 2700 and 3100 cm⁻¹ for (a) neat DOS, (b) neat OTS, (c) crystalline DOS, (d) crystalline OTS, (e) C18MF, and (f) C18TF. For neat DOS and OTS, four main Raman bands are observed at ~2852, 2886, 2931, and 2965 cm⁻¹, assigned to the $\nu_s(\text{CH}_2)$, $\nu_a(\text{CH}_2)$, $\nu_s(\text{CH}_2)_{\text{FR}}$, and $\nu_a(\text{CH}_3)$

modes, respectively. In the crystalline state, the vibrational modes of the methylene groups shift to slightly lower frequency, and their relative peak intensities change significantly. Previous studies^{37,38} have shown that the peak intensity ratio of the methylene asymmetric (at ~2880 cm⁻¹) to symmetric (at ~2852 cm⁻¹) $\nu(\text{CH}_2)$ bands and their peak frequencies are sensitive to the degree of alkyl chain order. In the liquid state, for example, the intensity ratio of these methylene bands is ~0.6–0.9, whereas in the crystalline solid, this ratio increases to ~1.6–2.0. Since the $\nu(\text{C}-\text{H})$ vibrational envelope in these spectra contains a number of modes, the $[I_{\nu_a(\text{CH}_2)}/I_{\nu_s(\text{CH}_2)}]$ value is used as a qualitative indicator of conformation within the alkyl chain. For DOS and OTS, this ratio has a value of ~0.9 in the liquid state (Figure 2a,b) and increases to ~2.0 in the crystalline state (Figure 2c,d). These values are consistent with the expected degree of order for the alkyl chains in each state.

For C18MF (Figure 2e) and C18TF (Figure 2f), the $\nu_s(\text{CH}_2)$ is observed at 2855 cm⁻¹, and the $\nu_a(\text{CH}_2)$ is observed as shoulder at 2885 cm⁻¹. Analysis of the Raman spectra shows that the intensity ratio of these two bands is slightly less than 1. This value is consistent with a highly disordered alkyl chain state similar to that of the liquid, a result which is also in agreement with the conclusions from the $\nu(\text{C}-\text{C})$ region.

In total, the Raman spectroscopy implies that the stationary phase alkyl chains exist in a disordered, liquid-like state on the silica surfaces of these packing materials. These findings are in *general* agreement with earlier IR results of Sander et al.¹⁹ They reported that the degree of conformational disorder of the alkyl chain is comparable to that observed in the corresponding liquid state at room temperature and above. However, careful examination of their data also suggests a reduction of gauche conformers for alkylsilanes bonded to stationary phase surfaces. This conclusion is contrary to that drawn here on the basis of the Raman spectral results. Due to a lack of information about the differences in properties (e.g., surface coverage, surface area, particle size, and pore size) between our packing materials and theirs and the effects that such differences may have on alkyl chain conformation, we do not yet fully understand the origins of these conformational

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differences. However, it should be noted that the chain surface coverages of C18MF and C18TF are less than $2 \mu\text{mol}/\text{m}^2$, which is relatively low when compared to the maximum possible coverage ($\sim 4.5 \mu\text{mol}/\text{m}^2$). Thus, these surface coverages represent submonolayer quantities of alkyl chains on the silica. Consequently, a more disordered alkyl chain conformation is expected on the basis of predictions from molecular dynamics simulations.^{3,39} Detailed studies of the effect of surface coverage on alkyl chain conformation are beyond the scope of this work; these studies are ongoing in this laboratory and will be reported at a later date.

CONCLUSION

The results presented here demonstrate the first use of Raman spectroscopy for the characterization of alkylsilane stationary phases on commercially available chromatographic packing materials. The high spectral quality indicates that Raman spectroscopy is a valuable tool for the characterization of these packing

materials. Our findings suggest that the alkyl chains of the chromatographic stationary phase exist in a highly disordered state which is essentially identical to that of the neat liquid. These findings are consistent with previously reported results obtained with different characterization techniques. Work is in progress to further investigate fundamental issues (such as the effects of solvents, temperature, surface coverage, and the presence of solute) in chromatographic retention on a variety of real packing materials.

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