SUPPORTING INFORMATION

Dry and Wet Wrinkling of a Silk Fibroin Biopolymer by a Shape-Memory Material with Insight into Mechanical Effects on Secondary Structures in Silk Network

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1. SUPPORTING METHODS

Supporting Method I - *SMP substrates for shape memory biaxial contraction.* To prepare SMP samples in which primarily compressive stresses are present in the substrate during shape recovery, we utilized a 4D-printing technique recently developed in our lab, known as programming via printing (PvP).[58] PvP involves using fused deposition modeling (FDM) to 3D-print an SMP substrate while simultaneously programming strains in the substrate at the fiber level during the printing process. This eliminates the need for a separate programming step following fabrication, and the SMP substrate can immediately undergo shape transformation after printing once exposed to an external stimulus (e.g., heat). The TPU pellets were dried for a minimum of 24 h in a vacuum oven at 50 °C prior to undergoing extrusion into filaments (Composer 450, 3Devo., The Netherlands). To print the SMP filament into desired substrates, STL files were converted to G-code using a slicing software (Ultimaker Cura., The Netherlands). Next, the SMP filament was loaded into a 3D printer (Ender 3 Pro., Creality) with a 0.4 mm nozzle diameter for depositing the semi-molten material onto the build plate in a layer-by-layer process. To ensure proper sample adhesion to the build plate, the plate was covered with Kapton tape and maintained at 25 °C. Rectangular samples (dimensions: 20 mm by 10 mm, 1.5 mm thick) were printed with fiber orientations of 0° and 90° alternating layer-by-layer for biaxial contraction. The samples were printed with a nozzle temperature of 200 °C, print speed of 30 mm/s, and print flow rate of 125%. Once printed, samples were stored in a desiccator until use.

Supporting Method II - *SMP substrates for shape memory bending actuated tension*. To prepare SMP samples in which primarily tensile stresses are present in the substrate during shape recovery, 3D printing was conducted but samples had to undergo conventional programming. The TPU filament was used to 3D print a "U-shaped" SMP substrate. To print the filament into the desired SMP substrate, the "U-shape" design was created in computer aided design software and STL files were converted to g-code using Ultimaker Cura. Next, the TPU filament was loaded into the Ender 3 Pro with 0.4 mm nozzle diameter. To optimize bed adhesion, the build plate was covered with Kapton tape and maintained at 50 °C. The U-shaped SMP substrates were printed with a nozzle temperature of 220 °C, print speed of 30 mm/s, and print

flow rate of 125%. To set the secondary shape of the SMP, the ends of the substrate were clamped, then the U-shaped SMP was preheated in an isothermal oven at 50 °C for 5 min, then tension was applied to the clamps to straighten the substrate, and then cooled in a freezer at -20 °C to fix the straightened, temporary shape. Once fixed, samples were removed from the clamps and stored in a desiccator until use.

Supporting Method III - *SF Thin Film Application for SMP substrates undergoing shape memory biaxial contraction or shape memory bending actuated tension*. Since one of the major limitations of spin coating is that it cannot create films on architecturally complex and/or curved surfaces, dip coating was used for application of a SF film to the surface of these SMP substrates. The stock concentration of SF was adjusted to 3% w/v by adding ultrapure water. To coat the SMP substrates with SF, samples were dip coated in a 20 ml scintillation vial for 2 min at room temperature ensuring that we prevent pre-recovery of the SMP. Then, by adapting a previously published fabrication procedure,[59] the silk coated SMP was immediately post-treated in a solution of 70% methanol for 5 sec. The treated samples were stored in a desiccator to dry overnight.

2. ADDITIONAL SUPPORTING FIGURES

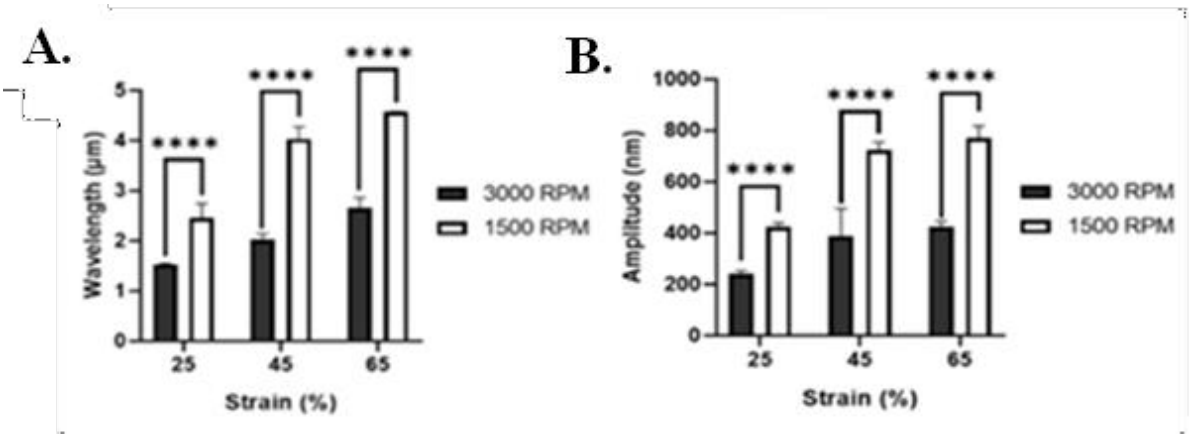


FIGURE S1. Detailed characterization of the effects of SF spin-coating speeds on wrinkle (A) wavelength and (B) amplitude at 70 °C. (n=3, Student's t-test for two group comparisons, *p< 0.05, **p< 0.01, ***p<0.001, ****p<0.0001).

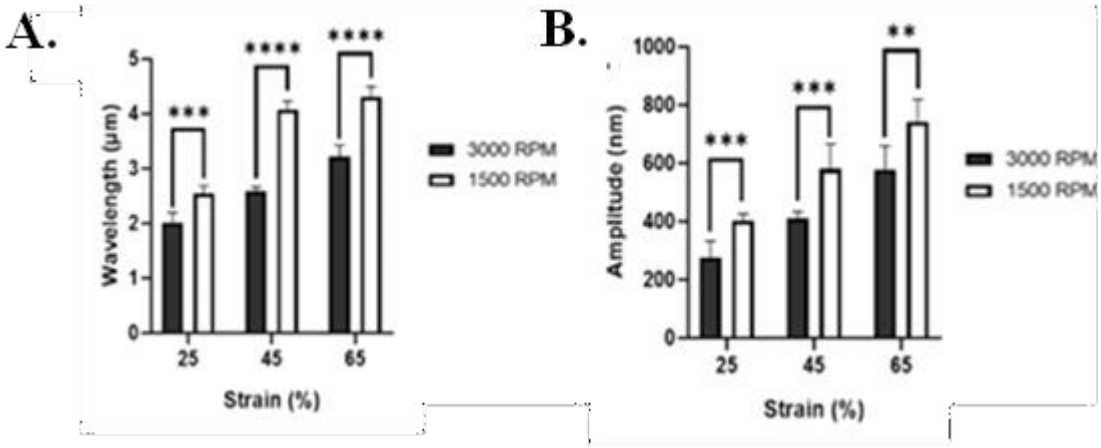
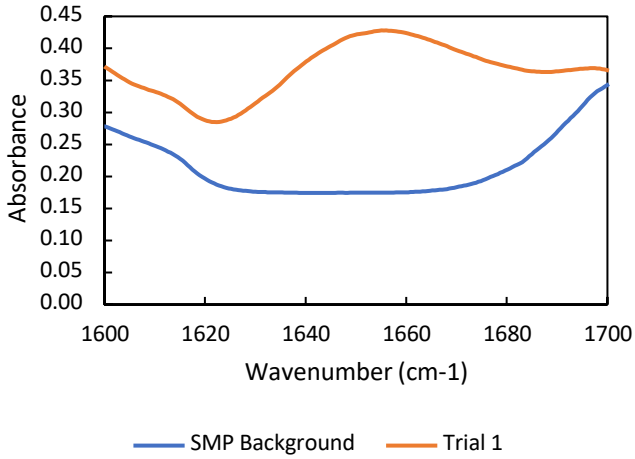


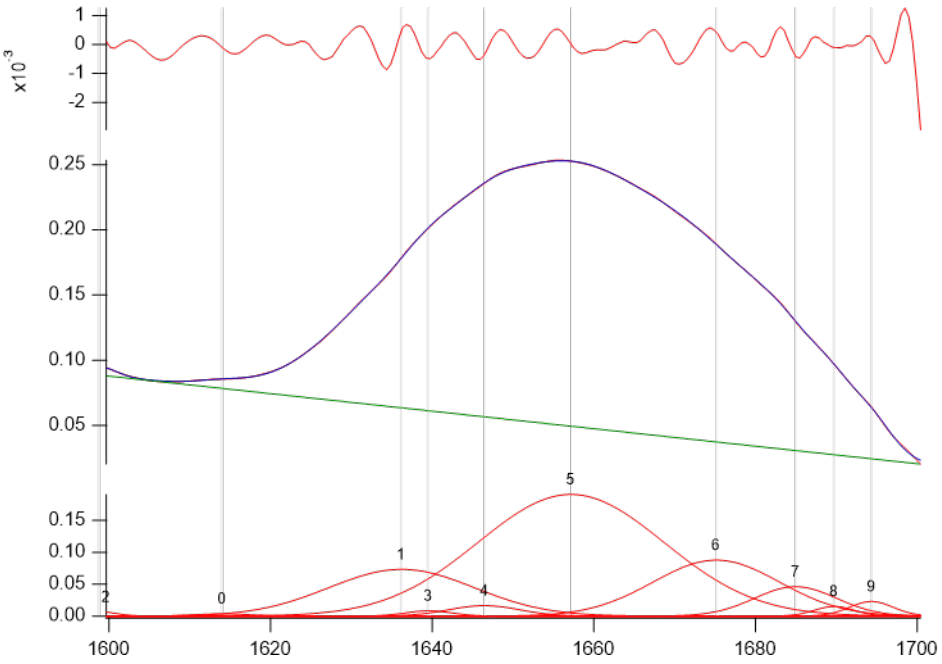
FIGURE S2. Detailed characterization of the effects of SF spin-coating speeds on wrinkle (A) wavelength and (B) amplitude at 40 °C. (n=3, Student's t-test for two group comparisons, *p< 0.05, **p< 0.01, ***p<0.001, ****p<0.0001).

No Methanol Trial 1

FTIR Background Substraction

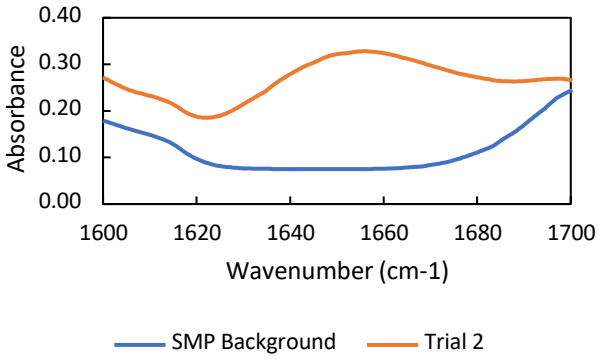


FTIR Deconvolution

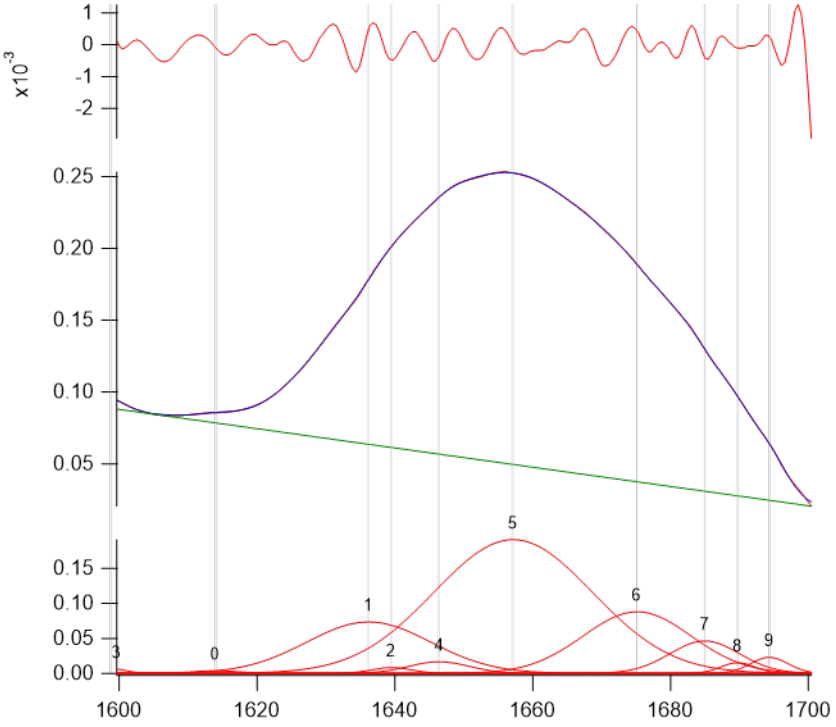


No Methanol Trial 2

FTIR Background Substraction



FTIR Deconvolution



No Methanol Trial 3

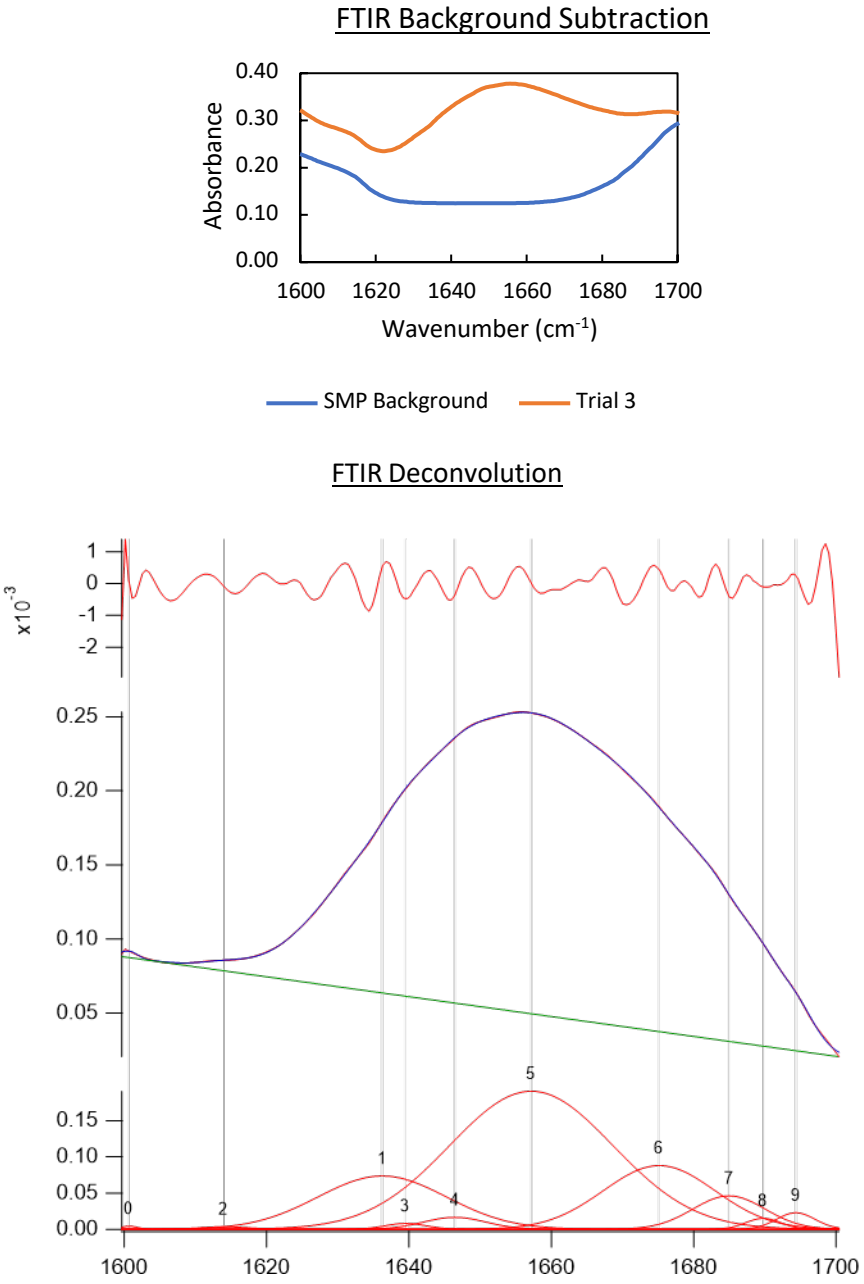
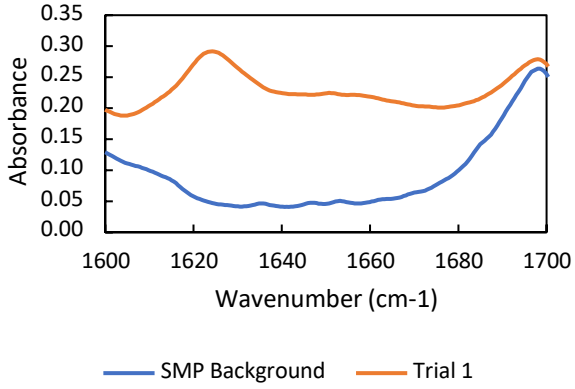


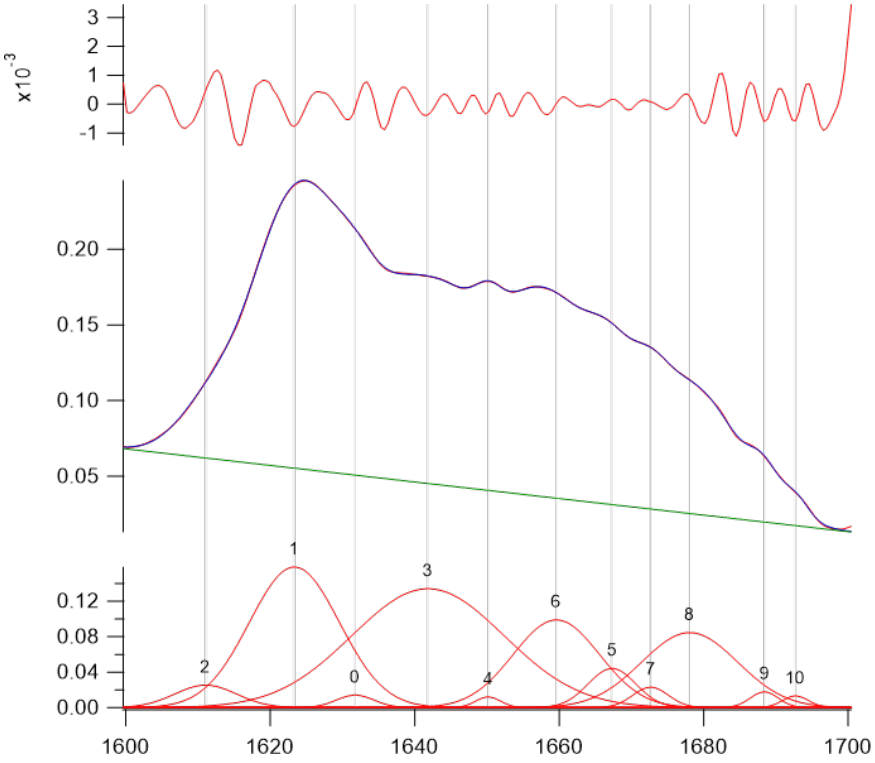
FIGURE S3. Deconvolution of FTIR spectra within the Amide I region (1600-1700 cm^{-1}). (Top panels) Multiple linear regression is used to subtract the contribution of the SMP substrate (blue) from the total signal (SF + SMP) with no methanol treatment (orange) through decomposition. Three independent replicates were measured for each condition (n=3). (Bottom panels) Deconvolution results of FTIR spectra for SF-SMP with no methanol treatment. In the middle panel, raw data (red) and reproduced spectra (blue) are depicted, with the blue line representing the summation of the deconvoluted peaks. Residual fit errors are depicted in the top panels. The bottom panel shows the results from the deconvolution process; secondary structures are assigned based on peak locations within this panel. The normalized peak areas were used to determine the secondary structures found in the film.

30 Minute Methanol Trial

FTIR Background Subtraction

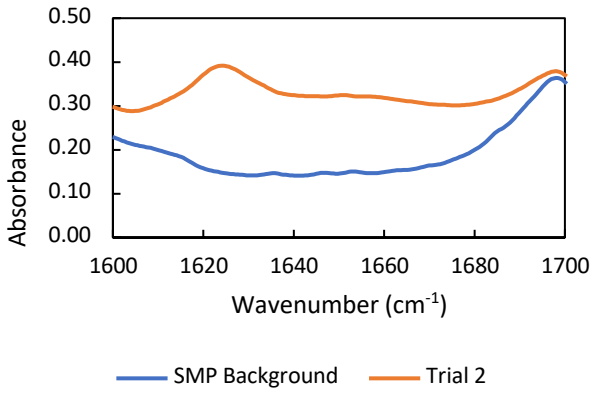


FTIR Deconvolution

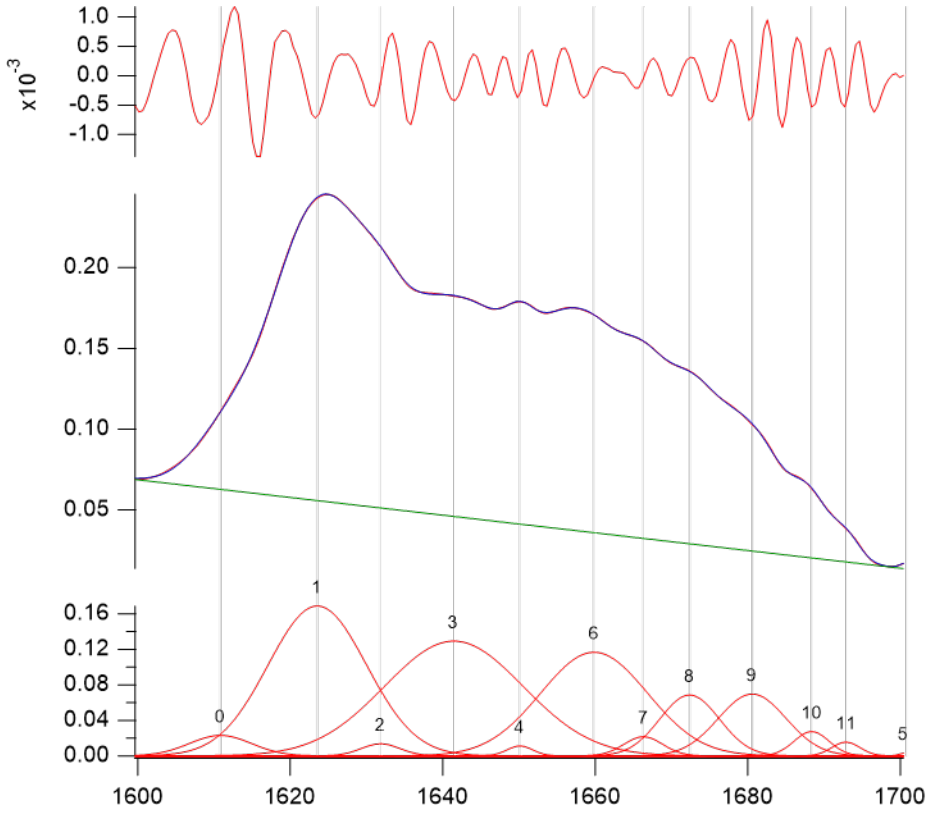


30 Minute Methanol Trial

FTIR Background Subtraction



FTIR Deconvolution



30 Minute Methanol Trial

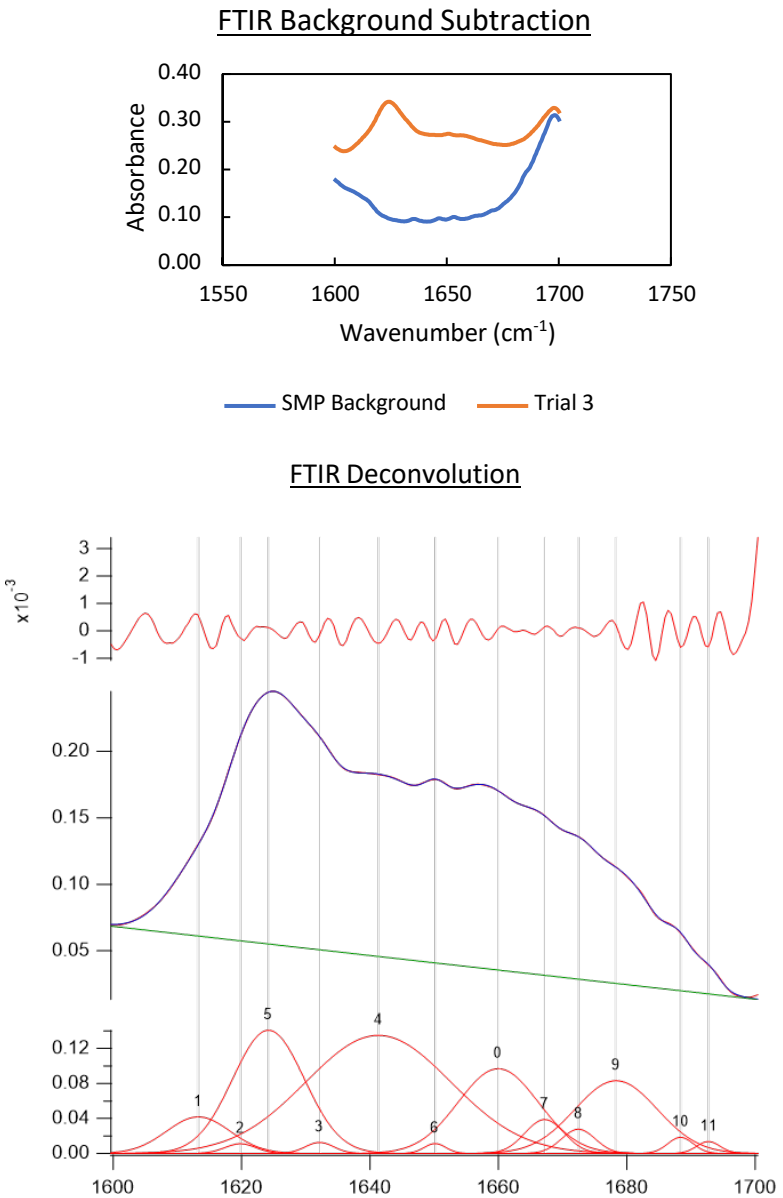
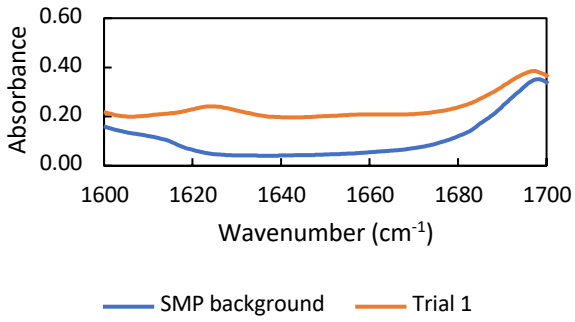


FIGURE S4. Deconvolution of FTIR spectra within the Amide I region (1600-1700 cm^{-1}). (Top panels) Multiple linear regression is used to subtract the contribution of the SMP substrate (blue) from the total signal (SF + SMP) with 30 min methanol treatment (orange) through decomposition. Three independent replicates were measured for each condition (n=3). (Bottom panels) Deconvolution results of FTIR spectra for SF-SMP with 30 min methanol treatment. In the middle panel, raw data (red) and reproduced spectra (blue) are depicted, with the blue line representing the summation of the deconvoluted peaks. Residual fit errors are depicted in the top panels. The bottom panel shows the results from the deconvolution process; secondary structures are assigned based on peak locations within this panel. The normalized peak areas were used to determine the secondary structures found in the film.

40 Minute Methanol Trial

FTIR Background Subtraction



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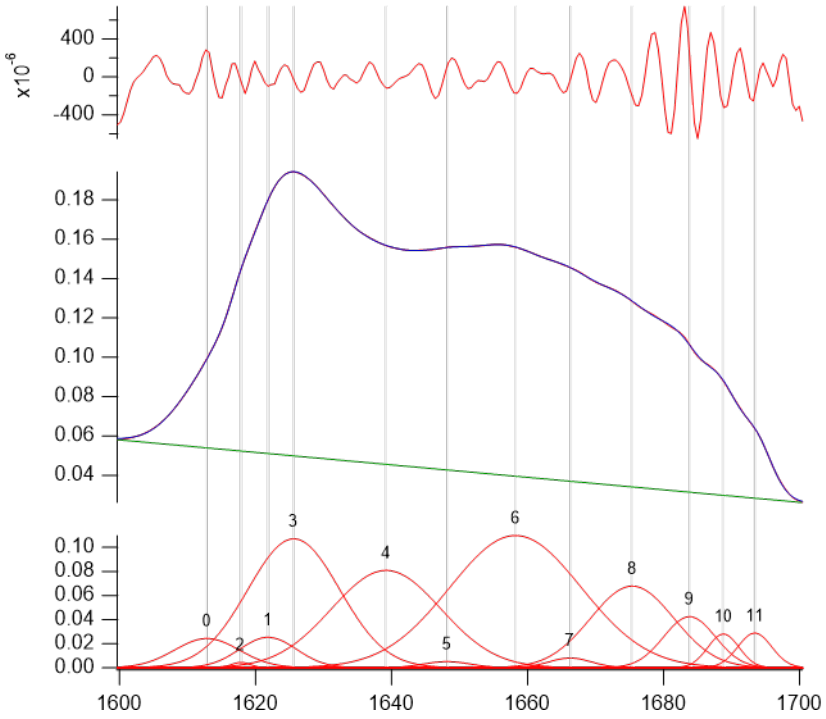
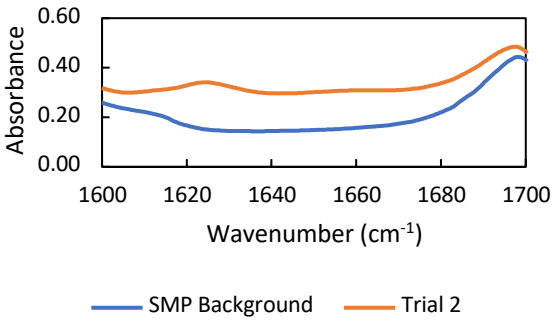
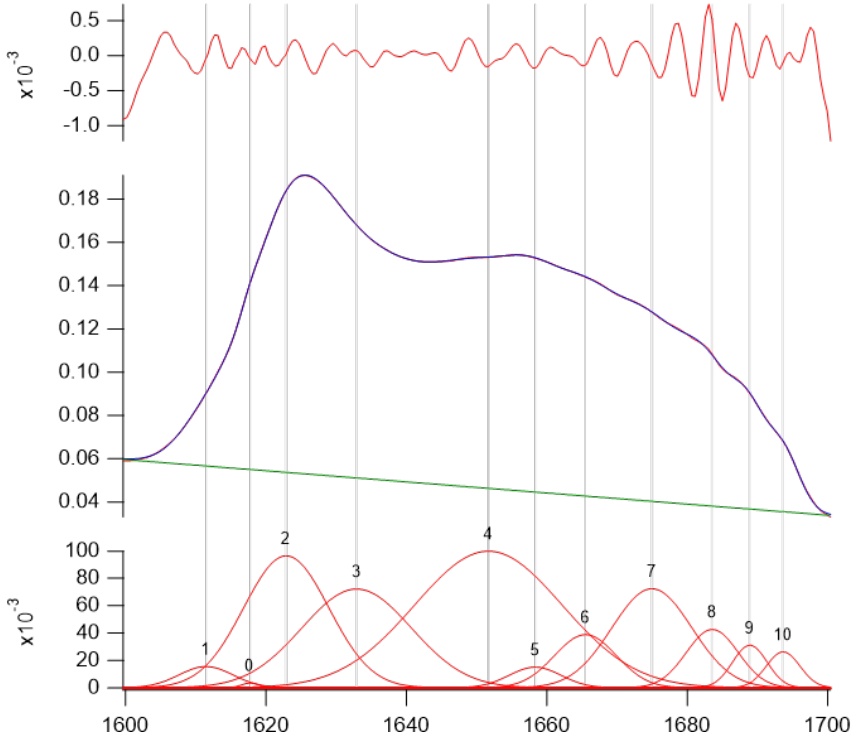


40 Minute Methanol Trial

FTIR Background Subtraction



FTIR Deconvolution



40 Minute Methanol Trial

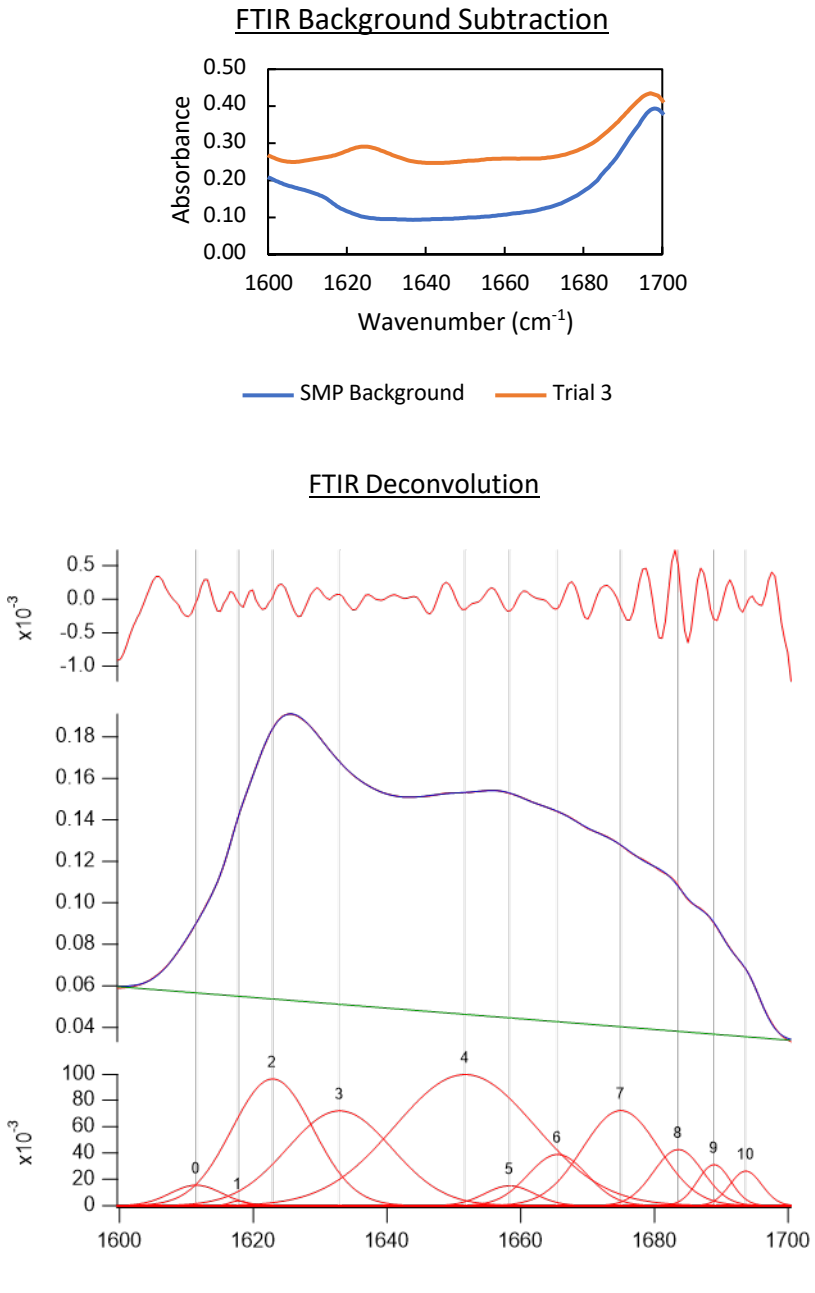
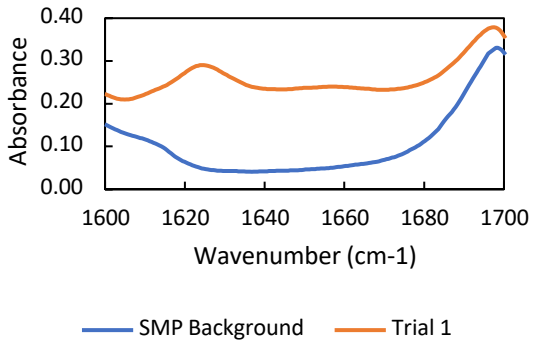


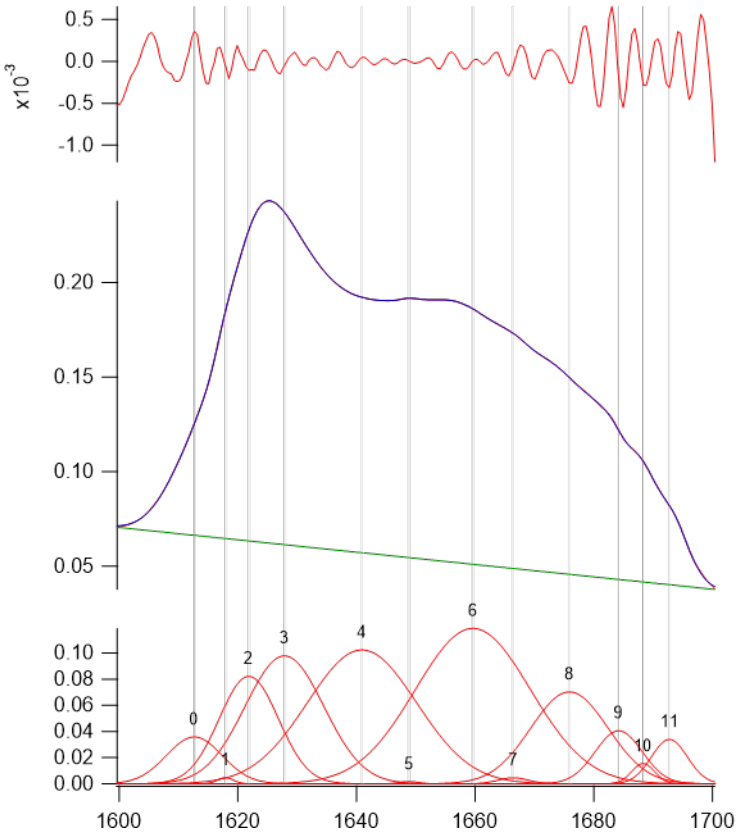
FIGURE S5. Deconvolution of FTIR spectra within the Amide I region (1600-1700 cm^{-1}). (Top panels) Multiple linear regression is used to subtract the contribution of the SMP substrate (blue) from the total signal (SF + SMP) with 40 min methanol treatment (orange) through decomposition. Three independent replicates were measured for each condition (n=3). (Bottom panels) Deconvolution results of FTIR spectra for SF-SMP with 40 min methanol treatment. In the middle panel, raw data (red) and reproduced spectra (blue) are depicted, with the blue line representing the summation of the deconvoluted peaks. Residual fit errors are depicted in the top panels. The bottom panel shows the results from the deconvolution process; secondary structures are assigned based on peak locations within this panel. The normalized peak areas were used to determine the secondary structures found in the film.

50 Minute Methanol Trial 1

FTIR Background Subtraction

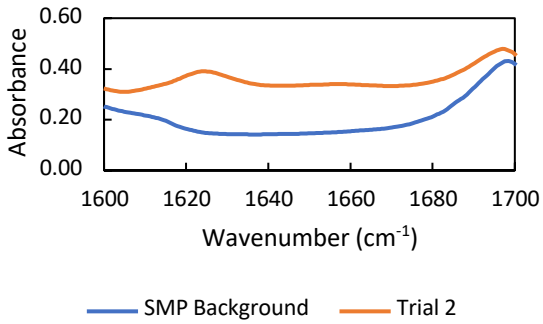


FTIR Deconvolution

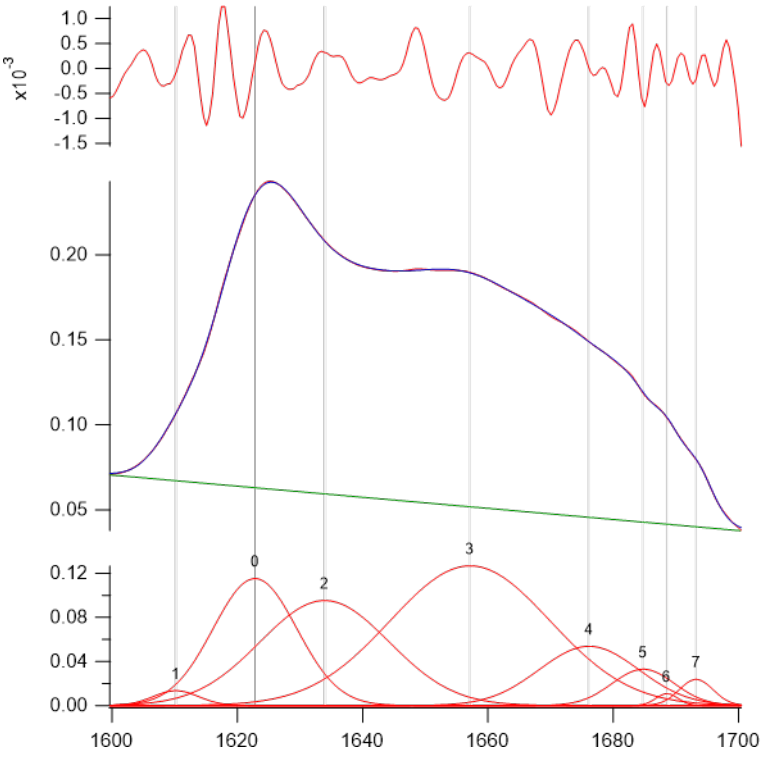


50 Minute Methanol Trial

FTIR Background Subtraction



FTIR Deconvolution



50 Minute Methanol Trial

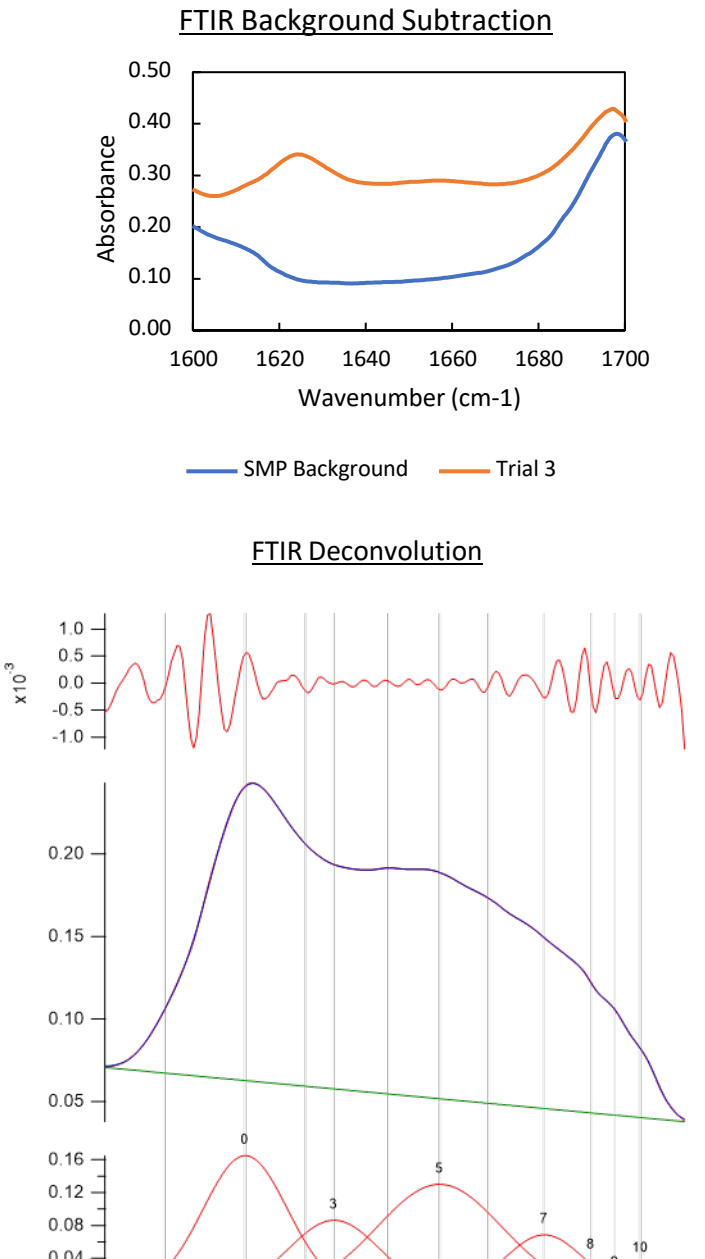
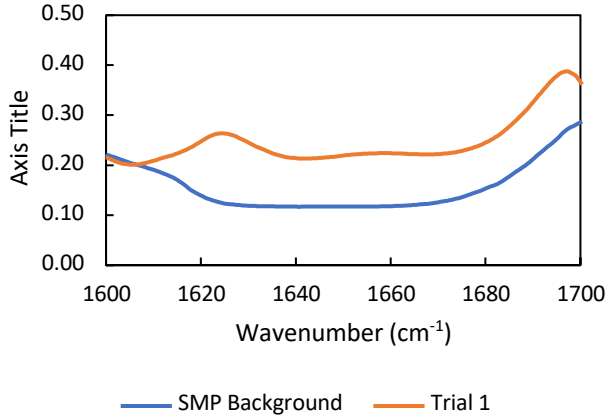


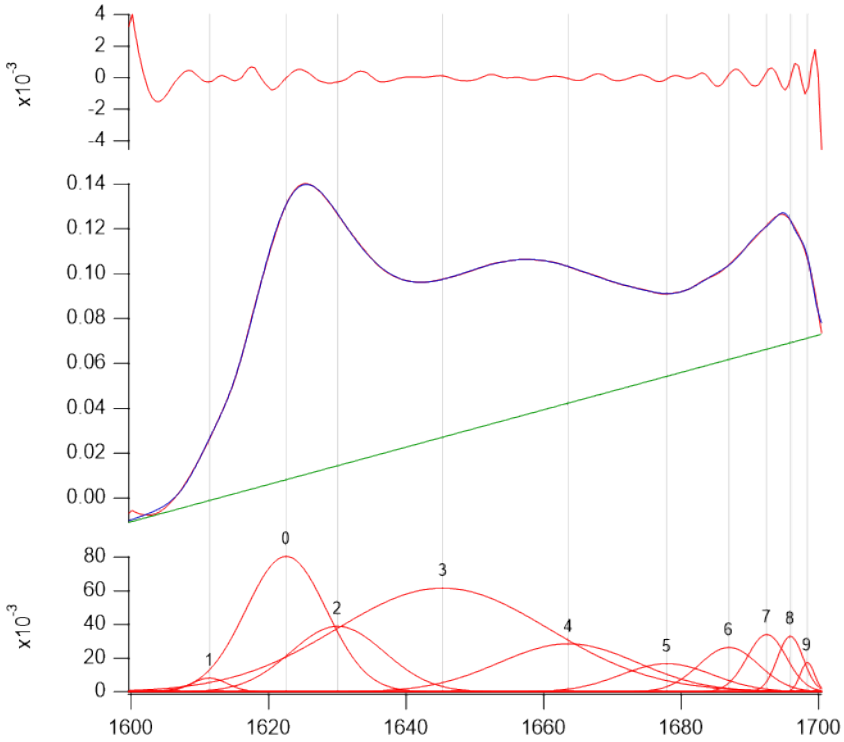
FIGURE S6. Deconvolution of FTIR spectra within the Amide I region (1600-1700 cm^{-1}). (Top panels) Multiple linear regression is used to subtract the contribution of the SMP substrate (blue) from the total signal (SF + SMP) with 50 min methanol treatment (orange) through decomposition. Three independent replicates were measured for each condition (n=3). (Bottom panels) Deconvolution results of FTIR spectra for SF-SMP with 50 min methanol treatment. In the middle panel, raw data (red) and reproduced spectra (blue) are depicted, with the blue line representing the summation of the deconvoluted peaks. Residual fit errors are depicted in the top panels. The bottom panel shows the results from the deconvolution process; secondary structures are assigned based on peak locations within this panel. The normalized peak areas were used to determine the secondary structures found in the film.

60 Minute Methanol Trial 1

FTIR Background Subtraction

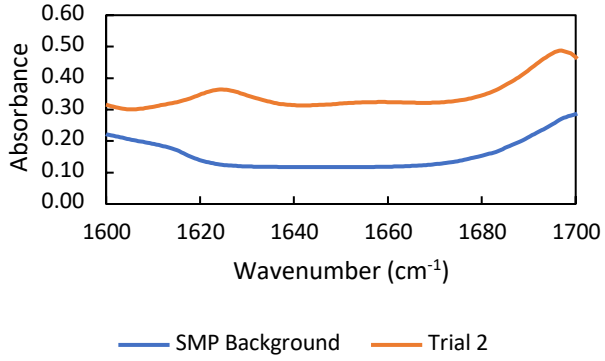


FTIR Deconvolution

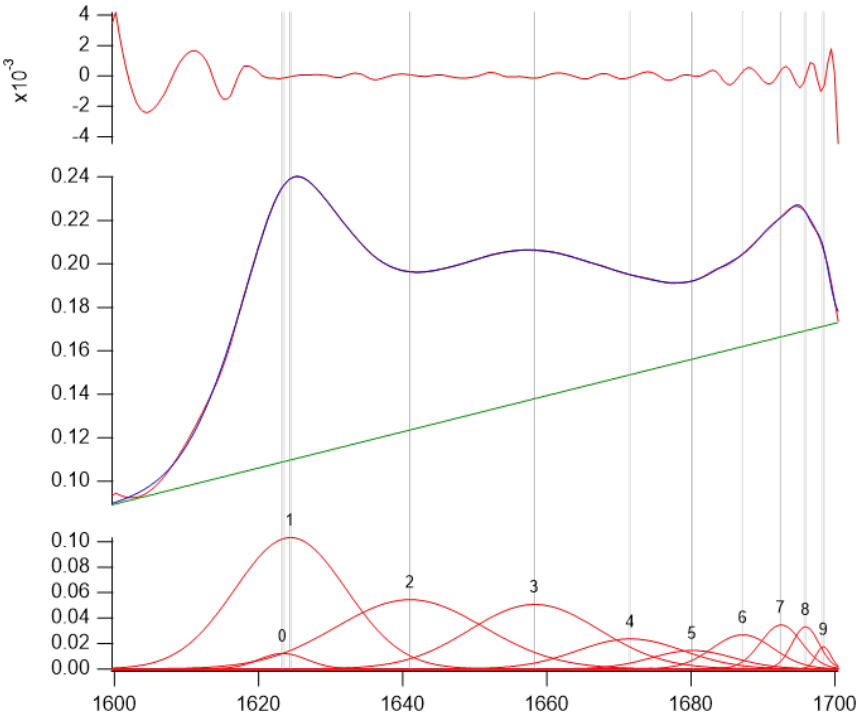


60 Minute Methanol Trial

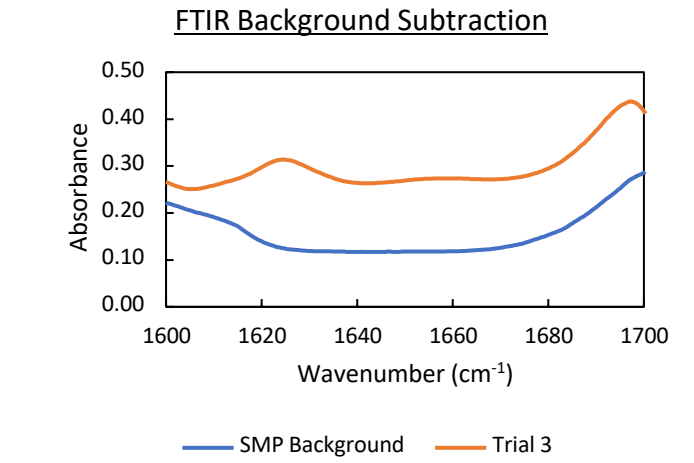
FTIR Background Subtraction



FTIR Deconvolution



60 Minute Methanol Trial



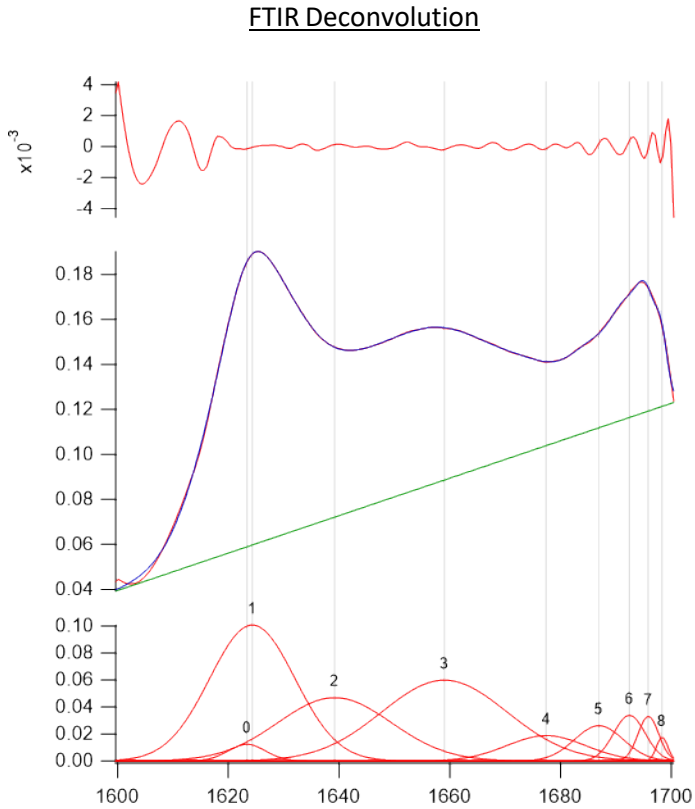


FIGURE S7. Deconvolution of FTIR spectra within the Amide I region (1600-1700 cm^{-1}). (Top panels) Multiple linear regression is used to subtract the contribution of the SMP substrate (blue) from the total signal (SF + SMP) with 60 min methanol treatment (orange) through decomposition. Three independent replicates were measured for each condition (n=3). (Bottom panels) Deconvolution results of FTIR spectra for SF-SMP with 60 min methanol treatment. In the middle panel, raw data (red) and reproduced spectra (blue) are depicted, with the blue line representing the summation of the deconvoluted peaks. Residual fit errors are depicted in the top panels. The bottom panel shows the results from the deconvolution process; secondary structures are assigned based on peak locations within this panel. The normalized peak areas were used to determine the secondary structures found in the film.

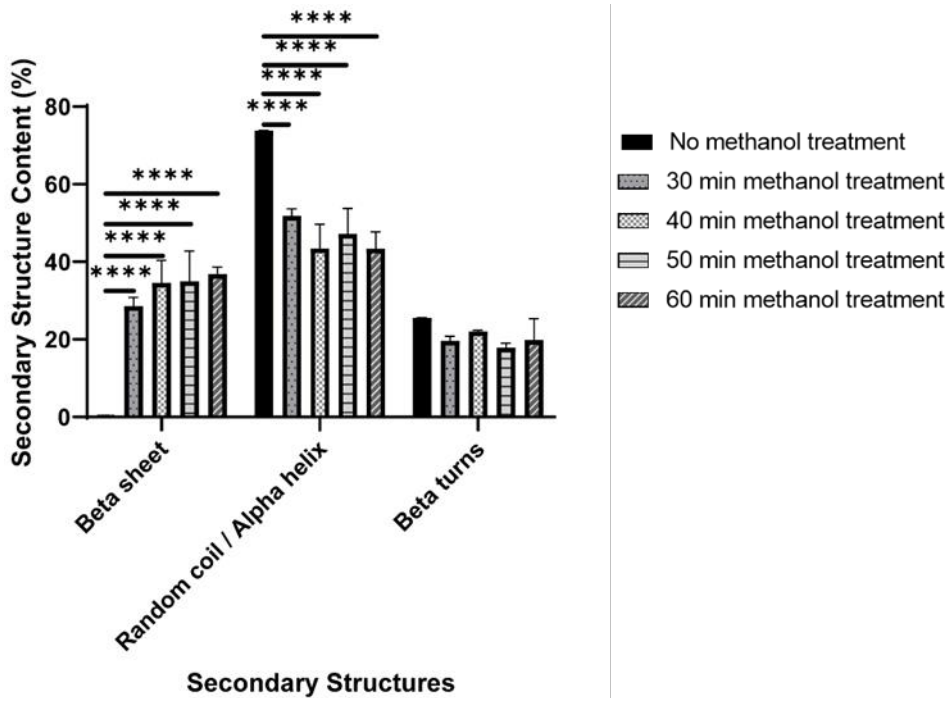
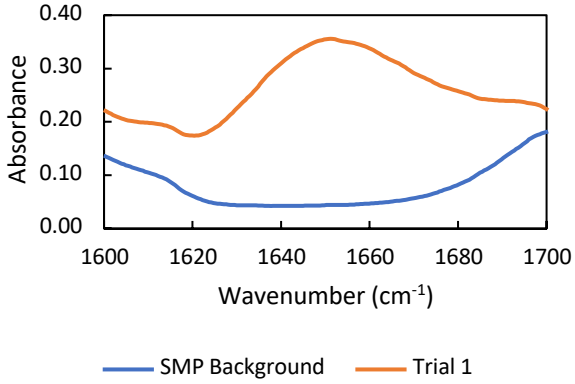


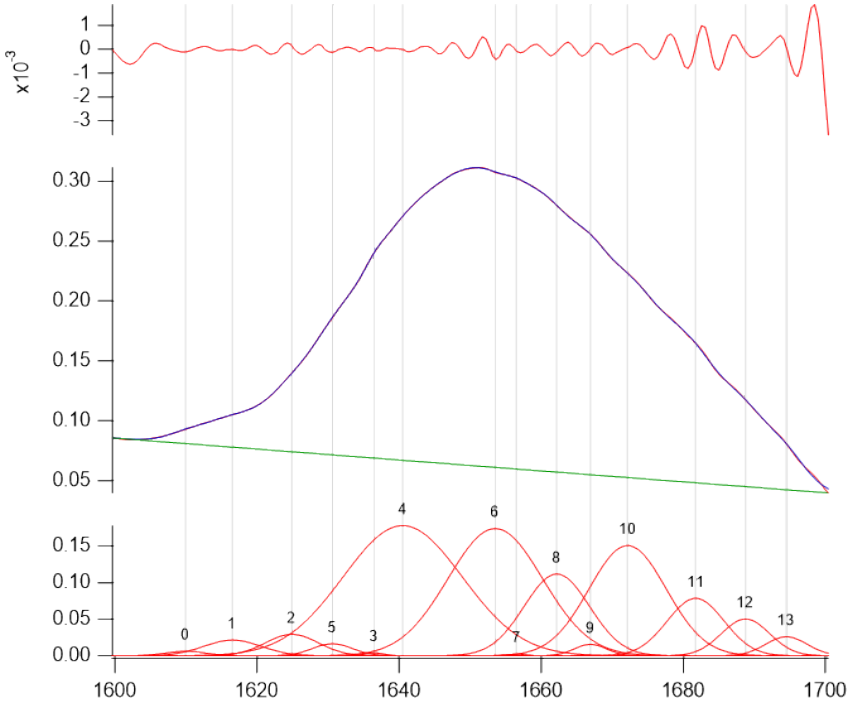
FIGURE S8. Methanol treatment provides secondary structural changes in the silk network and induces β-sheet formation. Detailed comparison of the secondary structure content with no methanol treatment and with varying methanol treatment times of 30 min, 40 min, 50 min, and 60 min.

Unstrained SF-SMP, No MeOH, 40 °C Trial

FTIR Background Subtraction

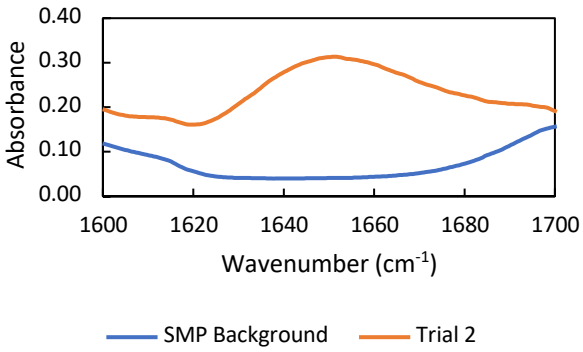


FTIR Deconvolution

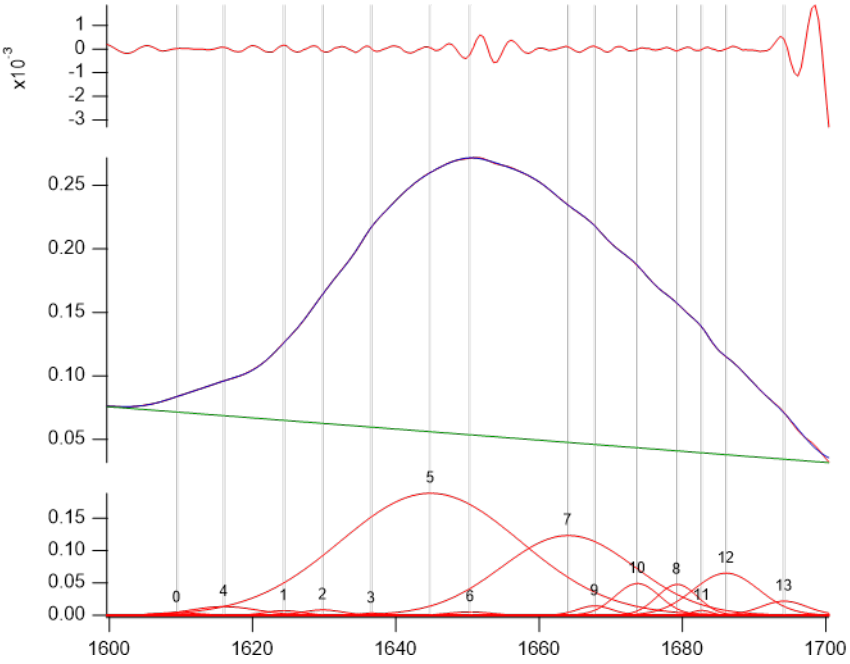


Unstrained SF-SMP, No MeOH, 40 °C Trial

FTIR Background Subtraction



FTIR Deconvolution



Unstrained SF-SMP, No MeOH, 40 °C Trial

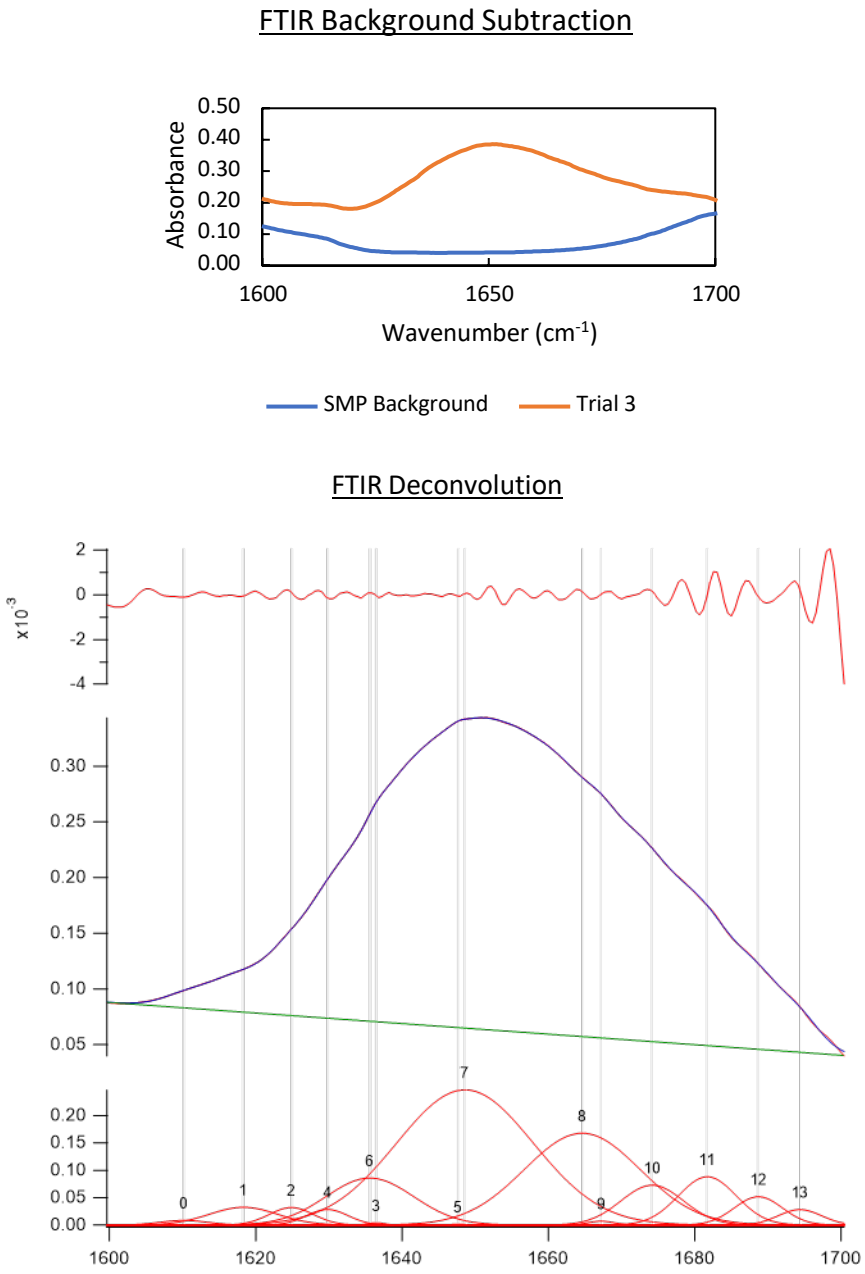
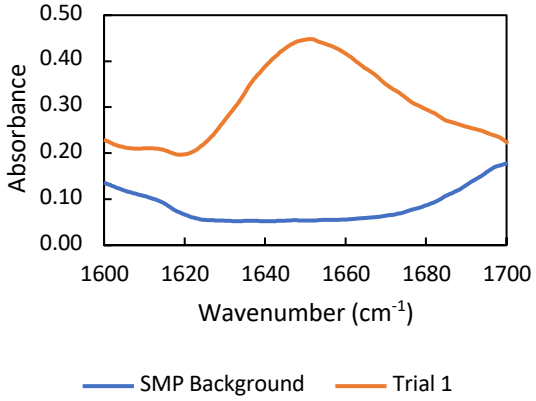


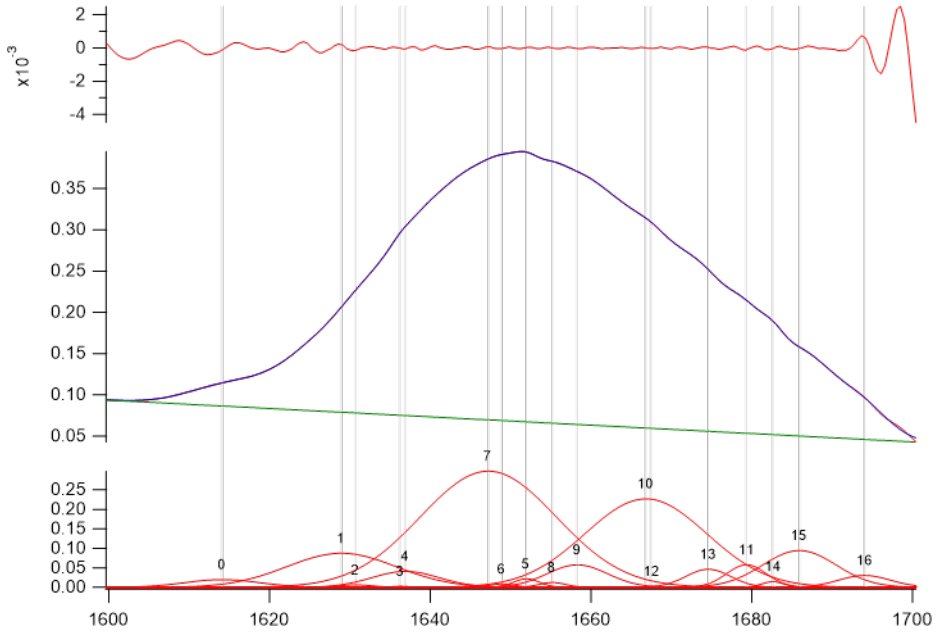
FIGURE S9. Deconvolution of FTIR spectra within the Amide I region (1600-1700 cm^{-1}) following 40 °C heating. (Top panels) Multiple linear regression is used to subtract the contribution of the SMP substrate (blue) from the total signal (SF + unstrained SMP) with no methanol treatment (orange) through decomposition. Three independent replicates were measured for each condition (n=3). (Bottom panels) Deconvolution results of FTIR spectra for SF-SMP with no methanol treatment. In the middle panel, raw data (red) and reproduced spectra (blue) are depicted, with the blue line representing the summation of the deconvoluted peaks. Residual fit errors are depicted in the top panels. The bottom panel shows the results from the deconvolution process; secondary structures are assigned based on peak locations within this panel. The normalized peak areas were used to determine the secondary structures found in the film.

Unstrained SF-SMP, No MeOH, 70 °C Trial

FTIR Background Subtraction

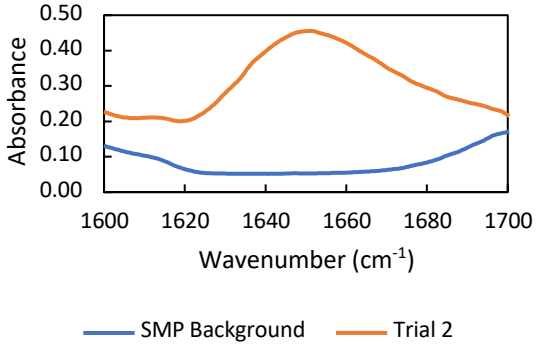


FTIR Deconvolution

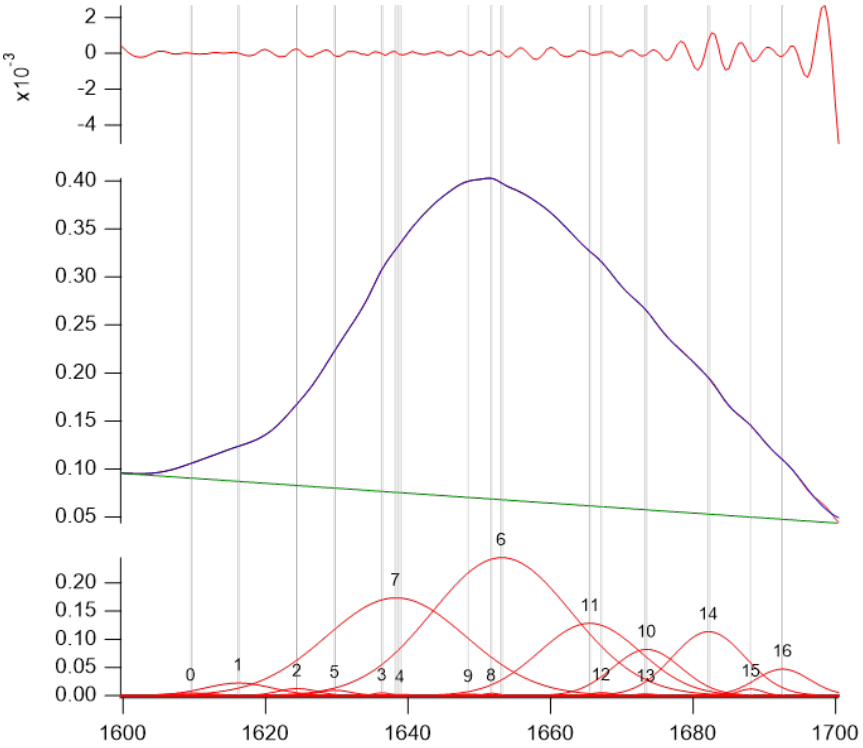


Unstrained SF-SMP, No MeOH, 70 °C Trial

FTIR Background Subtraction



FTIR Deconvolution



Unstrained SF-SMP, No MeOH, 70 °C Trial

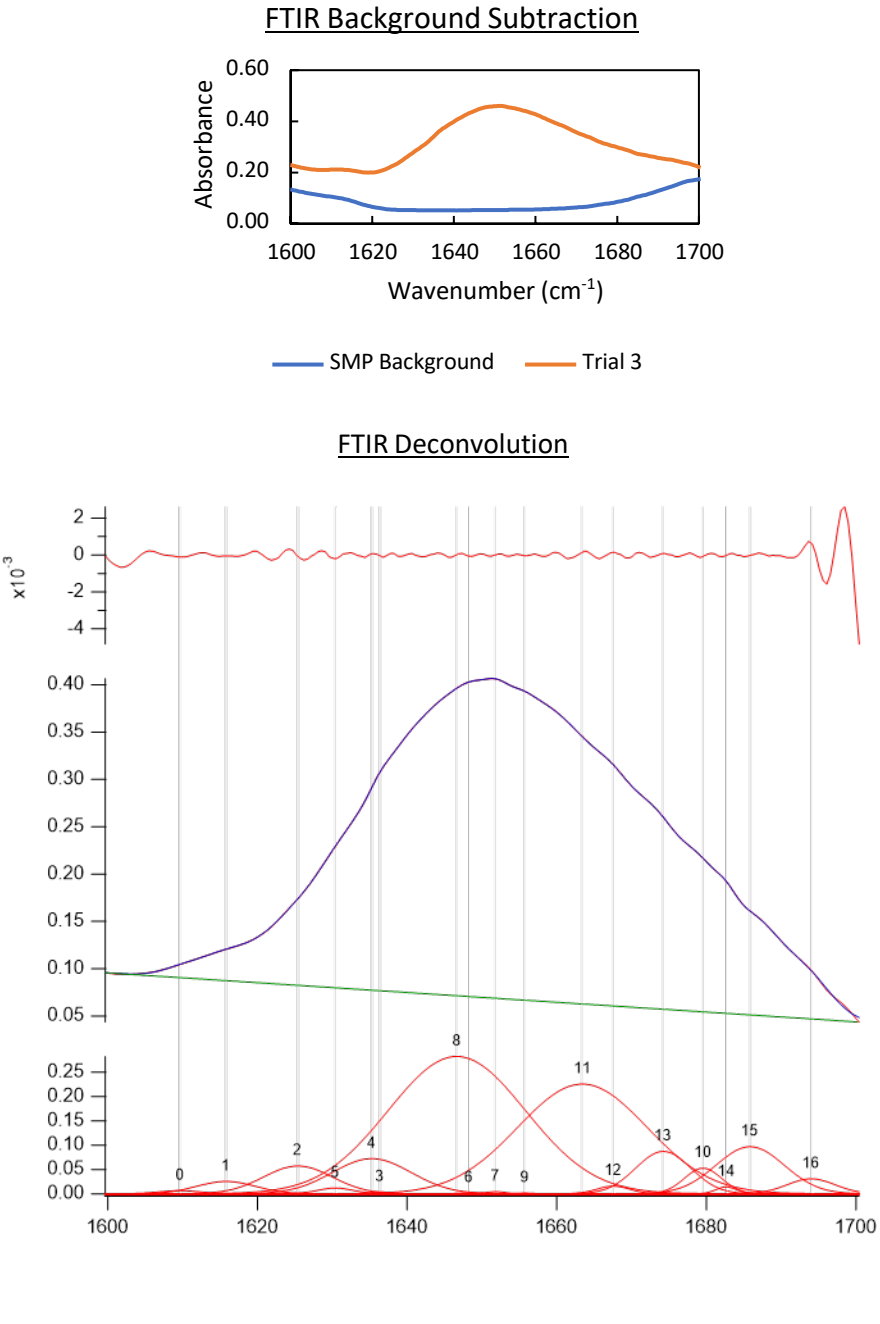
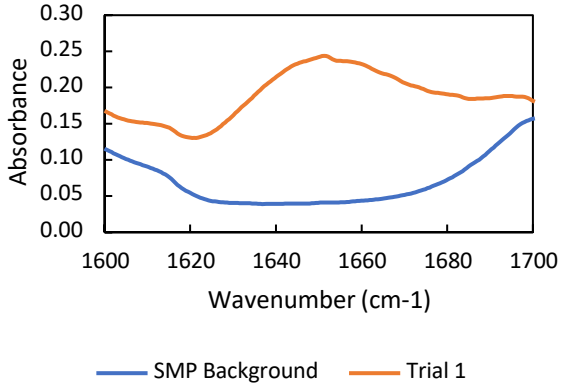


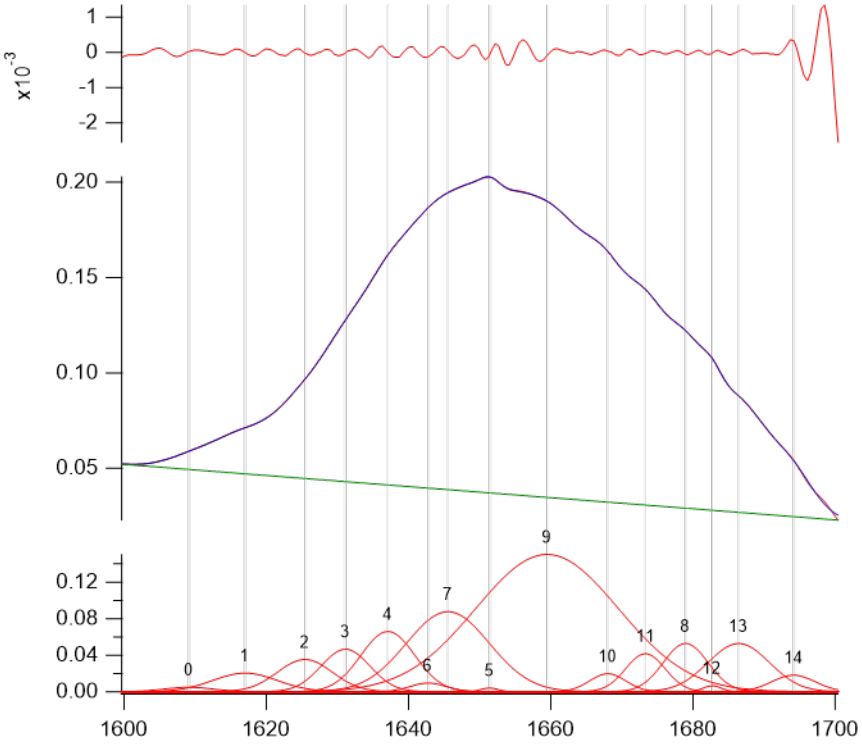
FIGURE S10. Deconvolution of FTIR spectra within the Amide I region (1600-1700 cm^{-1}) following 70 °C heating. (Top panels) Multiple linear regression is used to subtract the contribution of the SMP substrate (blue) from the total signal (SF + unstrained SMP) with no methanol treatment (orange) through decomposition. Three independent replicates were measured for each condition (n=3). (Bottom panels) Deconvolution results of FTIR spectra for SF-SMP with no methanol treatment. In the middle panel, raw data (red) and reproduced spectra (blue) are depicted, with the blue line representing the summation of the deconvoluted peaks. Residual fit errors are depicted in the top panels. The bottom panel shows the results from the deconvolution process; secondary structures are assigned based on peak locations within this panel. The normalized peak areas were used to determine the secondary structures found in the film.

Strained SF-SMP, No MeOH, 40 °C Trial

FTIR Background Subtraction

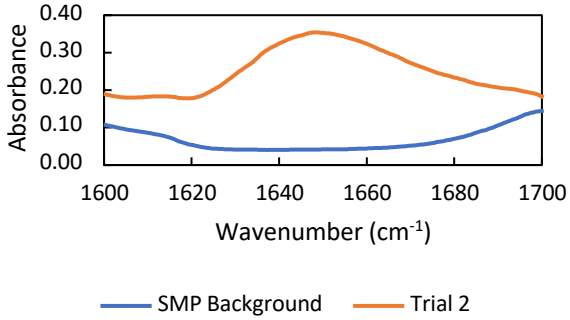


FTIR Deconvolution

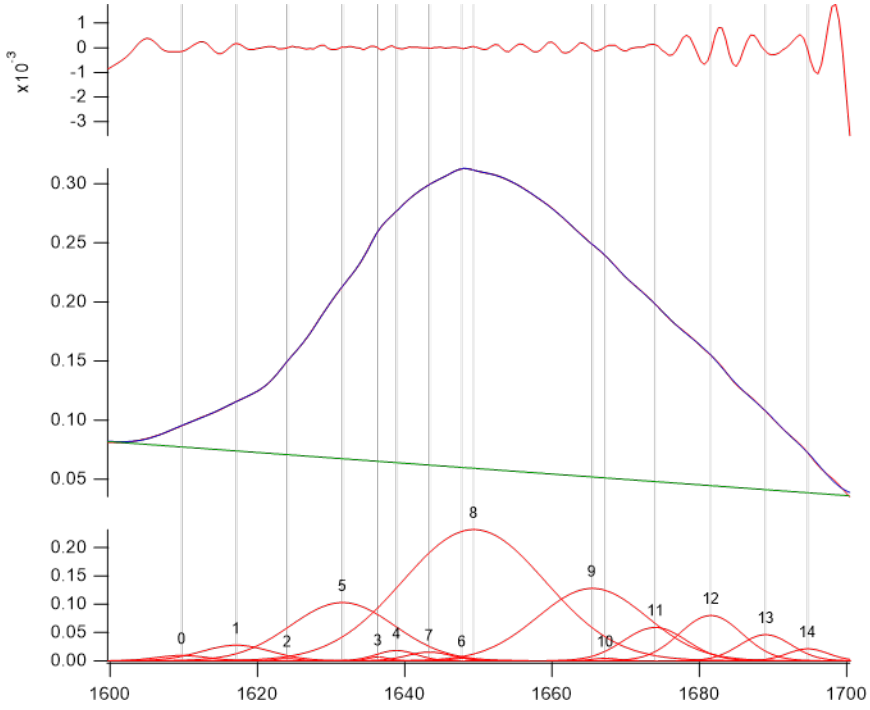


Strained SF-SMP, No MeOH, 40 °C Trial

FTIR Background Subtraction



FTIR Deconvolution



Strained SF-SMP, No MeOH, 40 °C Trial

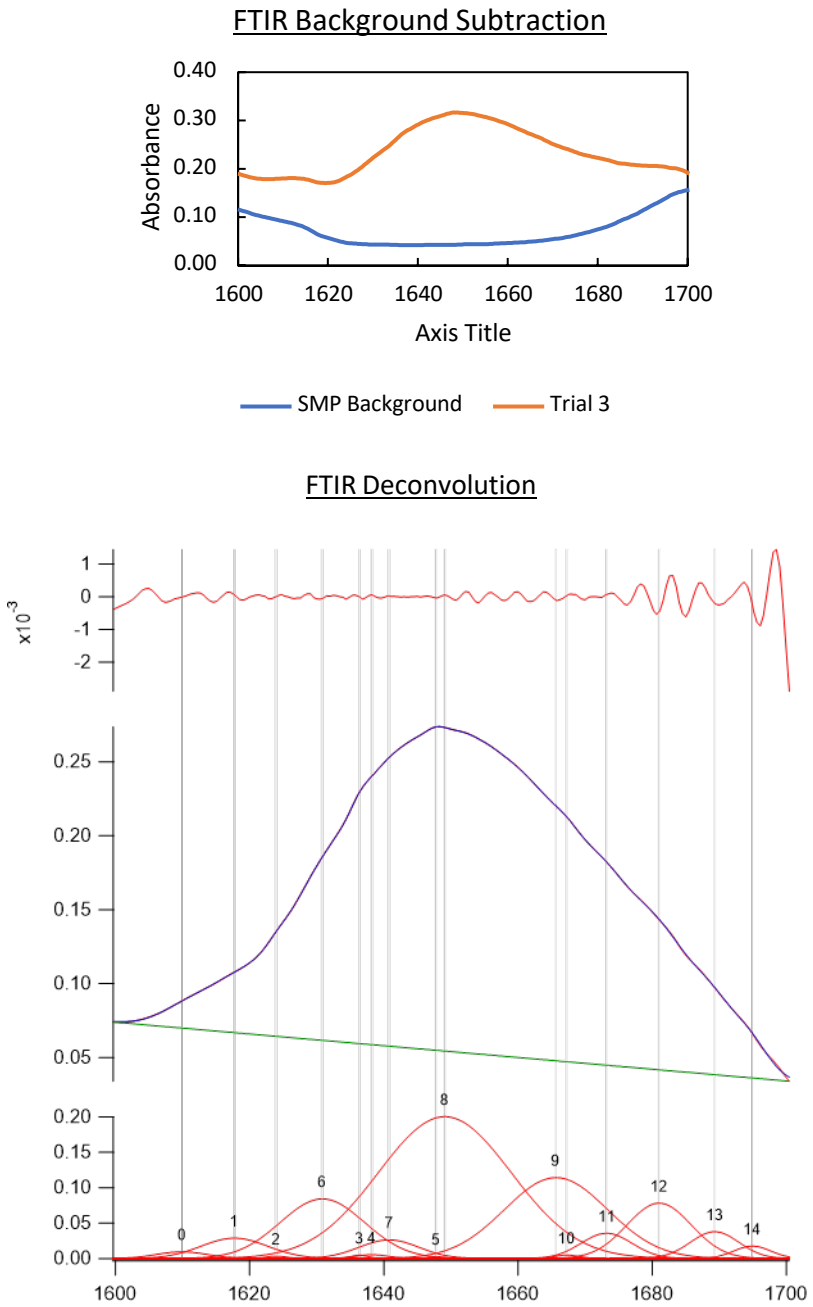
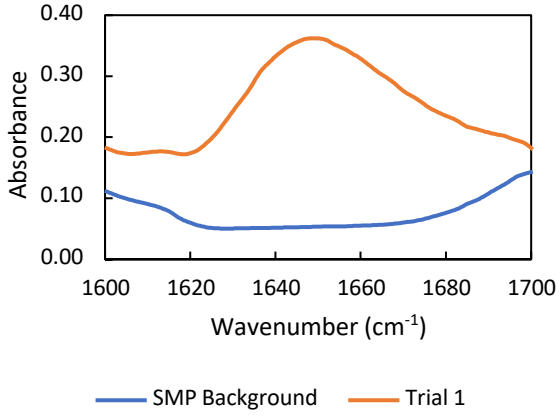


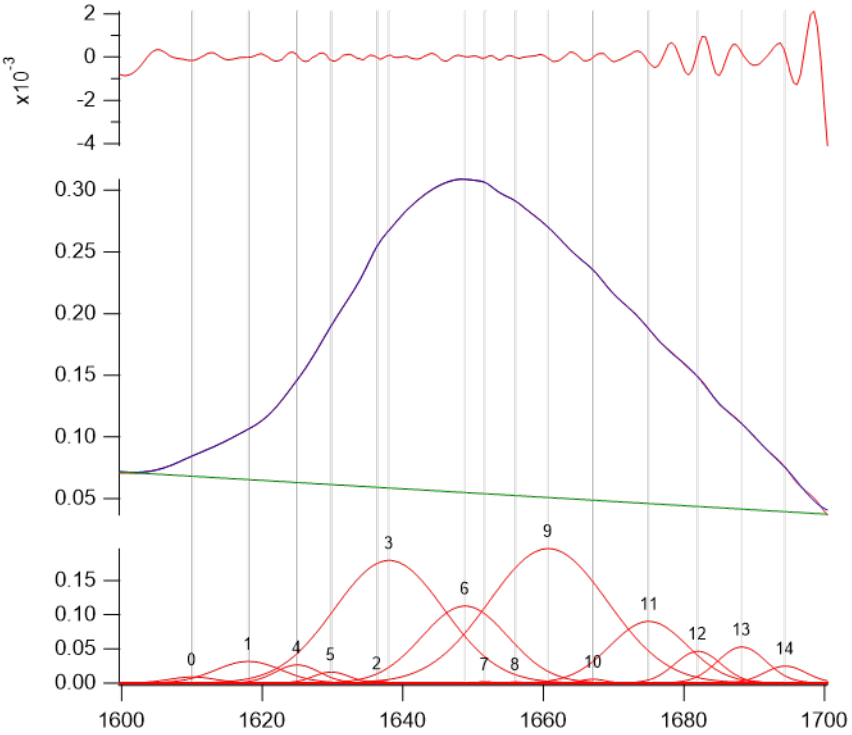
FIGURE S11. Deconvolution of FTIR spectra within the Amide I region (1600-1700 cm^{-1}) following 40 °C heating. (Top panels) Multiple linear regression is used to subtract the contribution of the SMP substrate (blue) from the total signal (SF + strained SMP) with no methanol treatment (orange) through decomposition. Three independent replicates were measured for each condition (n=3). (Bottom panels) Deconvolution results of FTIR spectra for SF-SMP with no methanol treatment. In the middle panel, raw data (red) and reproduced spectra (blue) are depicted, with the blue line representing the summation of the deconvoluted peaks. Residual fit errors are depicted in the top panels. The bottom panel shows the results from the deconvolution process; secondary structures are assigned based on peak locations within this panel. The normalized peak areas were used to determine the secondary structures found in the film.

Strained SF-SMP, No MeOH, 70 °C Trial 1

FTIR Background Subtraction

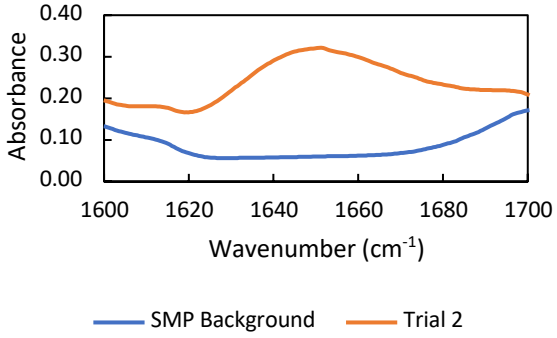


FTIR Deconvolution

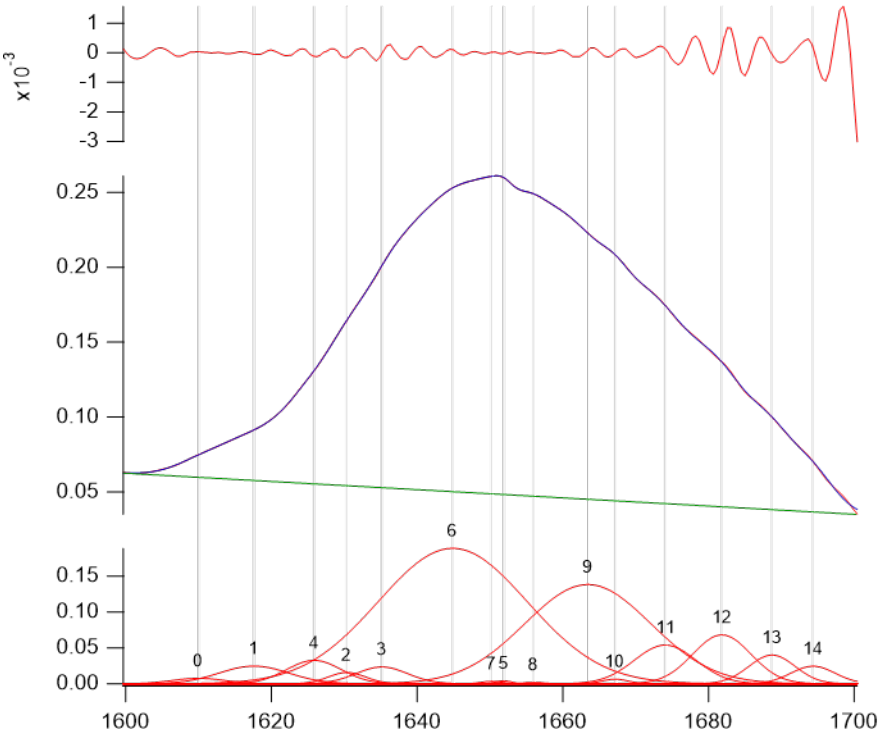


Strained SF-SMP, No MeOH, 70 °C Trial

FTIR Background Subtraction



FTIR Deconvolution



Strained SF-SMP, No MeOH, 70 °C Trial

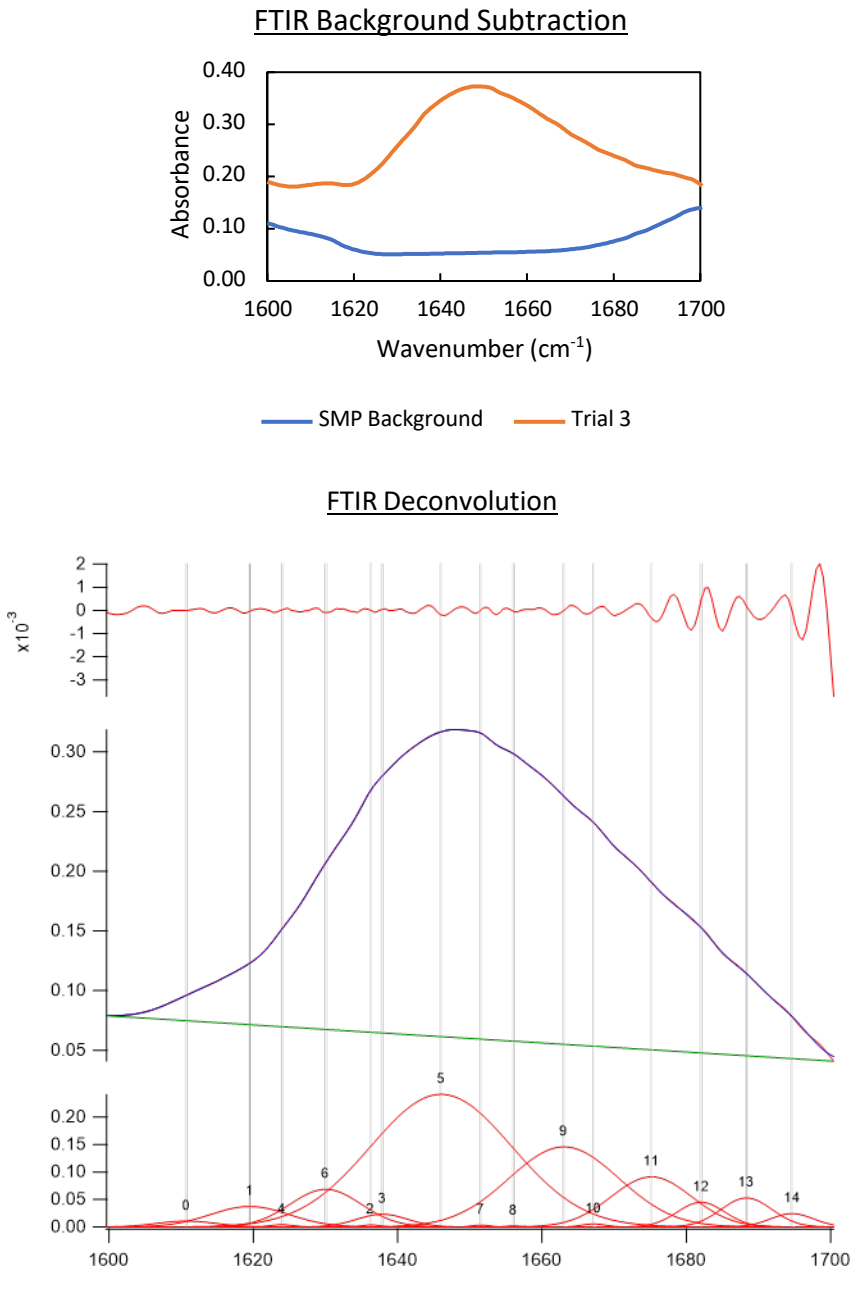
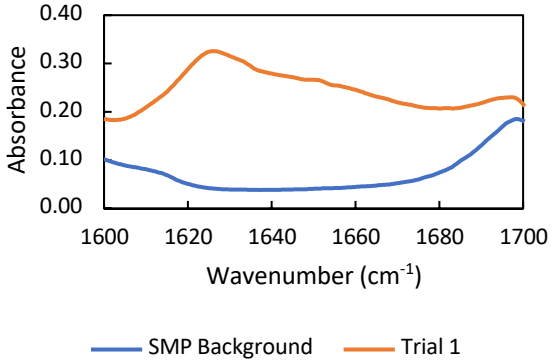


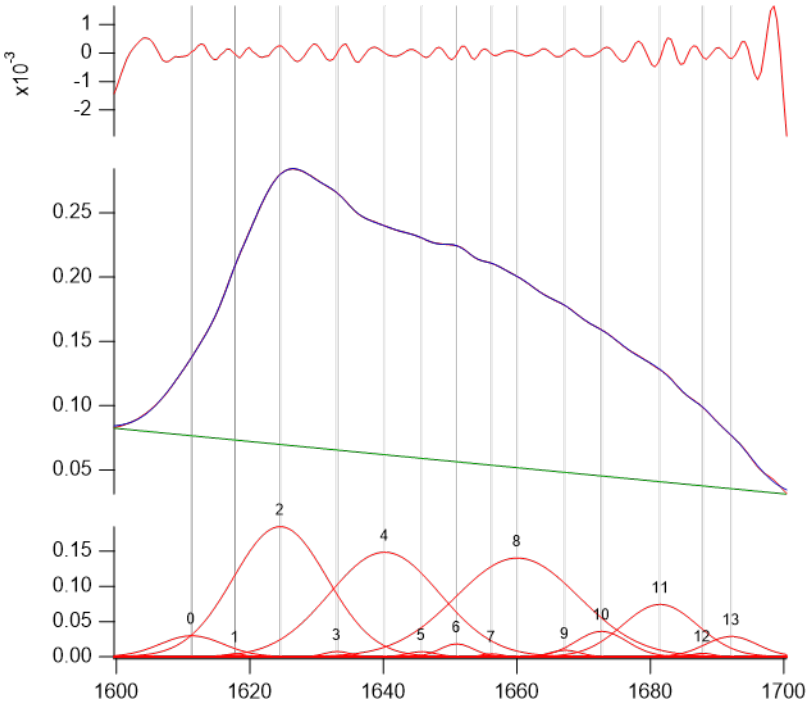
FIGURE S12. Deconvolution of FTIR spectra within the Amide I region (1600-1700 cm^{-1}) following 70 °C heating. (Top panels) Multiple linear regression is used to subtract the contribution of the SMP substrate (blue) from the total signal (SF + strained SMP) with no methanol treatment (orange) through decomposition. Three independent replicates were measured for each condition (n=3). (Bottom panels) Deconvolution results of FTIR spectra for SF-SMP with no methanol treatment. In the middle panel, raw data (red) and reproduced spectra (blue) are depicted, with the blue line representing the summation of the deconvoluted peaks. Residual fit errors are depicted in the top panels. The bottom panel shows the results from the deconvolution process; secondary structures are assigned based on peak locations within this panel. The normalized peak areas were used to determine the secondary structures found in the film.

Strained SF-SMP, With MeOH, 40 °C Trial

FTIR Background Subtraction

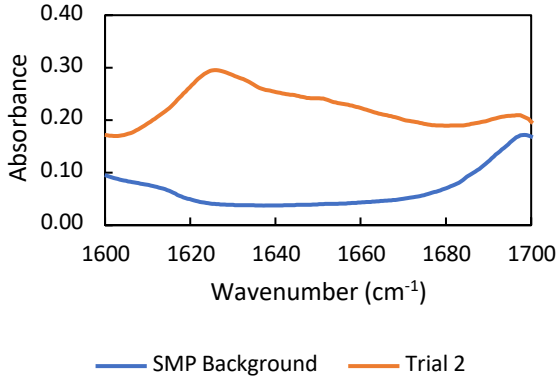


FTIR Deconvolution

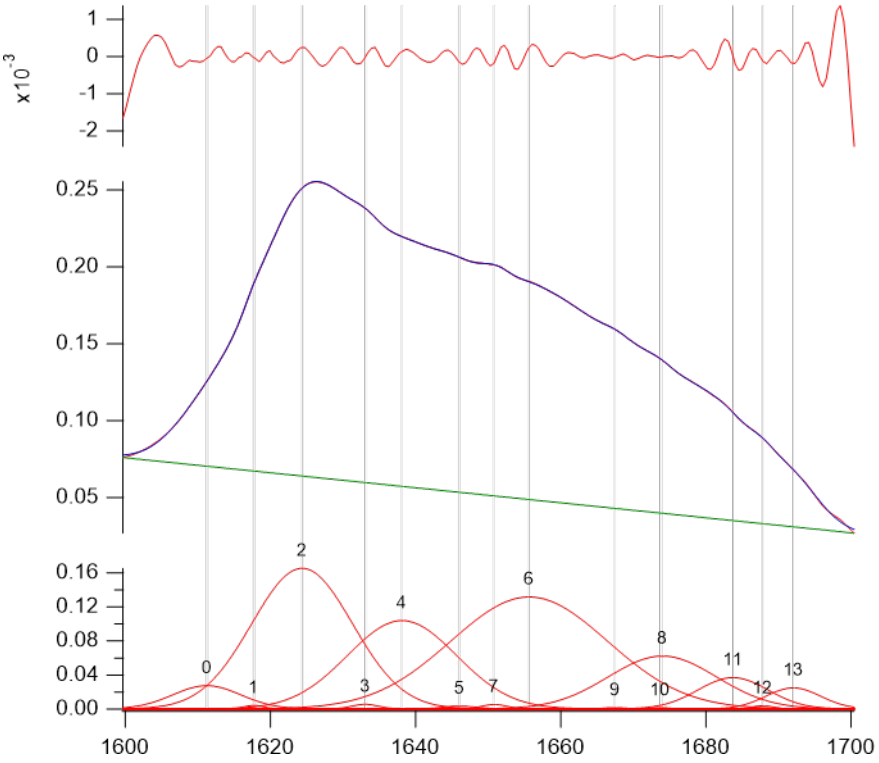


Strained SF-SMP, With MeOH, 40 °C Trial

FTIR Background Subtraction



FTIR Deconvolution



Strained SF-SMP, With MeOH, 40 °C Trial

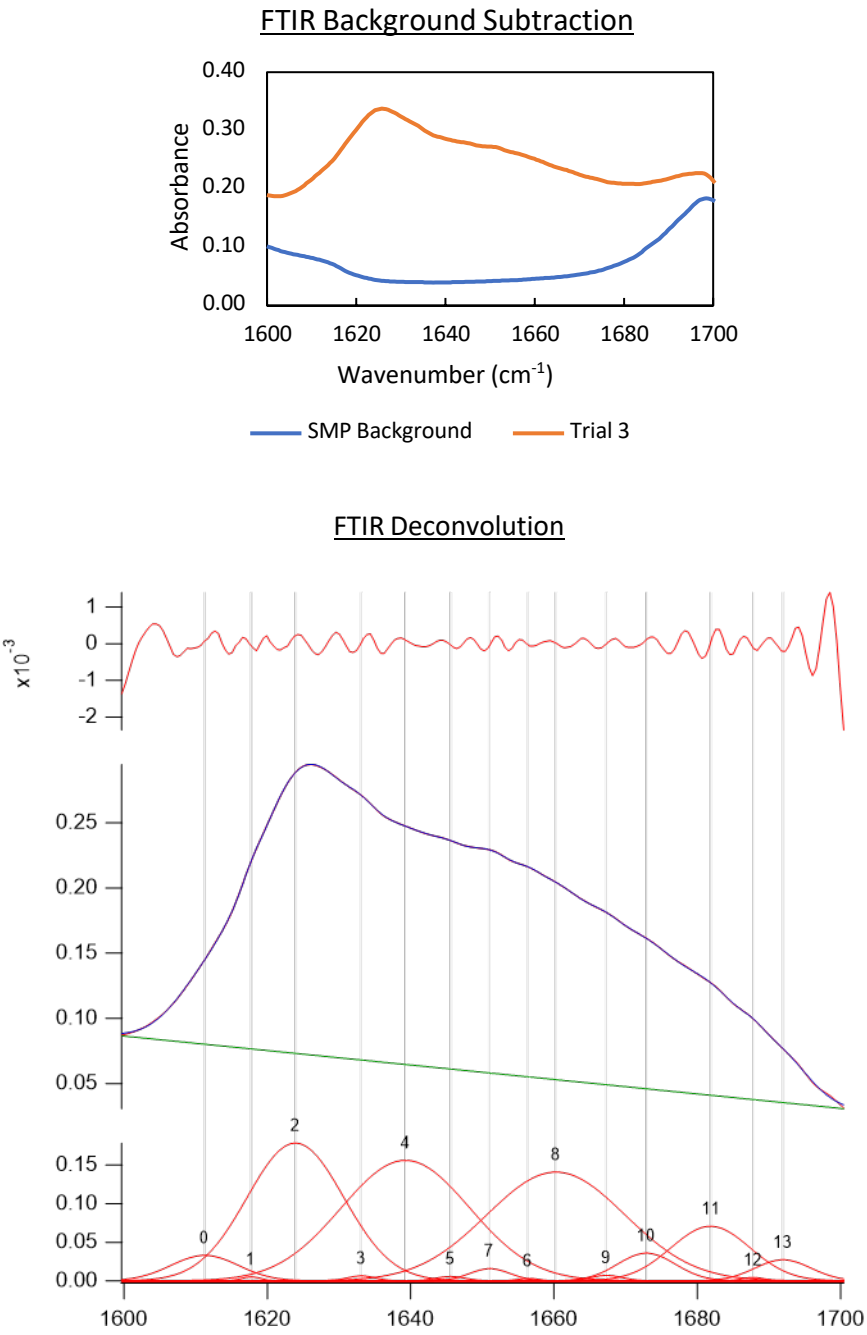
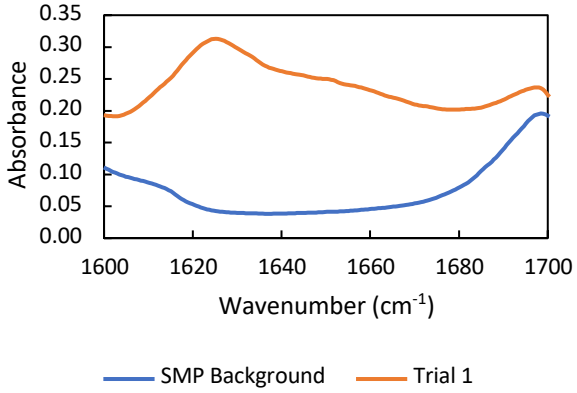


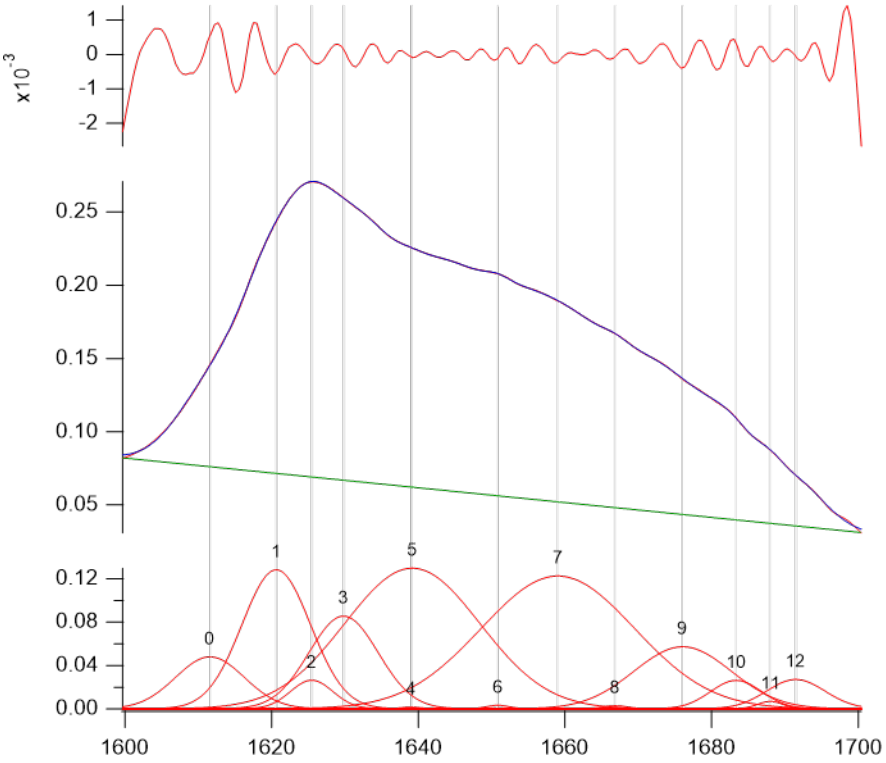
FIGURE S13. Deconvolution of FTIR spectra within the Amide I region (1600-1700 cm^{-1}) following 40 °C heating. (Top panels) Multiple linear regression is used to subtract the contribution of the SMP substrate (blue) from the total signal (SF + strained SMP) with methanol treatment (orange) through decomposition. Three independent replicates were measured for each condition (n=3). (Bottom panels) Deconvolution results of FTIR spectra for SF-SMP with methanol treatment. In the middle panel, raw data (red) and reproduced spectra (blue) are depicted, with the blue line representing the summation of the deconvoluted peaks. Residual fit errors are depicted in the top panels. The bottom panel shows the results from the deconvolution process; secondary structures are assigned based on peak locations within this panel. The normalized peak areas were used to determine the secondary structures found in the film.

Strained SF-SMP, With MeOH, 70 °C Trial 1

FTIR Background Subtraction

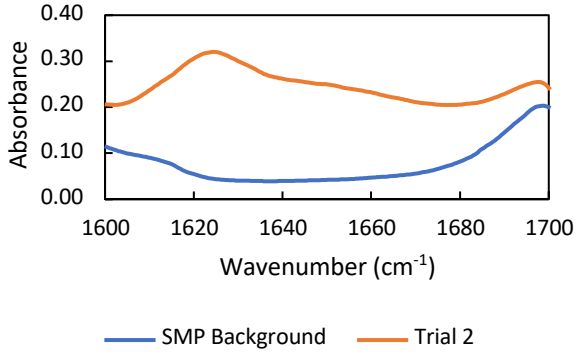


FTIR Deconvolution

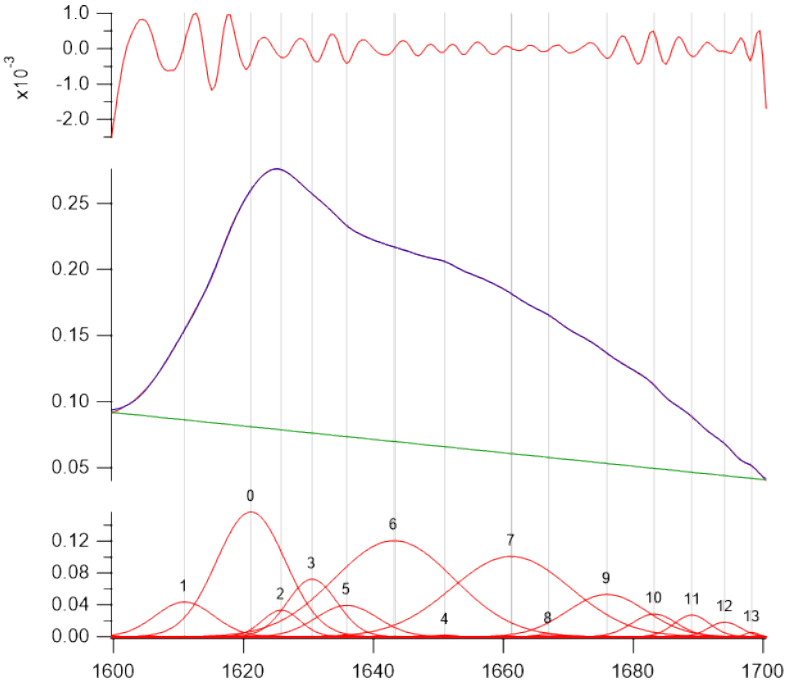


Strained SF-SMP, With MeOH, 70 °C Trial

FTIR Background Subtraction



FTIR Deconvolution



Strained SF-SMP, With MeOH, 70 °C Trial

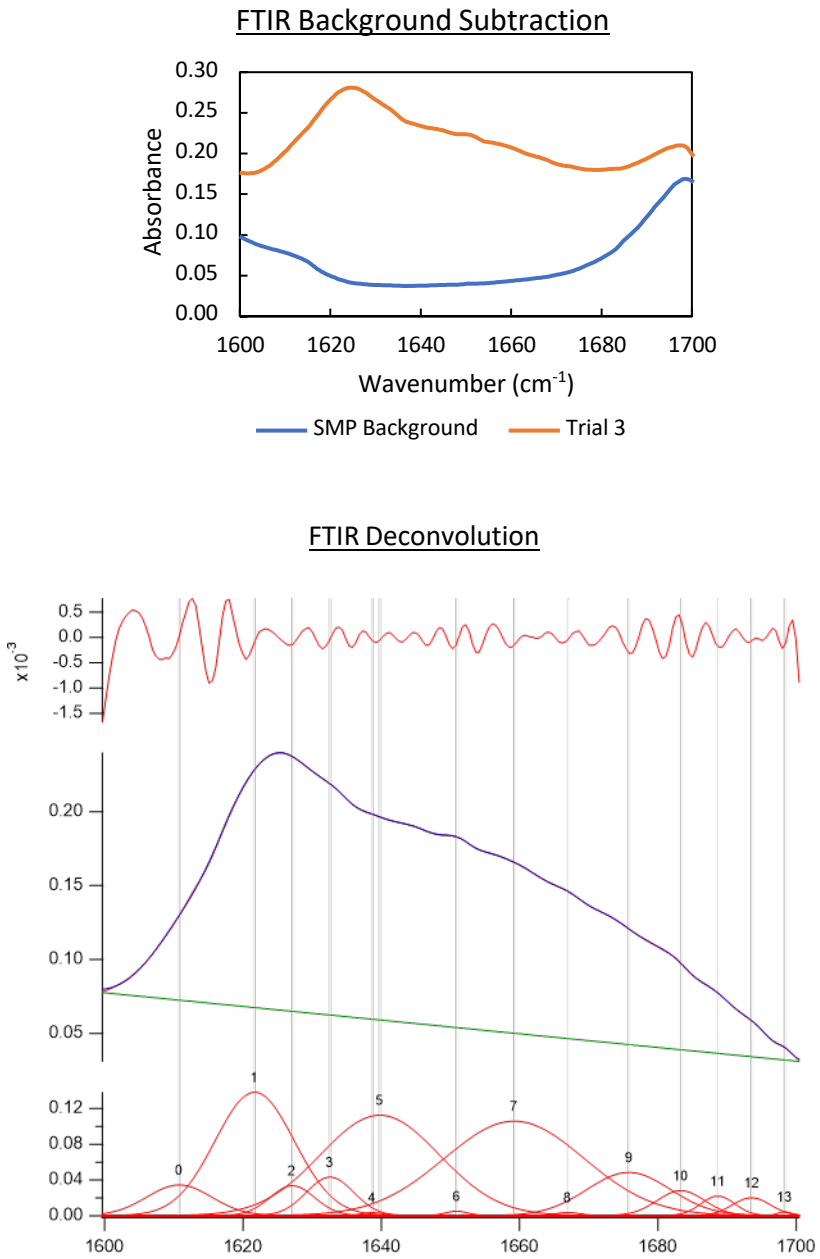
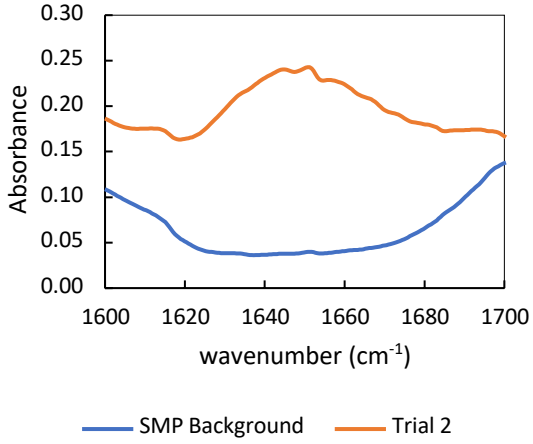


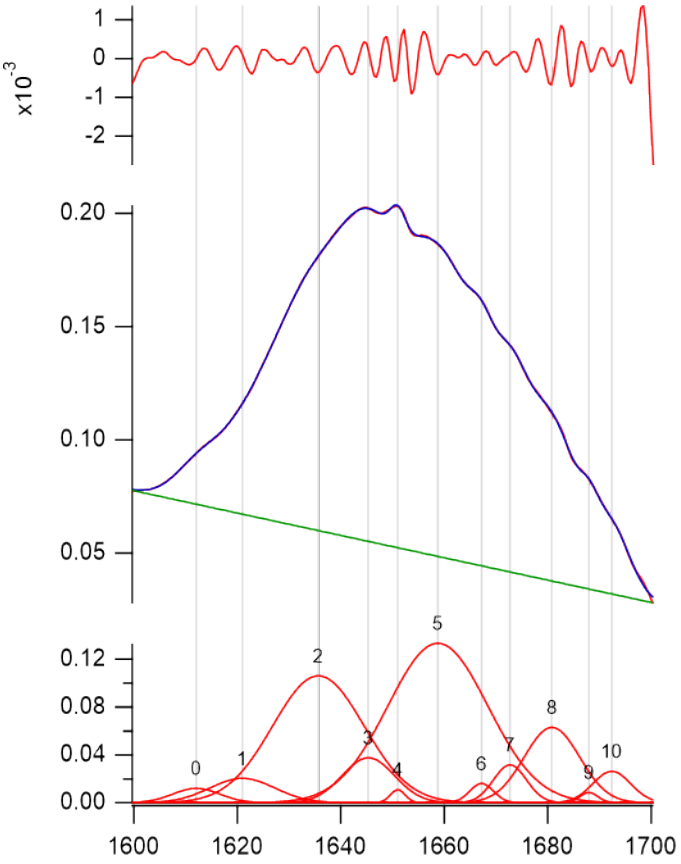
FIGURE S14. Deconvolution of FTIR spectra within the Amide I region (1600-1700 cm^{-1}) following 70 °C heating. (Top panels) Multiple linear regression is used to subtract the contribution of the SMP substrate (blue) from the total signal (SF + strained SMP) with methanol treatment (orange) through decomposition. Three independent replicates were measured for each condition (n=3). (Bottom panels) Deconvolution results of FTIR spectra for SF-SMP with methanol treatment. In the middle panel, raw data (red) and reproduced spectra (blue) are depicted, with the blue line representing the summation of the deconvoluted peaks. Residual fit errors are depicted in the top panels. The bottom panel shows the results from the deconvolution process; secondary structures are assigned based on peak locations within this panel. The normalized peak areas were used to determine the secondary structures found in the film.

Strained SF-SMP, No MeOH, 40 °C Trial

FTIR Background Subtraction

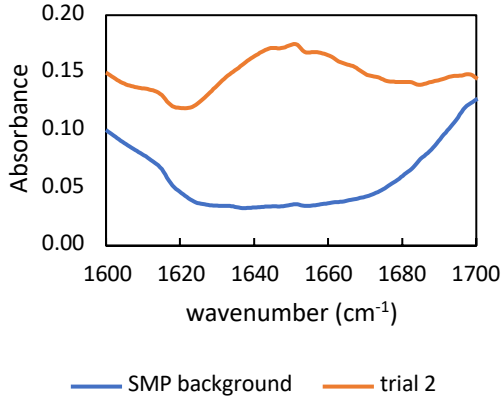


FTIR Deconvolution

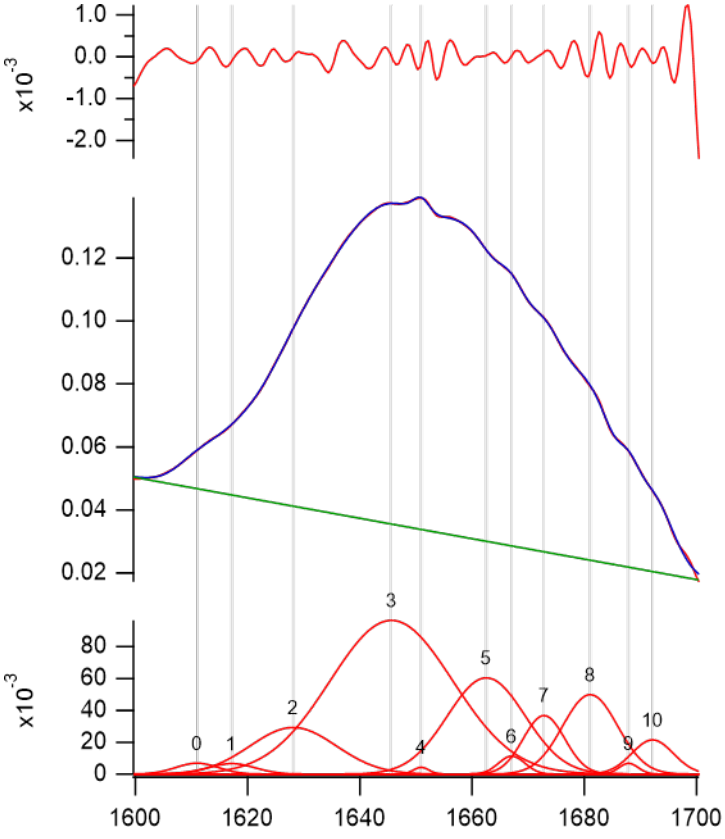


Strained SF-SMP, No MeOH, 40 °C Trial

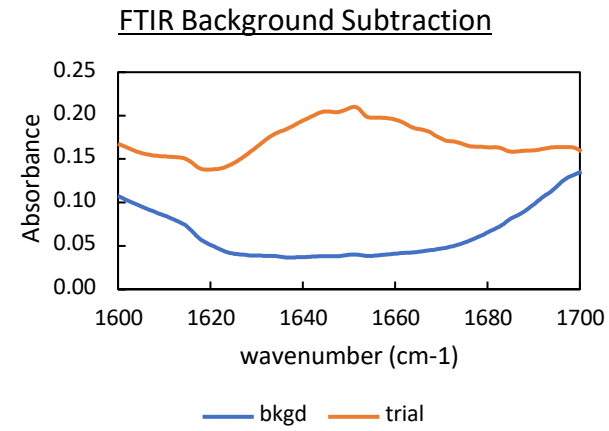
FTIR Background Subtraction



FTIR Deconvolution



Strained SF-SMP, No MeOH, 40 °C Trial



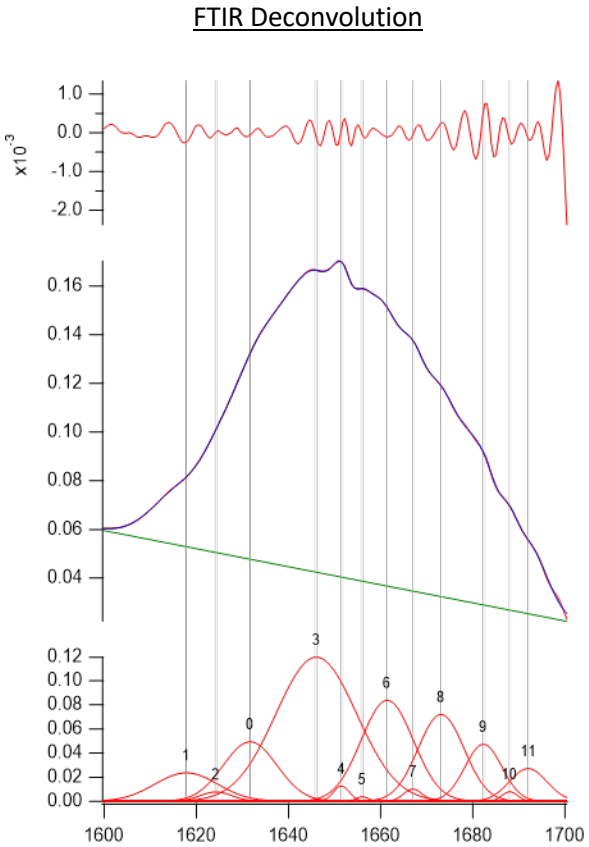
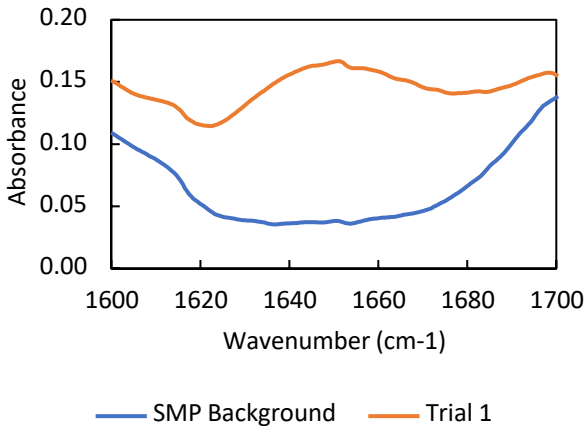


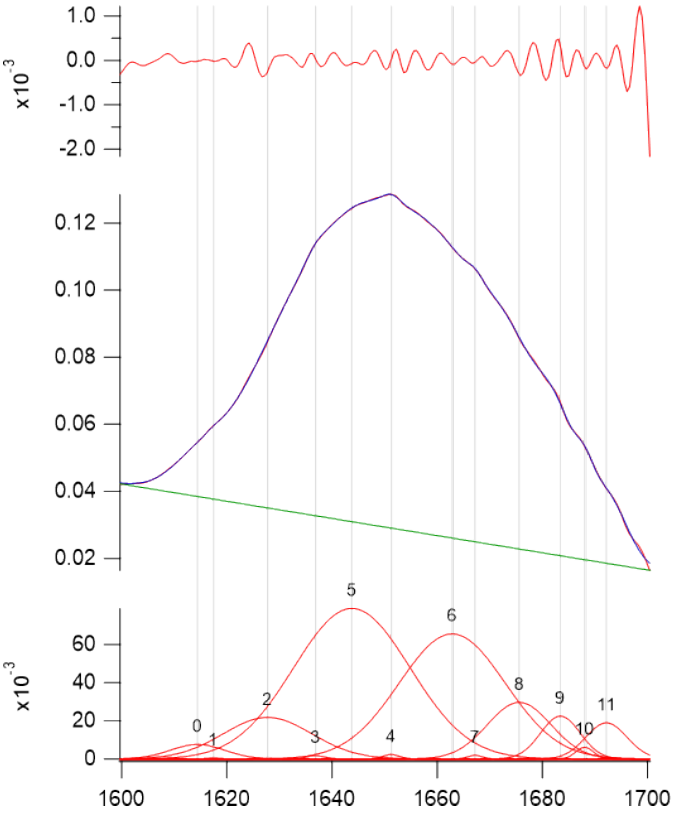
FIGURE S15. Deconvolution of FTIR spectra within the Amide I region (1600-1700 cm^{-1}) following 40 °C heating. (Top panels) Multiple linear regression is used to subtract the contribution of the SMP substrate (blue) from the total signal (SF + strained SMP) with no methanol treatment (orange) through decomposition. Three independent replicates were measured for each condition (n=3). (Bottom panels) Deconvolution results of FTIR spectra for SF-SMP with no methanol treatment. In the middle panel, raw data (red) and reproduced spectra (blue) are depicted, with the blue line representing the summation of the deconvoluted peaks. Residual fit errors are depicted in the top panels. The bottom panel shows the results from the deconvolution process; secondary structures are assigned based on peak locations within this panel. The normalized peak areas were used to determine the secondary structures found in the film.

Strained SF-SMP, No MeOH, 70 °C Trial 1

FTIR Background Subtraction

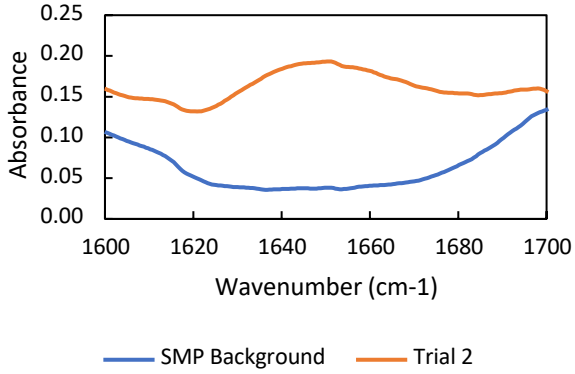


FTIR Deconvolution

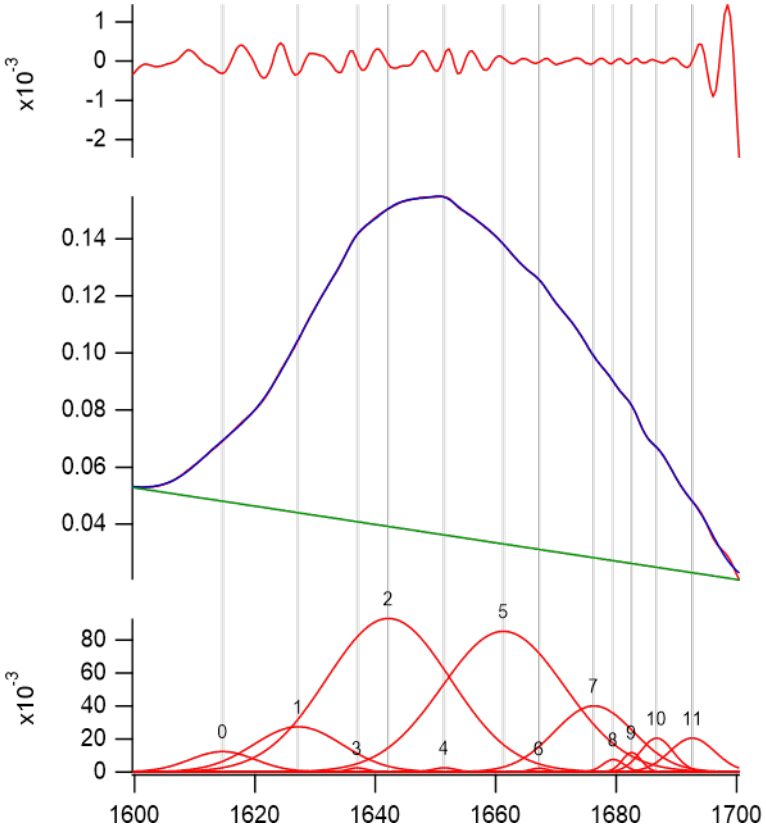


Strained SF-SMP, No MeOH, 70 °C Trial

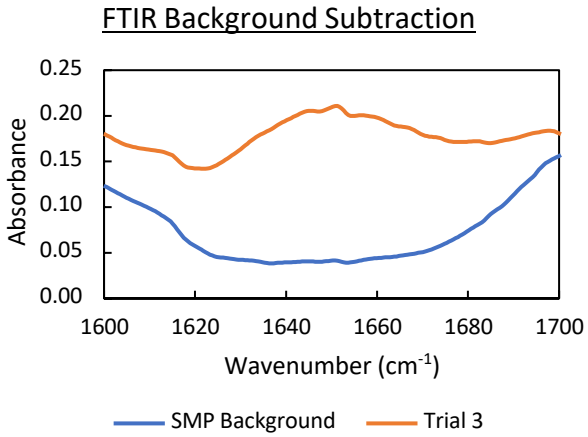
FTIR Background Subtraction



FTIR Deconvolution



Strained SF-SMP, No MeOH, 70 °C Trial



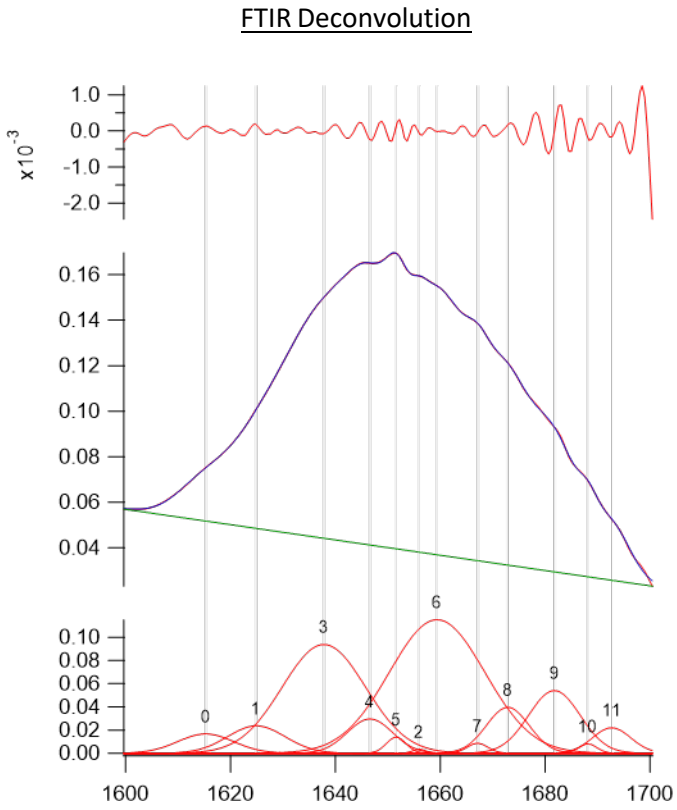
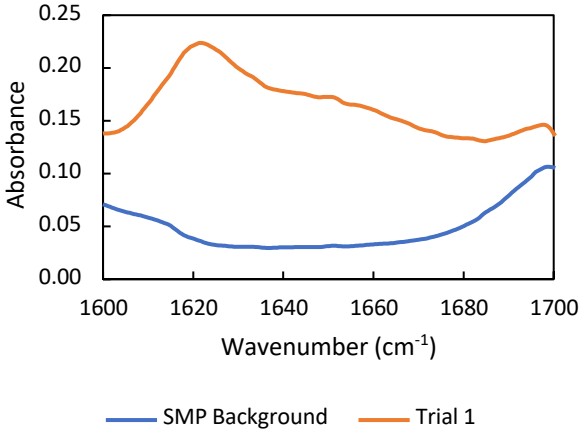


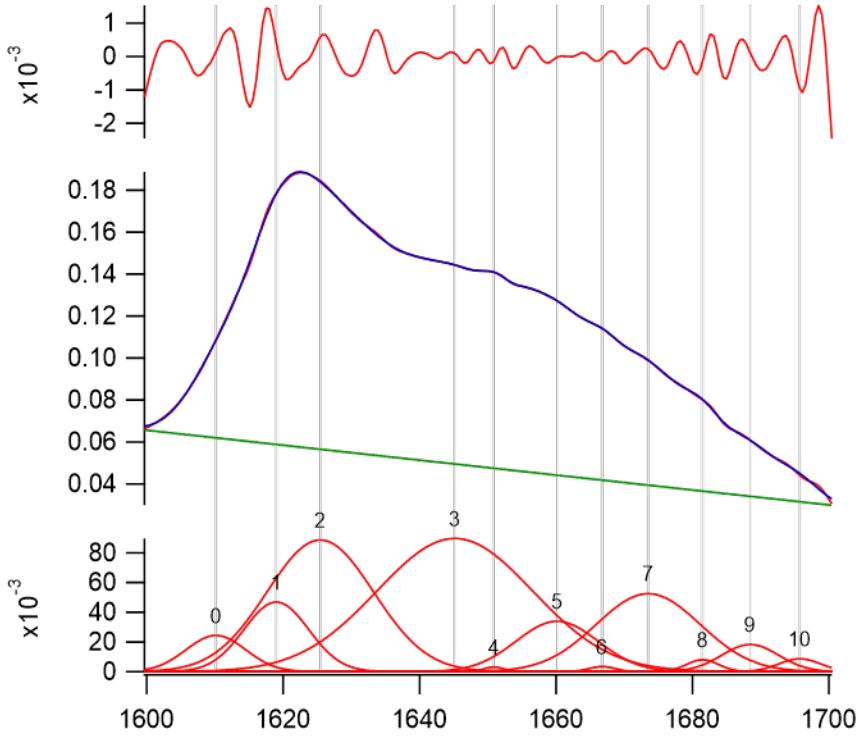
FIGURE S16. Deconvolution of FTIR spectra within the Amide I region (1600-1700 cm^{-1}) following 70 °C heating. (Top panels) Multiple linear regression is used to subtract the contribution of the SMP substrate (blue) from the total signal (SF + strained SMP) with no methanol treatment (orange) through decomposition. Three independent replicates were measured for each condition (n=3). (Bottom panels) Deconvolution results of FTIR spectra for SF-SMP with no methanol treatment. In the middle panel, raw data (red) and reproduced spectra (blue) are depicted, with the blue line representing the summation of the deconvoluted peaks. Residual fit errors are depicted in the top panels. The bottom panel shows the results from the deconvolution process; secondary structures are assigned based on peak locations within this panel. The normalized peak areas were used to determine the secondary structures found in the film.

Strained SF-SMP, With MeOH, 40 °C Trial

FTIR Background Subtraction

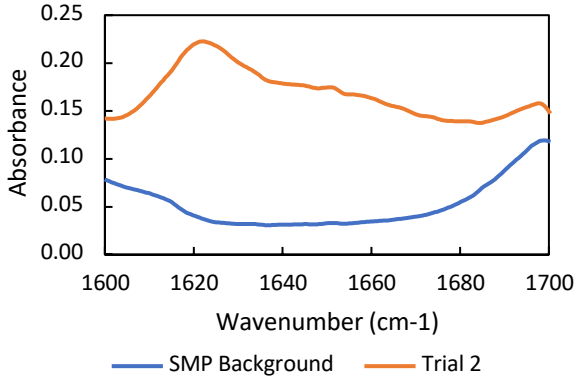


FTIR Deconvolution

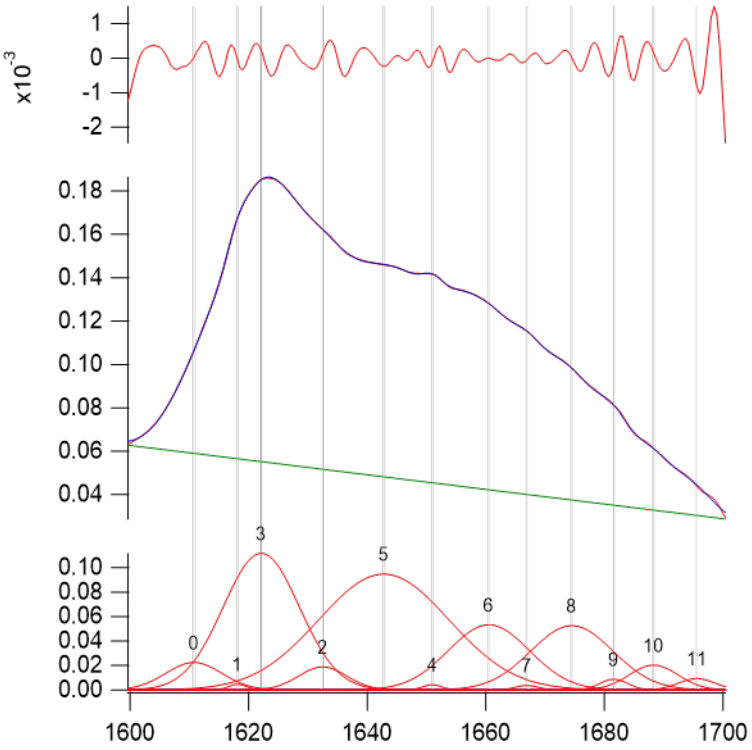


Strained SF-SMP, With MeOH, 40 °C Trial

FTIR Background Subtraction



FTIR Deconvolution



Strained SF-SMP, With MeOH, 40 °C Trial

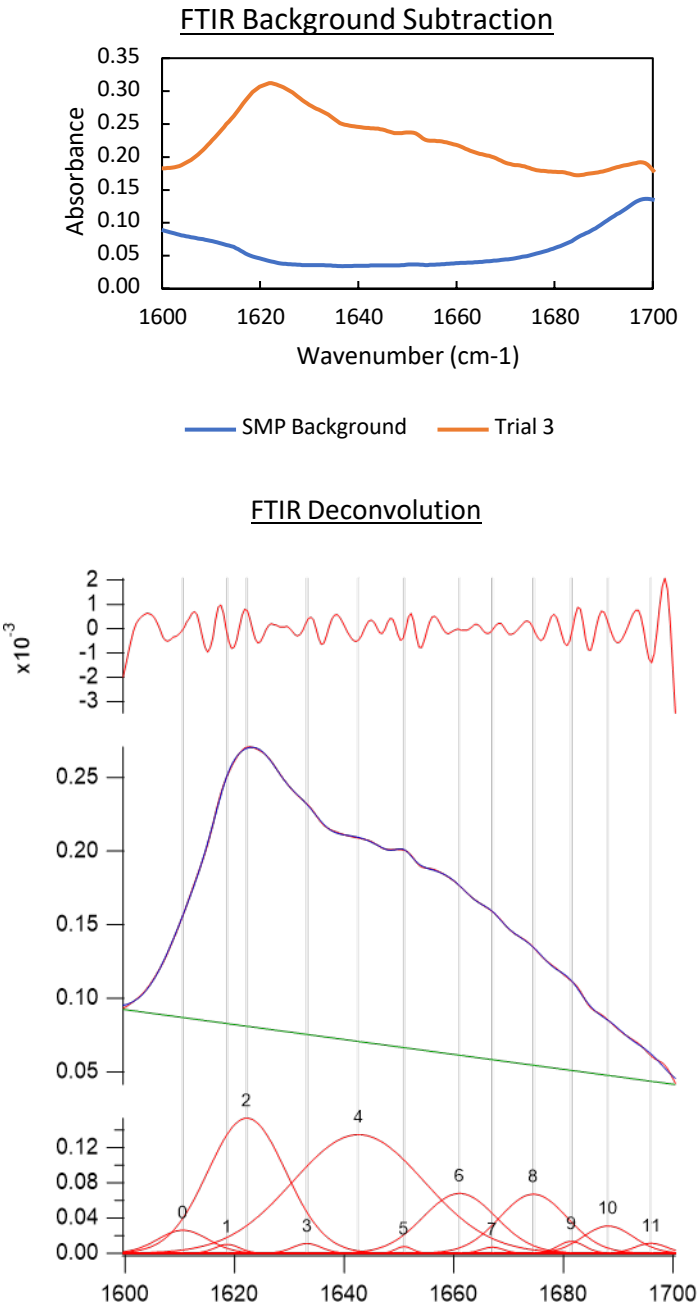
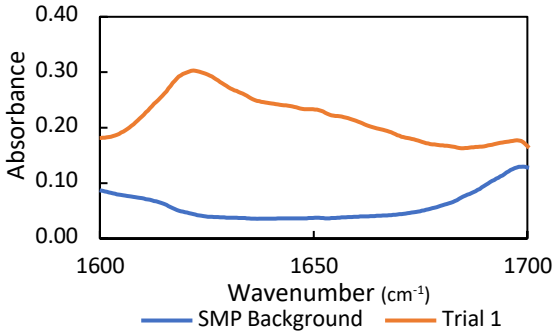


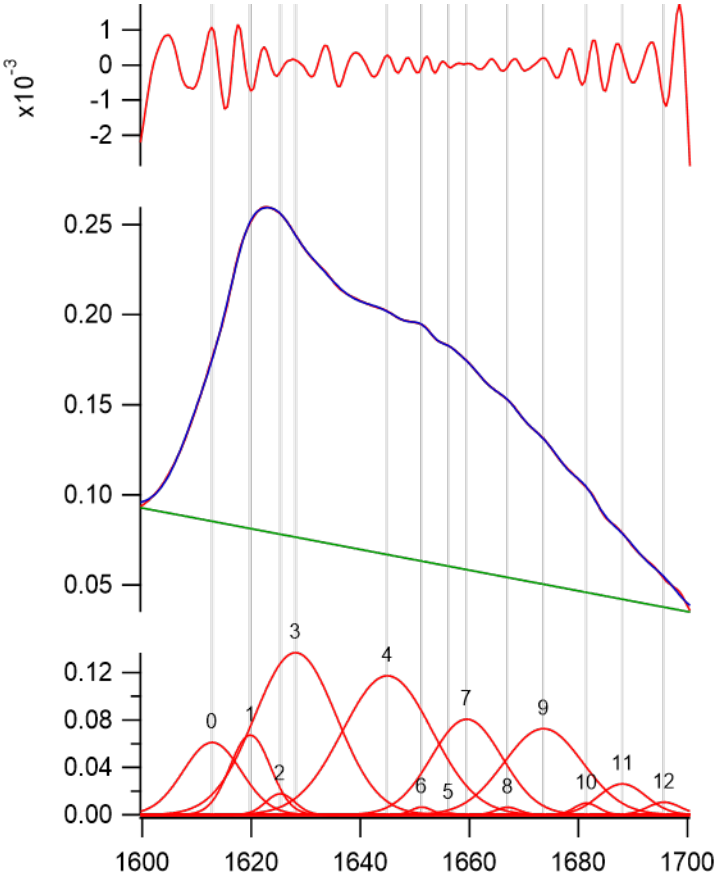
FIGURE S17. Deconvolution of FTIR spectra within the Amide I region (1600-1700 cm^{-1}) following 40 °C heating. (Top panels) Multiple linear regression is used to subtract the contribution of the SMP substrate (blue) from the total signal (SF + strained SMP) with methanol treatment (orange) through decomposition. Three independent replicates were measured for each condition (n=3). (Bottom panels) Deconvolution results of FTIR spectra for SF-SMP with methanol treatment. In the middle panel, raw data (red) and reproduced spectra (blue) are depicted, with the blue line representing the summation of the deconvoluted peaks. Residual fit errors are depicted in the top panels. The bottom panel shows the results from the deconvolution process; secondary structures are assigned based on peak locations within this panel. The normalized peak areas were used to determine the secondary structures found in the film.

Strained SF-SMP, With MeOH, 70 °C Trial 1

FTIR Background Subtraction

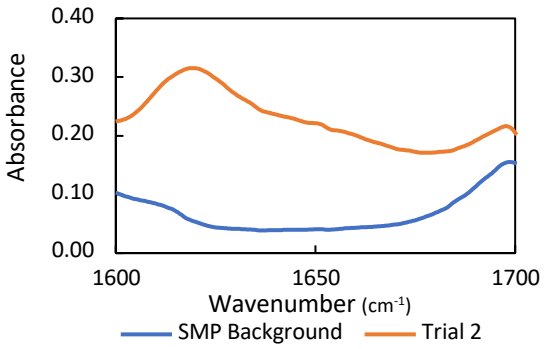


FTIR Deconvolution

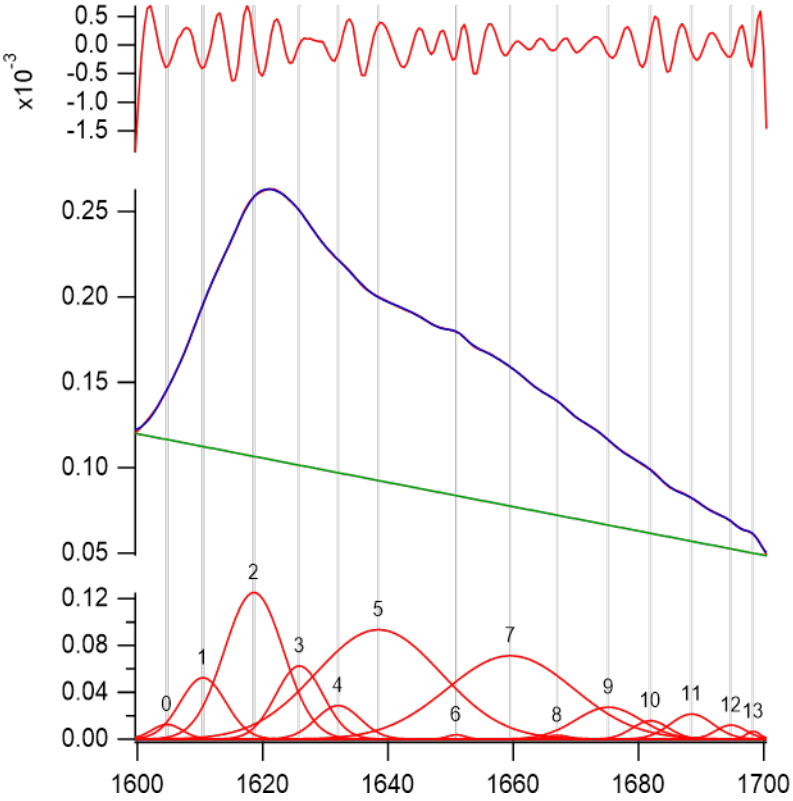


Strained SF-SMP, With MeOH, 70 °C Trial

FTIR Background Subtraction



FTIR Deconvolution



Strained SF-SMP, With MeOH, 70 °C Trial 52

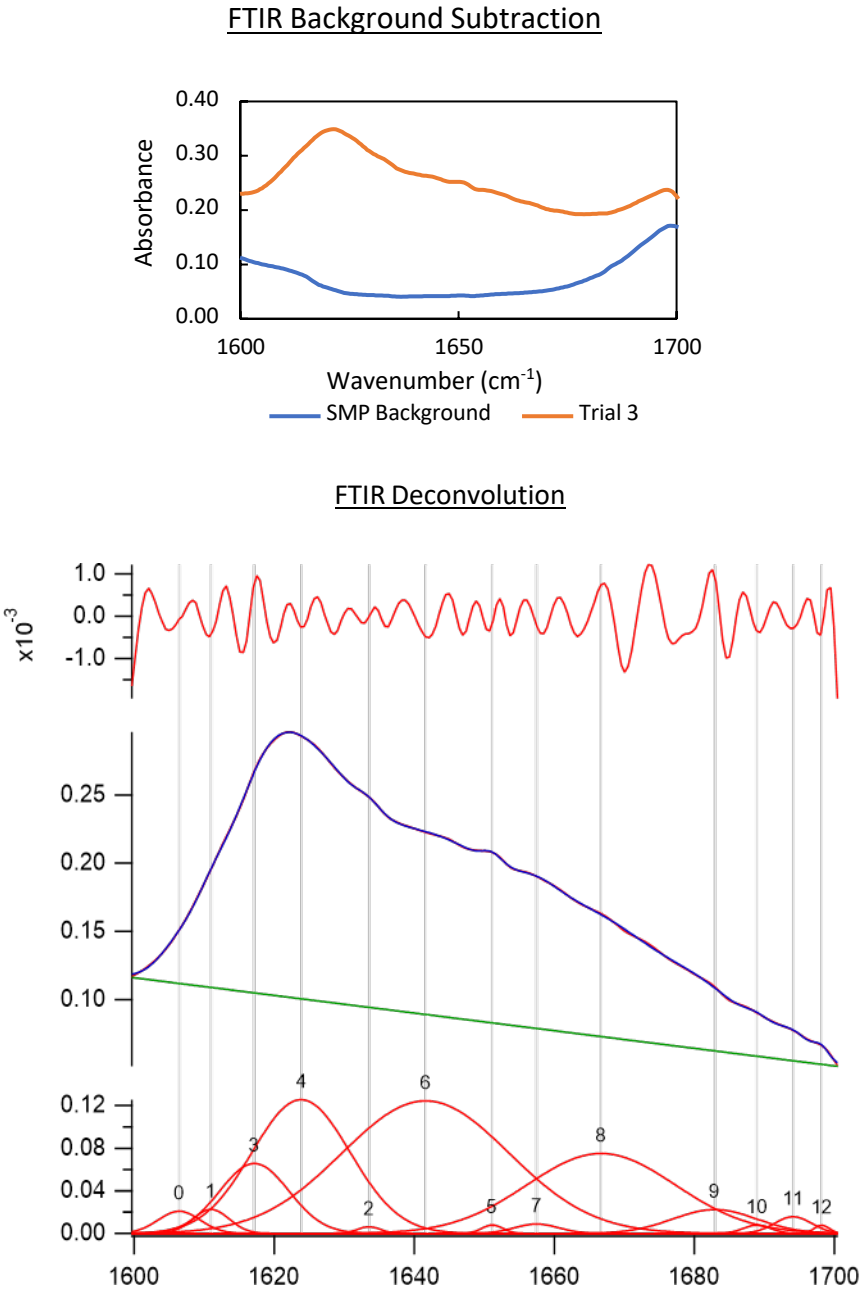
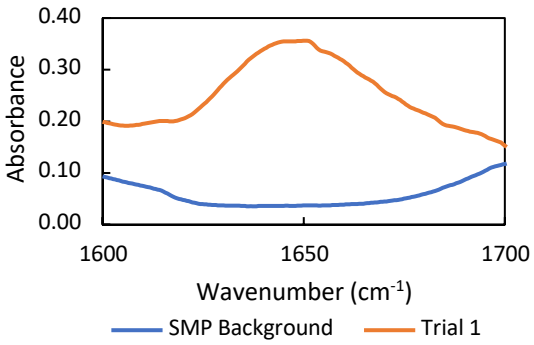


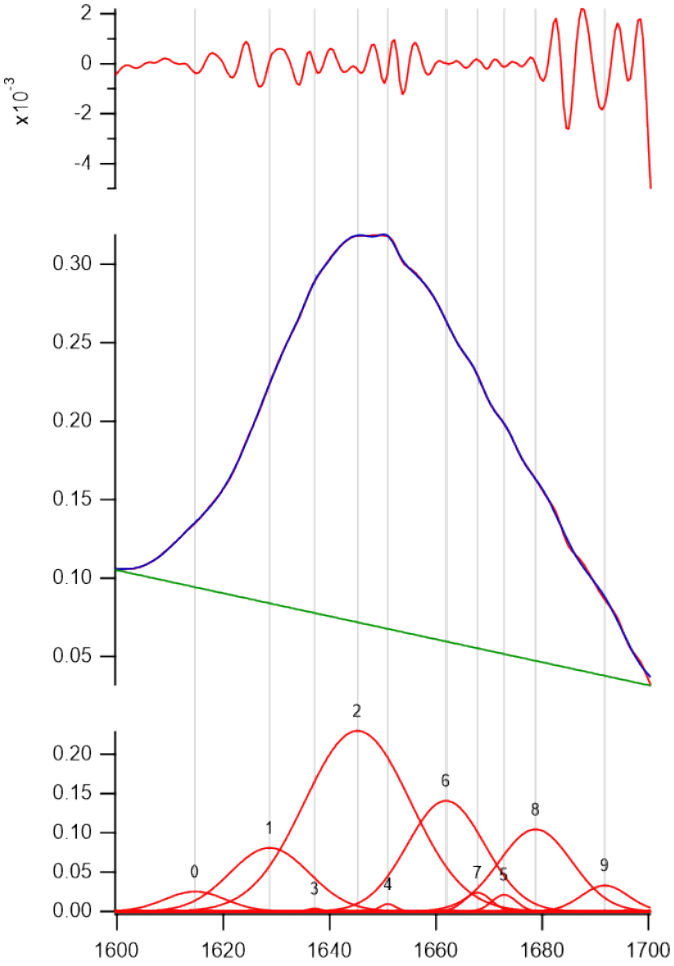
FIGURE S18. Deconvolution of FTIR spectra within the Amide I region (1600-1700 cm^{-1}) following 70 °C heating. (Top panels) Multiple linear regression is used to subtract the contribution of the SMP substrate (blue) from the total signal (SF + strained SMP) with methanol treatment (orange) through decomposition. Three independent replicates were measured for each condition (n=3). (Bottom panels) Deconvolution results of FTIR spectra for SF-SMP with methanol treatment. In the middle panel, raw data (red) and reproduced spectra (blue) are depicted, with the blue line representing the summation of the deconvoluted peaks. Residual fit errors are depicted in the top panels. The bottom panel shows the results from the deconvolution process; secondary structures are assigned based on peak locations within this panel. The normalized peak areas were used to determine the secondary structures found in the film.

Unstrained SF-SMP, No MeOH, 40 °C Trial 53

FTIR Background Subtraction

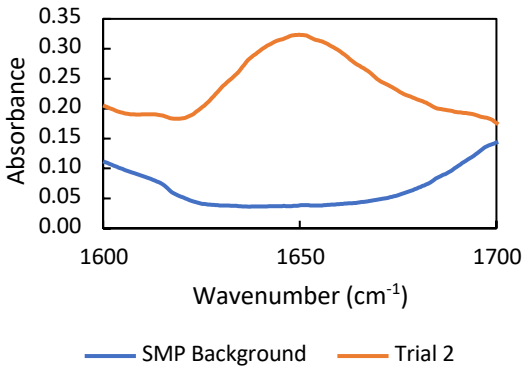


FTIR Deconvolution

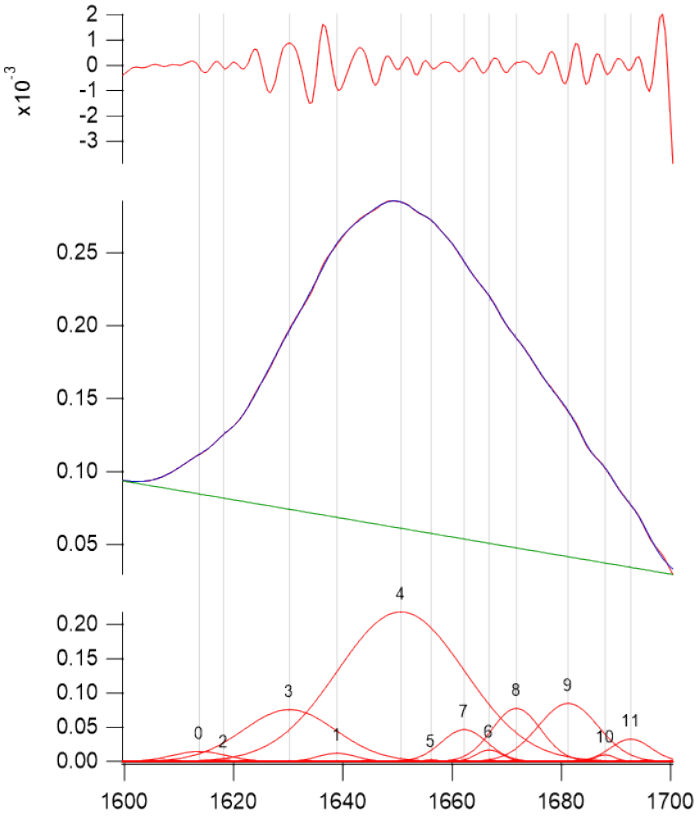


Unstrained SF-SMP, No MeOH, 40 °C Trial

FTIR Background Subtraction



FTIR Deconvolution



Unstrained SF-SMP, No MeOH, 40 °C Trial

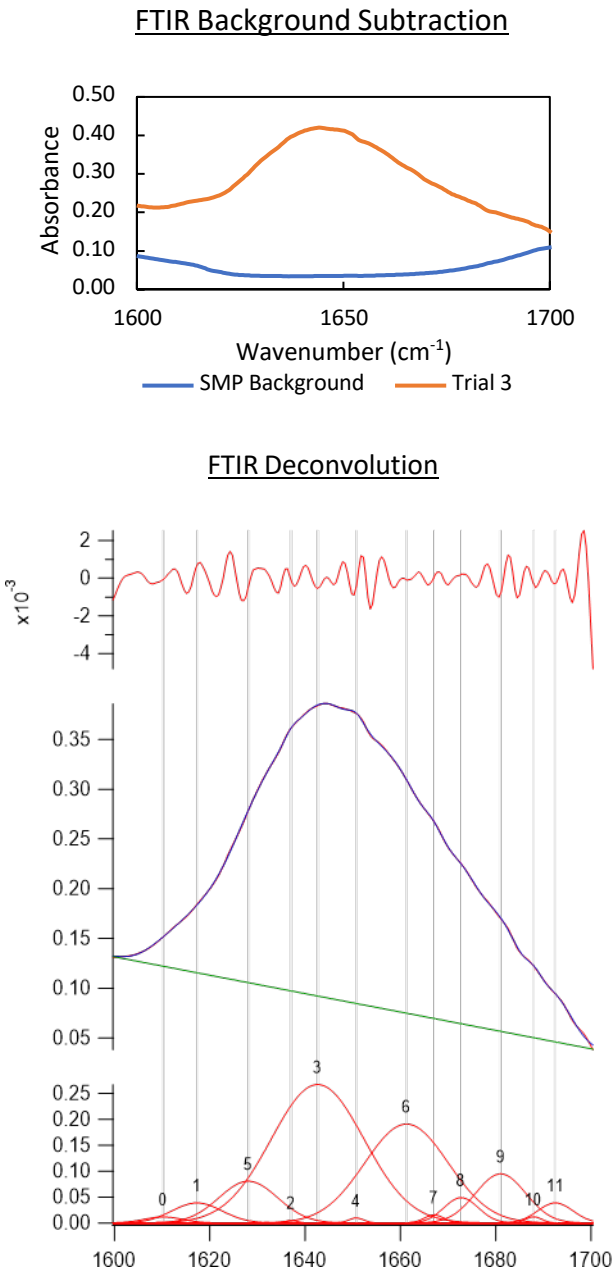
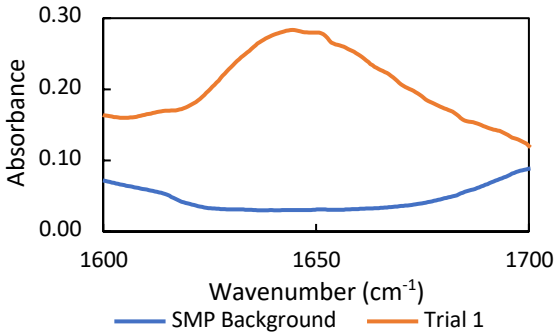


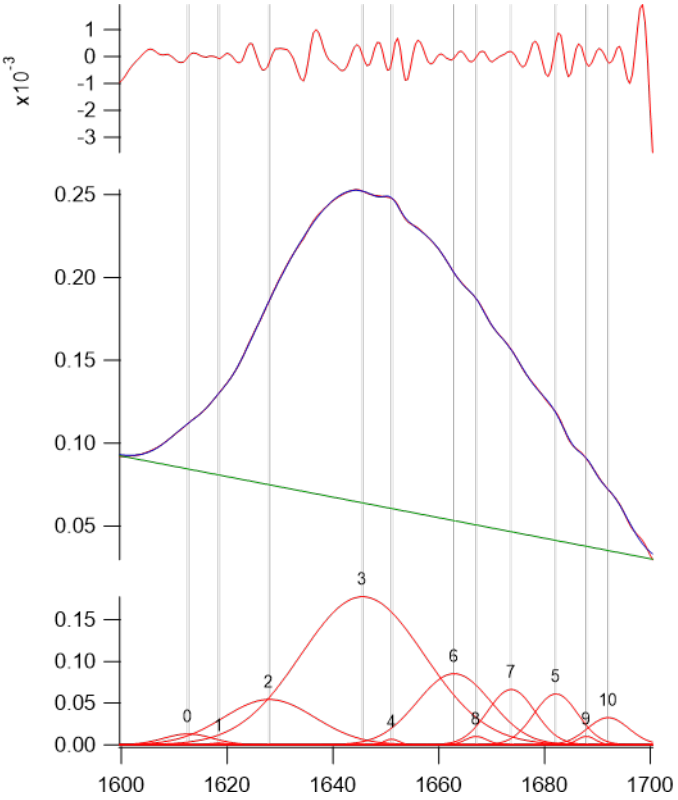
FIGURE S19. Deconvolution of FTIR spectra within the Amide I region (1600-1700 cm^{-1}) following 40 °C heating. (Top panels) Multiple linear regression is used to subtract the contribution of the SMP substrate (blue) from the total signal (SF + unstrained SMP) with no methanol treatment (orange) through decomposition. Three independent replicates were measured for each condition (n=3). (Bottom panels) Deconvolution results of FTIR spectra for SF-SMP with no methanol treatment. In the middle panel, raw data (red) and reproduced spectra (blue) are depicted, with the blue line representing the summation of the deconvoluted peaks. Residual fit errors are depicted in the top panels. The bottom panel shows the results from the deconvolution process; secondary structures are assigned based on peak locations within this panel. The normalized peak areas were used to determine the secondary structures found in the film.

Unstrained SF-SMP, No MeOH, 70 °C Trial

FTIR Background Subtraction

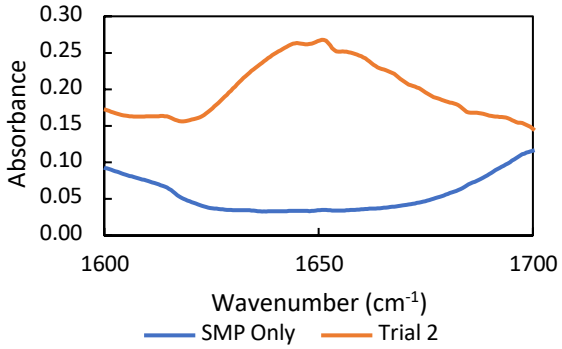


FTIR Deconvolution

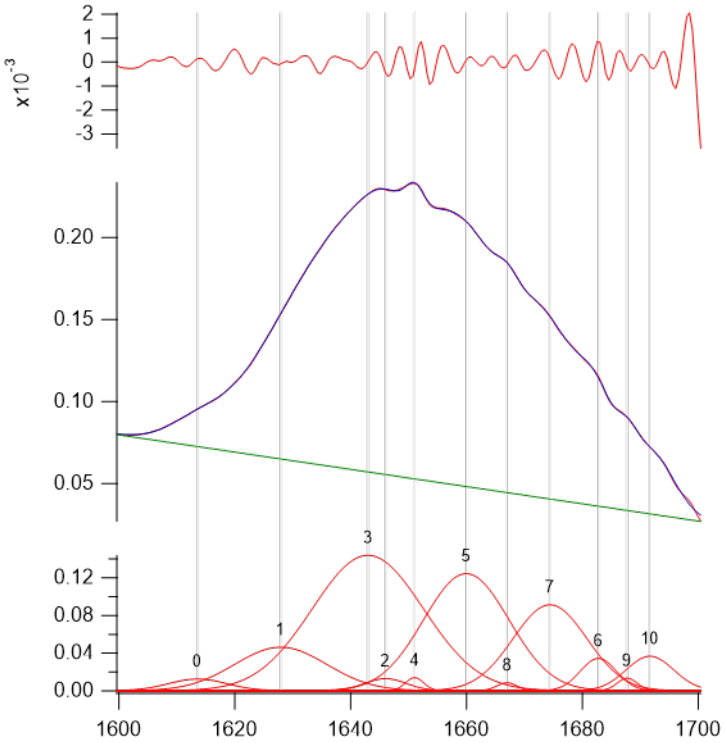


Unstrained SF-SMP, No MeOH, 70 °C Trial

FTIR Background Subtraction



FTIR Deconvolution



Unstrained SF-SMP, No MeOH, 70 °C Trial

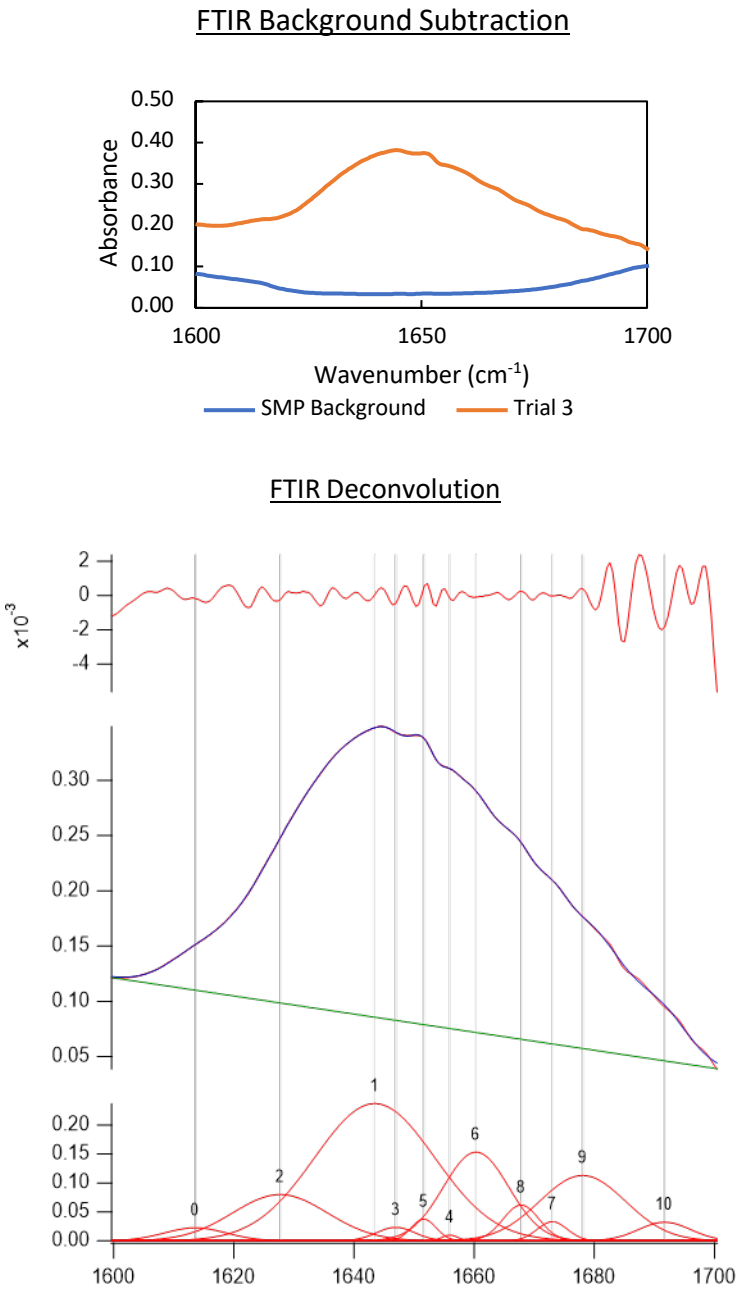
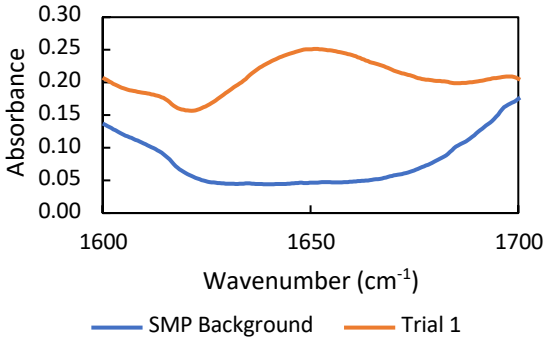


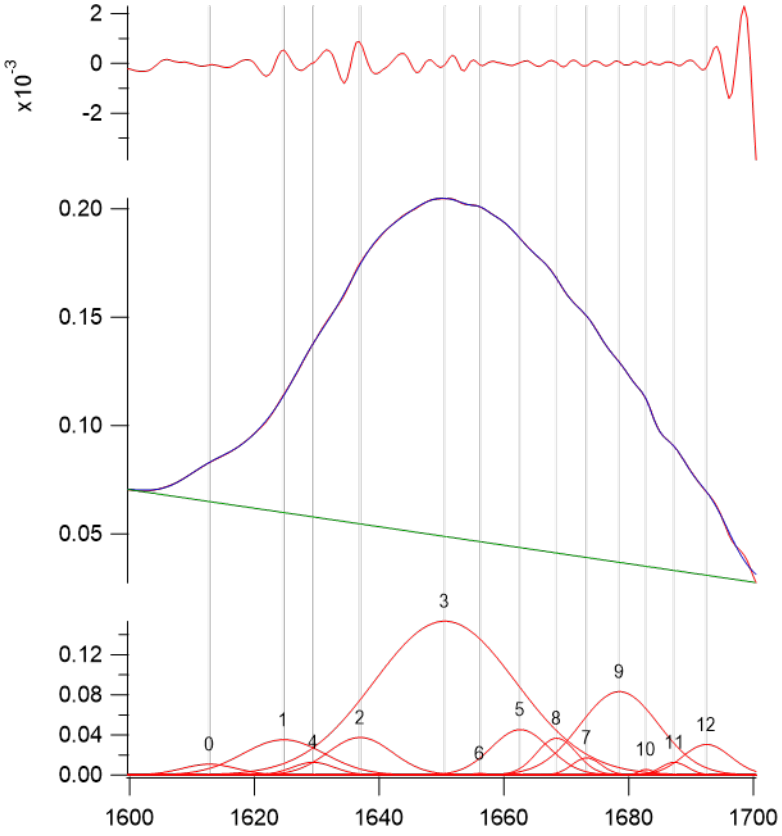
FIGURE S20. Deconvolution of FTIR spectra within the Amide I region (1600-1700 cm^{-1}) following 70 °C heating. (Top panels) Multiple linear regression is used to subtract the contribution of the SMP substrate (blue) from the total signal (SF + unstrained SMP) with no methanol treatment (orange) through decomposition. Three independent replicates were measured for each condition (n=3). (Bottom panels) Deconvolution results of FTIR spectra for SF-SMP with no methanol treatment. In the middle panel, raw data (red) and reproduced spectra (blue) are depicted, with the blue line representing the summation of the deconvoluted peaks. Residual fit errors are depicted in the top panels. The bottom panel shows the results from the deconvolution process; secondary structures are assigned based on peak locations within this panel. The normalized peak areas were used to determine the secondary structures found in the film.

Strained SF-SMP, No MeOH, 40 °C Trial

FTIR Background Subtraction

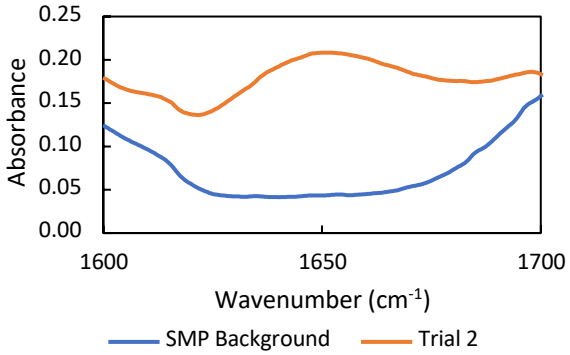


FTIR Deconvolution

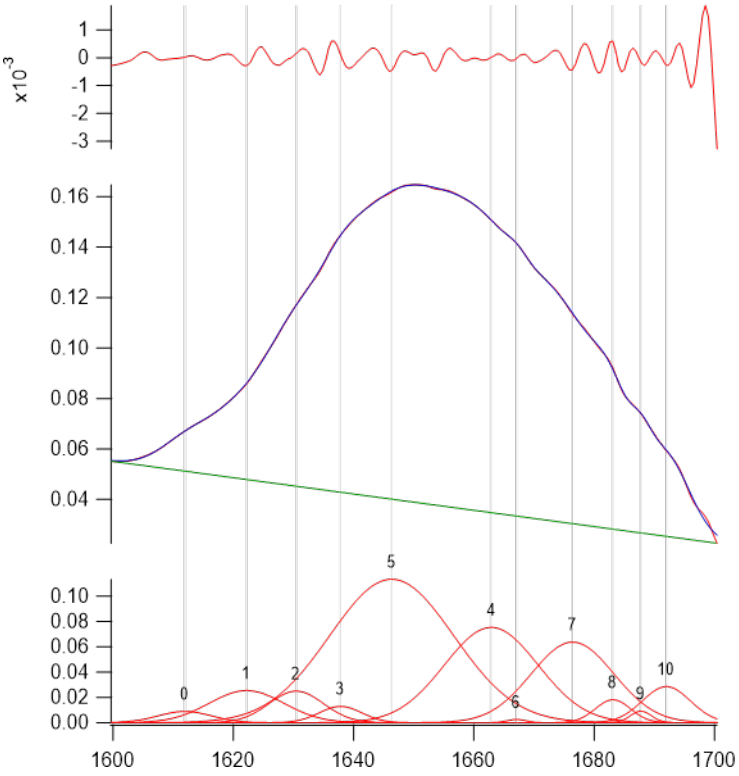


Strained SF-SMP, No MeOH, 40 °C Trial

FTIR Background Subtraction



FTIR Deconvolution



Strained SF-SMP, No MeOH, 40 °C Trial

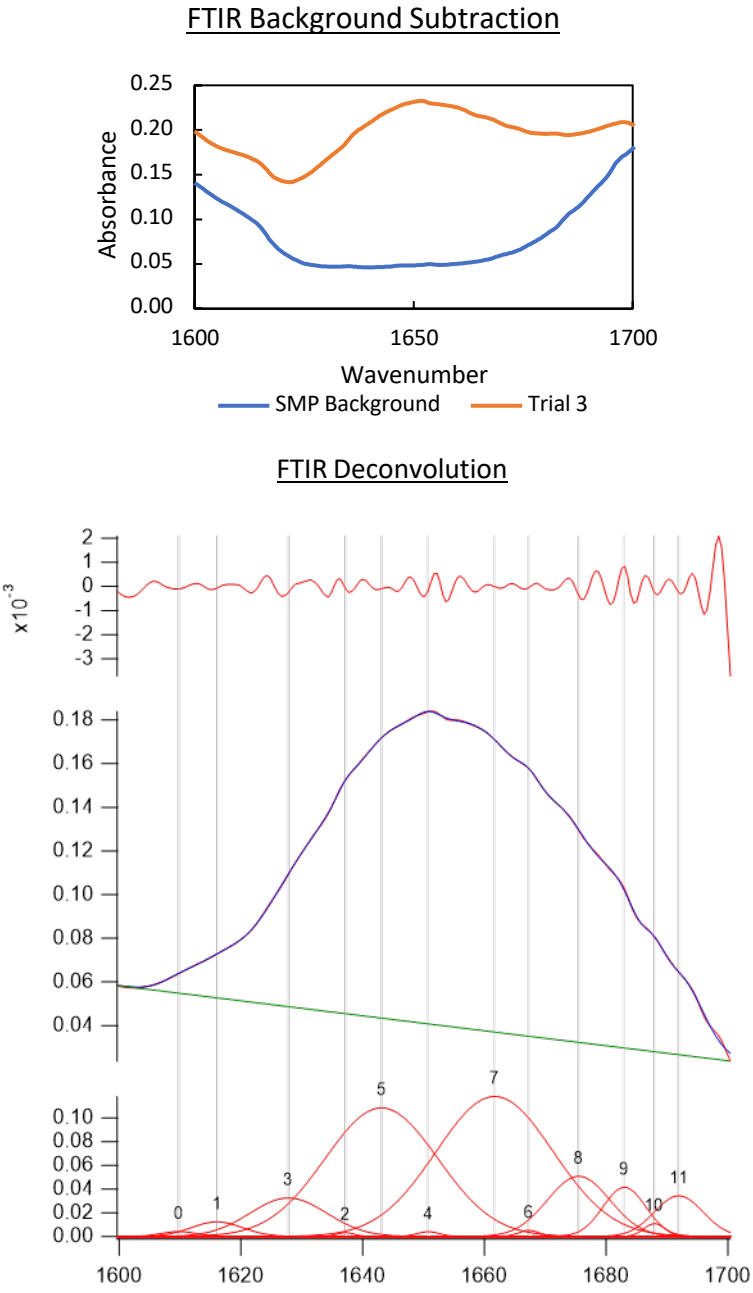
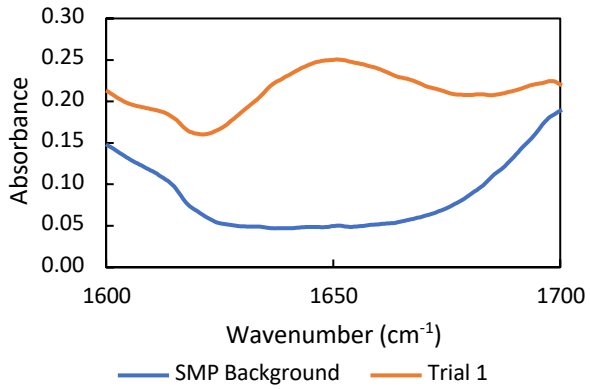


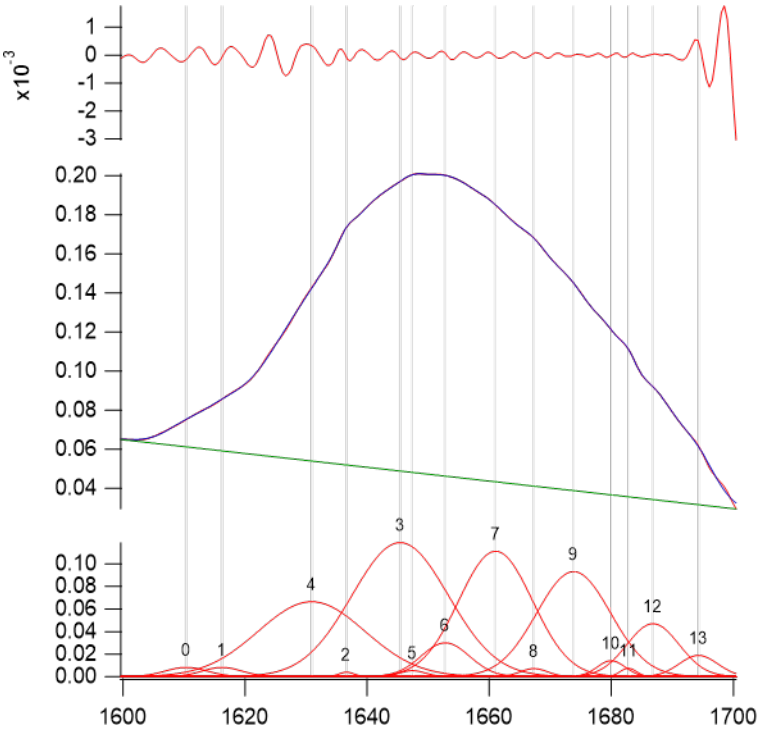
FIGURE S21. Deconvolution of FTIR spectra within the Amide I region (1600-1700 cm^{-1}) following 40 °C heating. (Top panels) Multiple linear regression is used to subtract the contribution of the SMP substrate (blue) from the total signal (SF + strained SMP) with no methanol treatment (orange) through decomposition. Three independent replicates were measured for each condition (n=3). (Bottom panels) Deconvolution results of FTIR spectra for SF-SMP with no methanol treatment. In the middle panel, raw data (red) and reproduced spectra (blue) are depicted, with the blue line representing the summation of the deconvoluted peaks. Residual fit errors are depicted in the top panels. The bottom panel shows the results from the deconvolution process; secondary structures are assigned based on peak locations within this panel. The normalized peak areas were used to determine the secondary structures found in the film.

Strained SF-SMP, No MeOH, 70 °C Trial 1

FTIR Background Subtraction

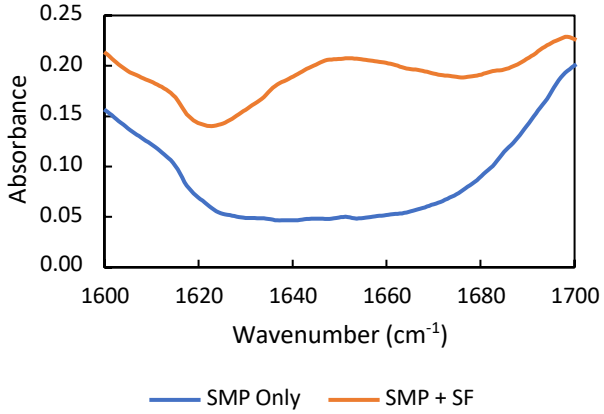


FTIR Deconvolution

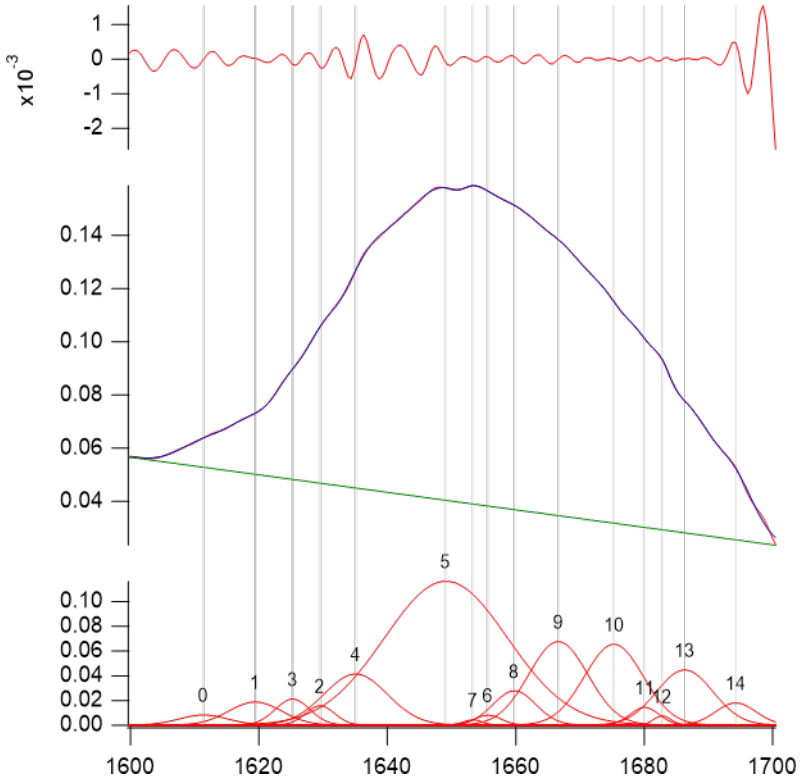


Strained SF-SMP, No MeOH, 70 °C Trial 63

FTIR Background Subtraction



FTIR Deconvolution



Strained SF-SMP, No MeOH, 70 °C Trial 64

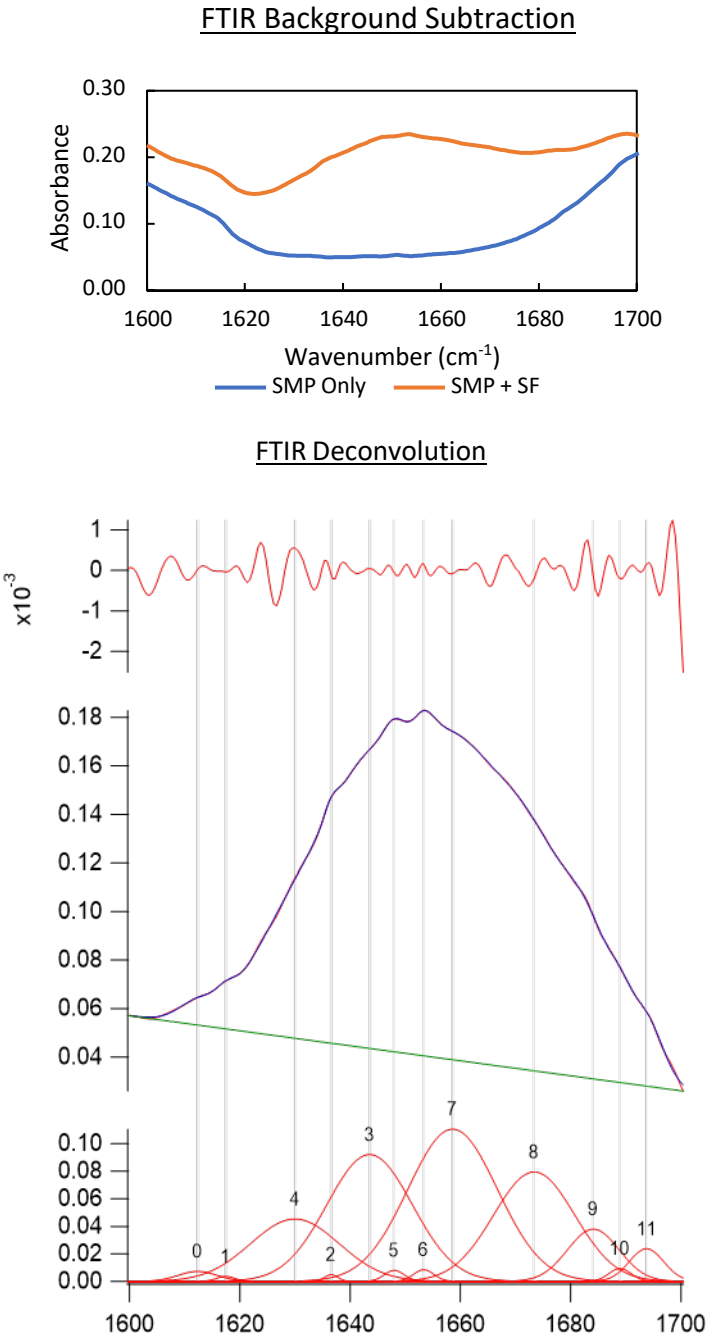
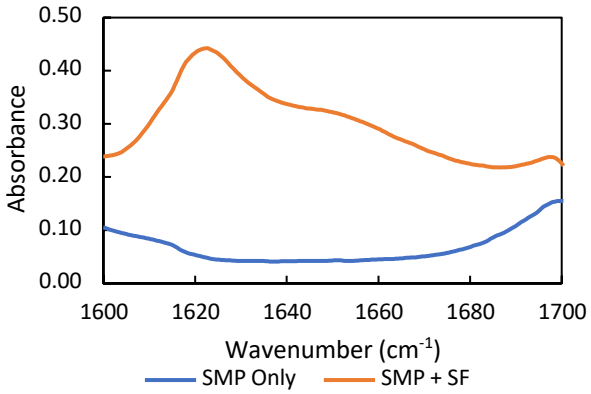


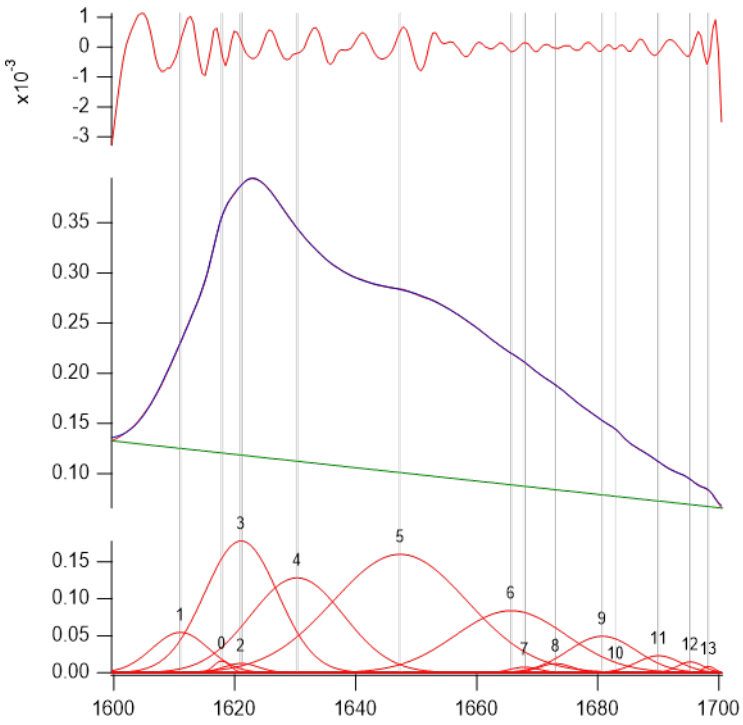
FIGURE S22. Deconvolution of FTIR spectra within the Amide I region (1600-1700 cm^{-1}) following 70 °C heating. (Top panels) Multiple linear regression is used to subtract the contribution of the SMP substrate (blue) from the total signal (SF + strained SMP) with no methanol treatment (orange) through decomposition. Three independent replicates were measured for each condition (n=3). (Bottom panels) Deconvolution results of FTIR spectra for SF-SMP with no methanol treatment. In the middle panel, raw data (red) and reproduced spectra (blue) are depicted, with the blue line representing the summation of the deconvoluted peaks. Residual fit errors are depicted in the top panels. The bottom panel shows the results from the deconvolution process; secondary structures are assigned based on peak locations within this panel. The normalized peak areas were used to determine the secondary structures found in the film.

Strained SF-SMP, With MeOH, 40 °C Trial 65

FTIR Background Subtraction

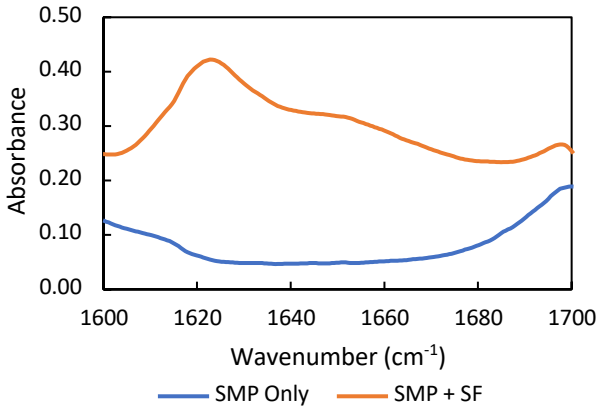


FTIR Deconvolution

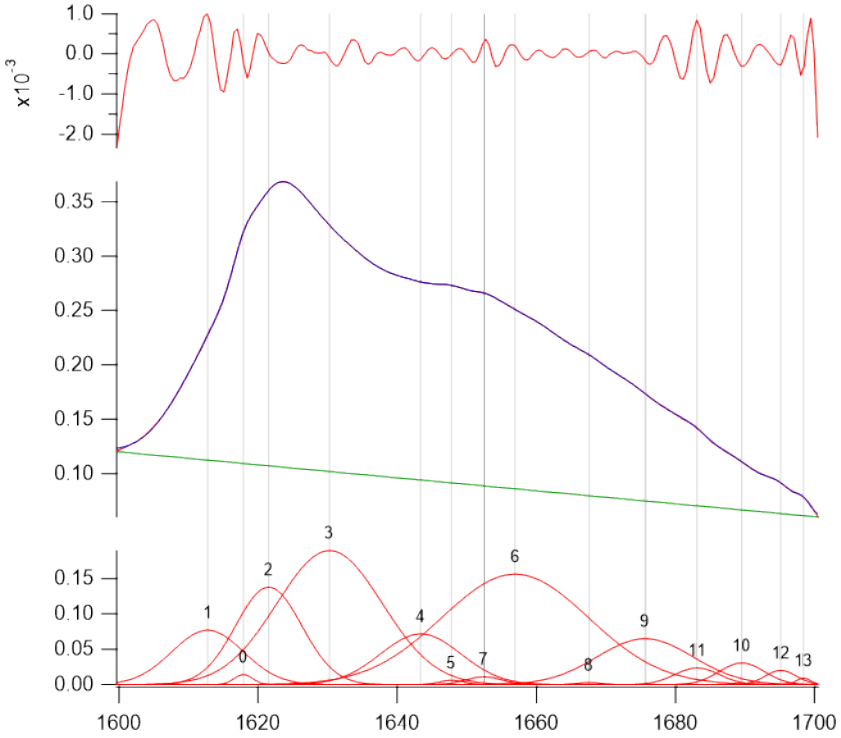


Strained SF-SMP, With MeOH, 40 °C Trial 66

FTIR Background Subtraction



FTIR Deconvolution



Strained SF-SMP, With MeOH, 40 °C Trial 67

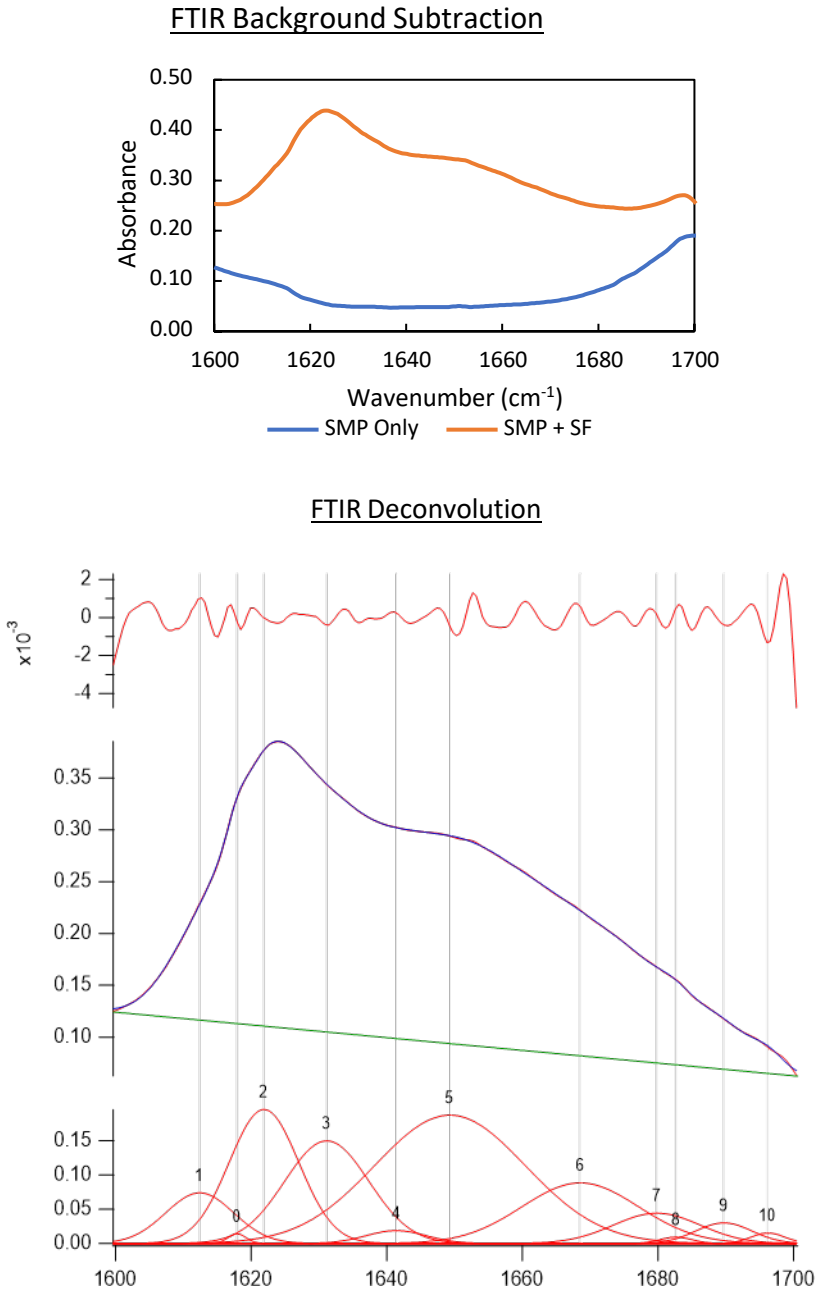
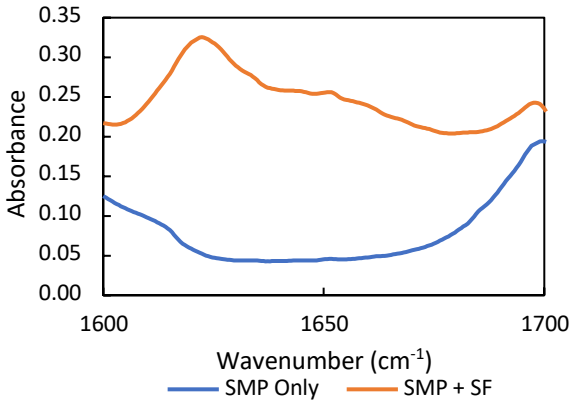


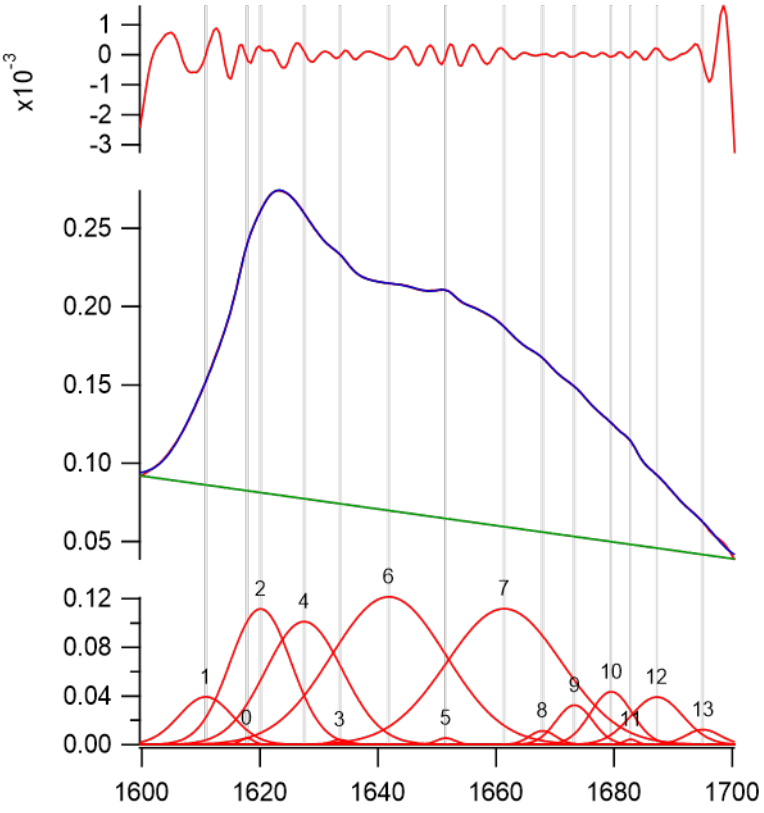
FIGURE S23. Deconvolution of FTIR spectra within the Amide I region (1600-1700 cm^{-1}) following 40 °C heating. (Top panels) Multiple linear regression is used to subtract the contribution of the SMP substrate (blue) from the total signal (SF + strained SMP) with methanol treatment (orange) through decomposition. Three independent replicates were measured for each condition (n=3). (Bottom panels) Deconvolution results of FTIR spectra for SF-SMP with methanol treatment. In the middle panel, raw data (red) and reproduced spectra (blue) are depicted, with the blue line representing the summation of the deconvoluted peaks. Residual fit errors are depicted in the top panels. The bottom panel shows the results from the deconvolution process; secondary structures are assigned based on peak locations within this panel. The normalized peak areas were used to determine the secondary structures found in the film.

Strained SF-SMP, With MeOH, 70 °C Trial 1

FTIR Background Subtraction

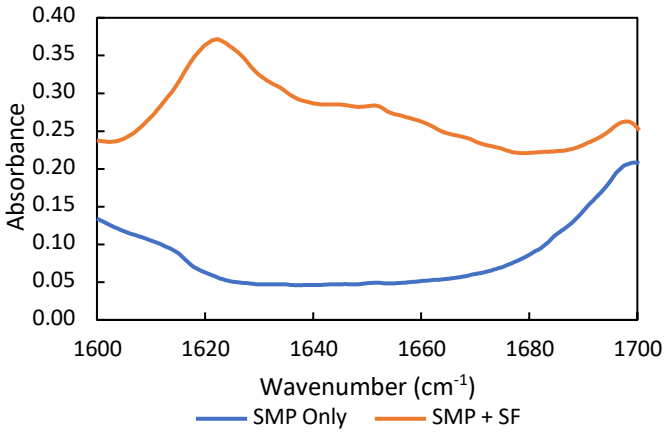


FTIR Deconvolution

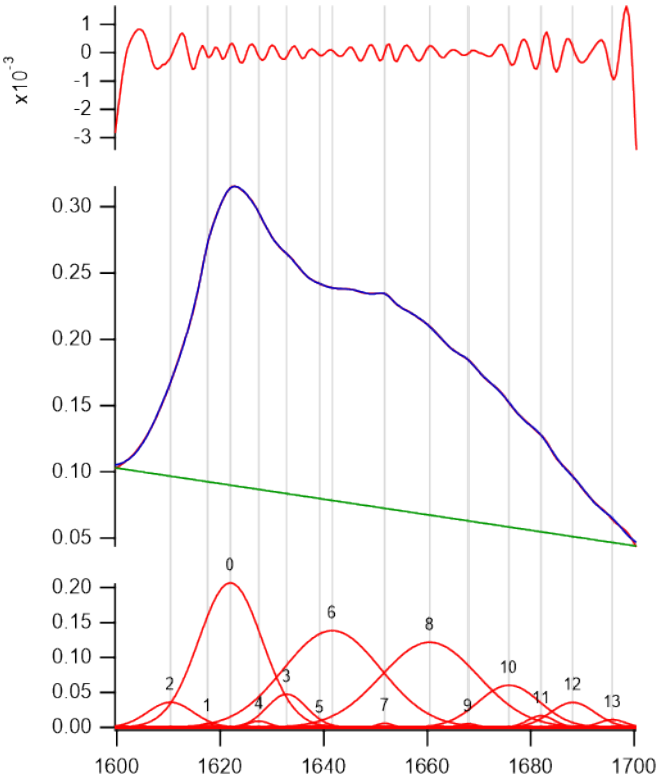


Strained SF-SMP, With MeOH, 70 °C Trial 69

FTIR Background Subtraction



FTIR Deconvolution



Strained SF-SMP, With MeOH, 70 °C Trial

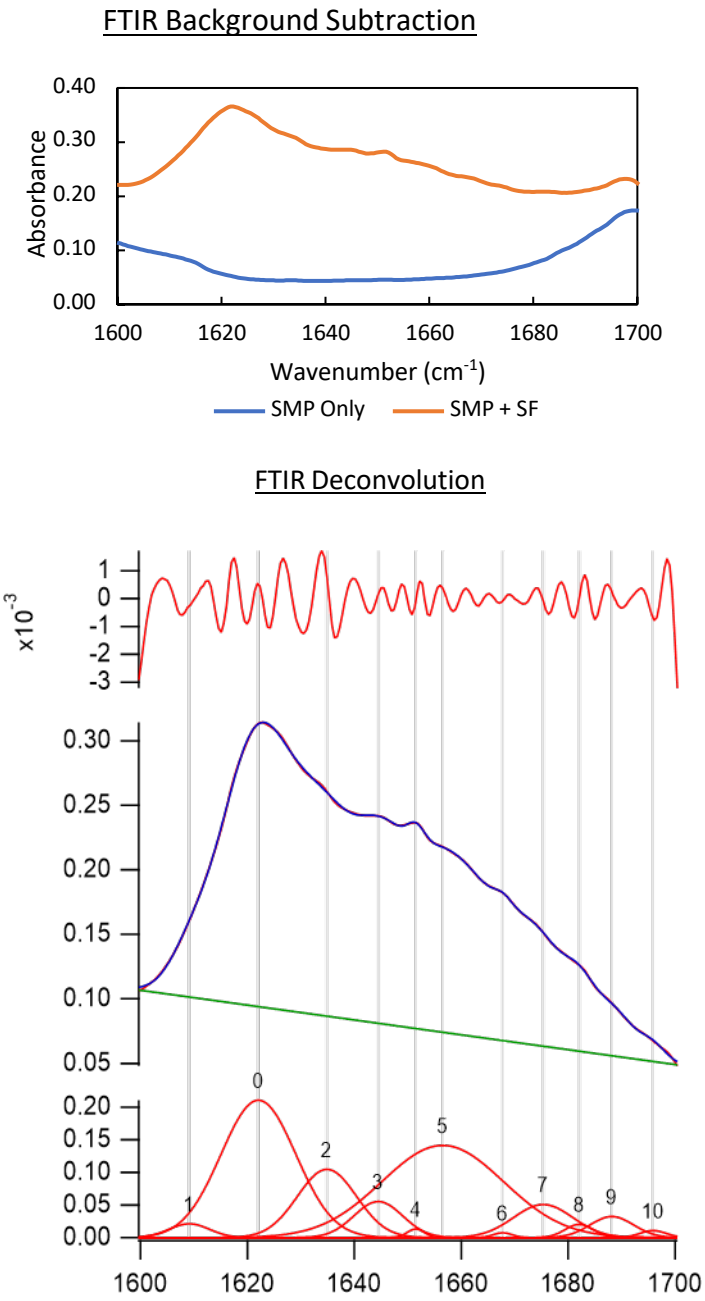
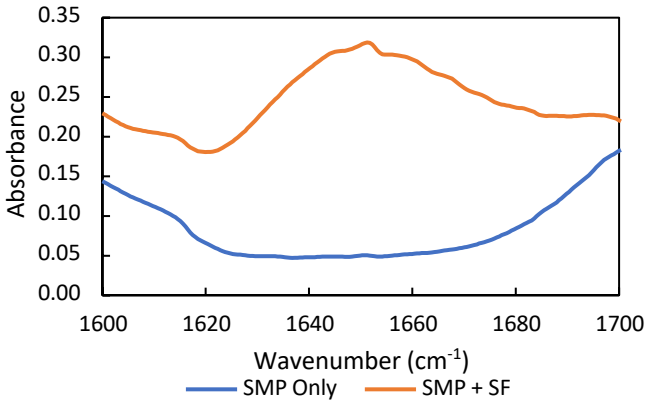


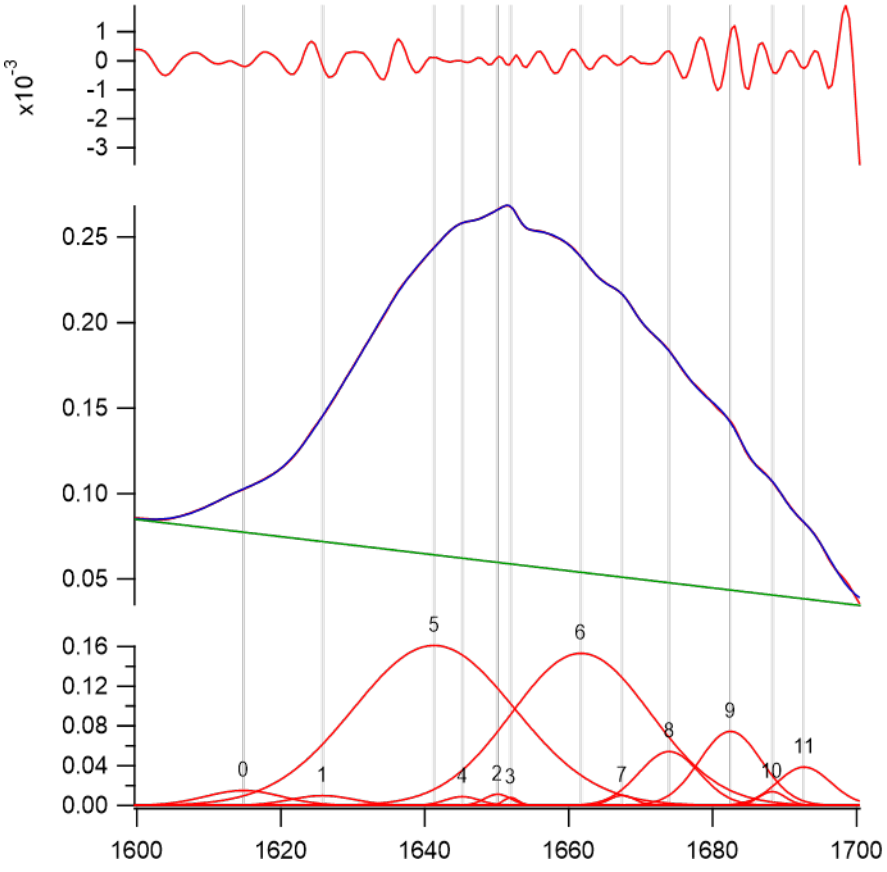
FIGURE S24. Deconvolution of FTIR spectra within the Amide I region (1600-1700 cm^{-1}) following 70 °C heating. (Top panels) Multiple linear regression is used to subtract the contribution of the SMP substrate (blue) from the total signal (SF + strained SMP) with methanol treatment (orange) through decomposition. Three independent replicates were measured for each condition (n=3). (Bottom panels) Deconvolution results of FTIR spectra for SF-SMP with methanol treatment. In the middle panel, raw data (red) and reproduced spectra (blue) are depicted, with the blue line representing the summation of the deconvoluted peaks. Residual fit errors are depicted in the top panels. The bottom panel shows the results from the deconvolution process; secondary structures are assigned based on peak locations within this panel. The normalized peak areas were used to determine the secondary structures found in the film.

Unstrained SF-SMP, No MeOH, 40 °C Trial 71

FTIR Background Subtraction

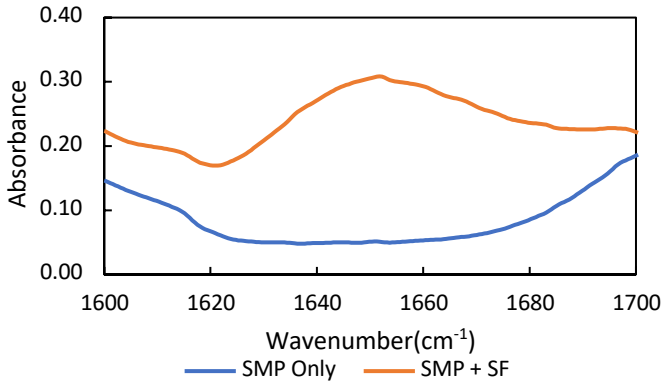


FTIR Deconvolution

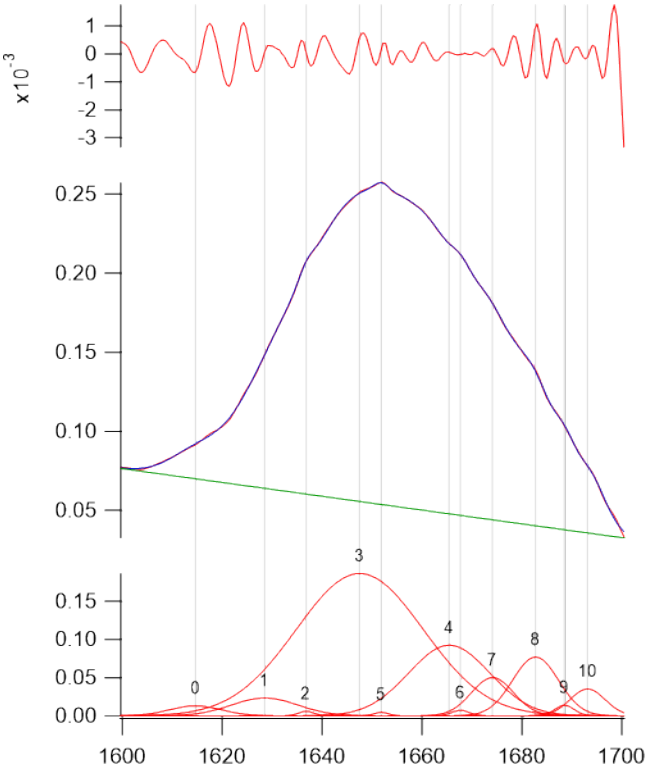


Unstrained SF-SMP, No MeOH, 40 °C Trial 72

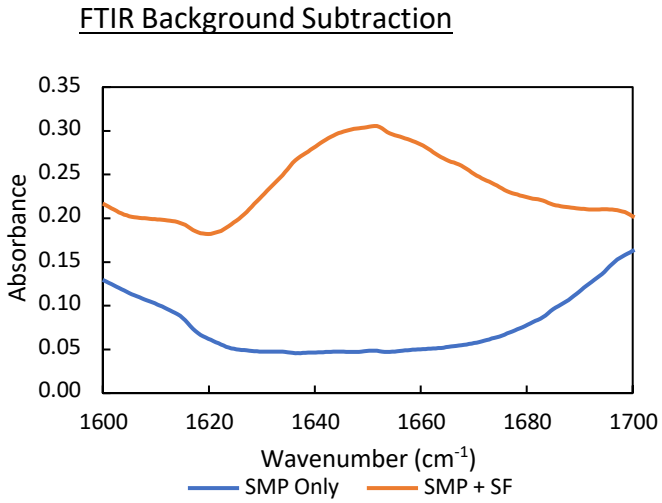
FTIR Background Subtraction



FTIR Deconvolution



Unstrained SF-SMP, No MeOH, 40 °C Trial



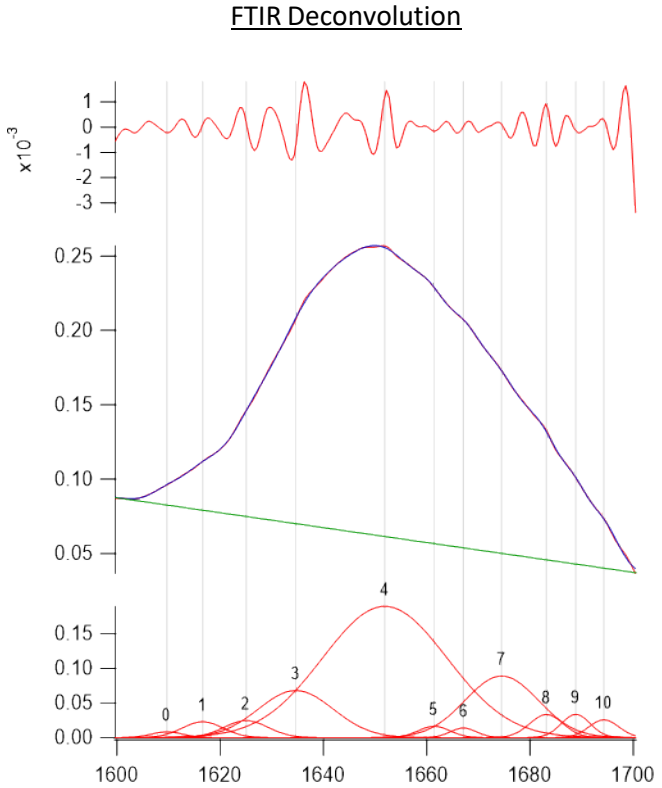
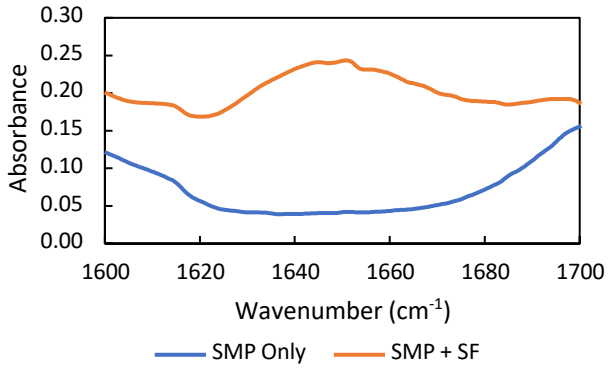


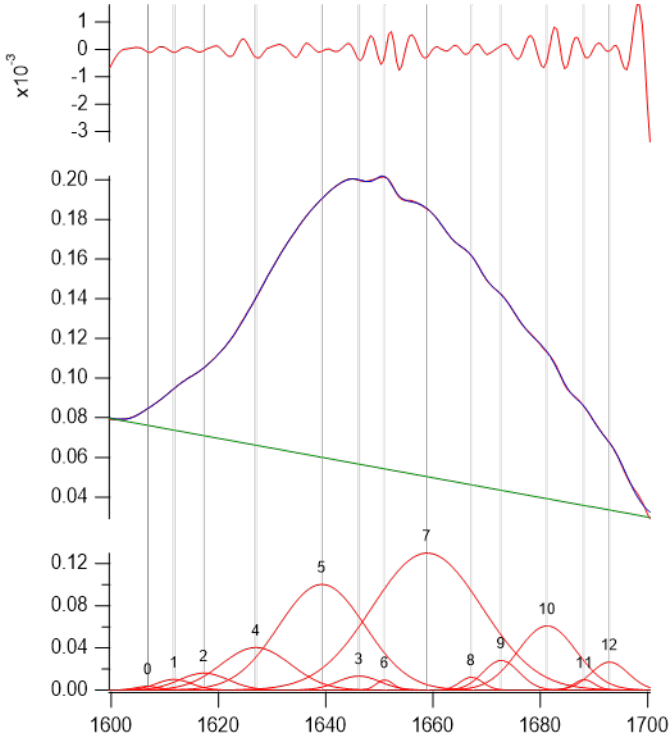
FIGURE S25. Deconvolution of FTIR spectra within the Amide I region (1600-1700 cm^{-1}) following 40 °C heating. (Top panels) Multiple linear regression is used to subtract the contribution of the SMP substrate (blue) from the total signal (SF + unstrained SMP) with no methanol treatment (orange) through decomposition. Three independent replicates were measured for each condition (n=3). (Bottom panels) Deconvolution results of FTIR spectra for SF-SMP with no methanol treatment. In the middle panel, raw data (red) and reproduced spectra (blue) are depicted, with the blue line representing the summation of the deconvoluted peaks. Residual fit errors are depicted in the top panels. The bottom panel shows the results from the deconvolution process; secondary structures are assigned based on peak locations within this panel. The normalized peak areas were used to determine the secondary structures found in the film.

Unstrained SF-SMP, No MeOH, 70 °C Trial

FTIR Background Subtraction

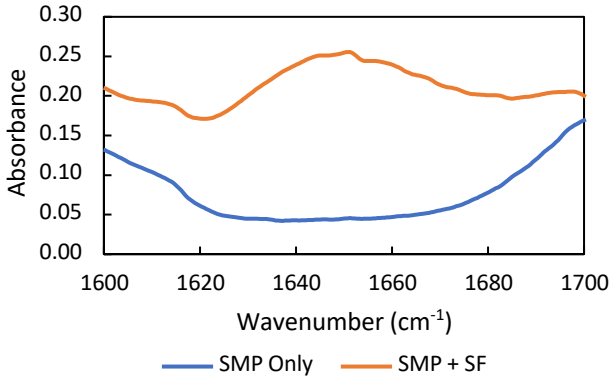


FTIR Deconvolution

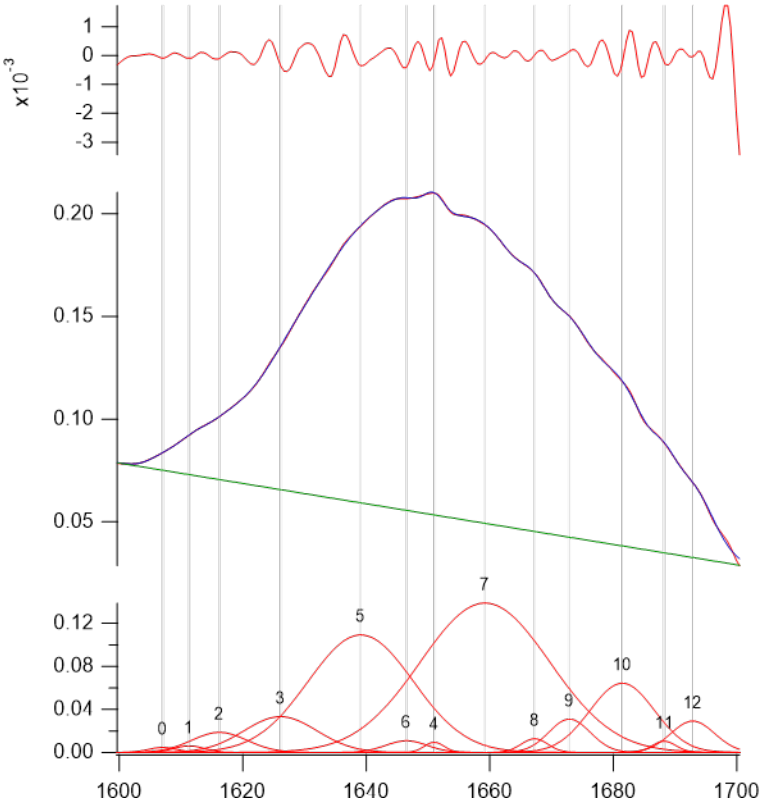


Unstrained SF-SMP, No MeOH, 70 °C Trial

FTIR Background Subtraction



FTIR Deconvolution



Unstrained SF-SMP, No MeOH, 70 °C Trial

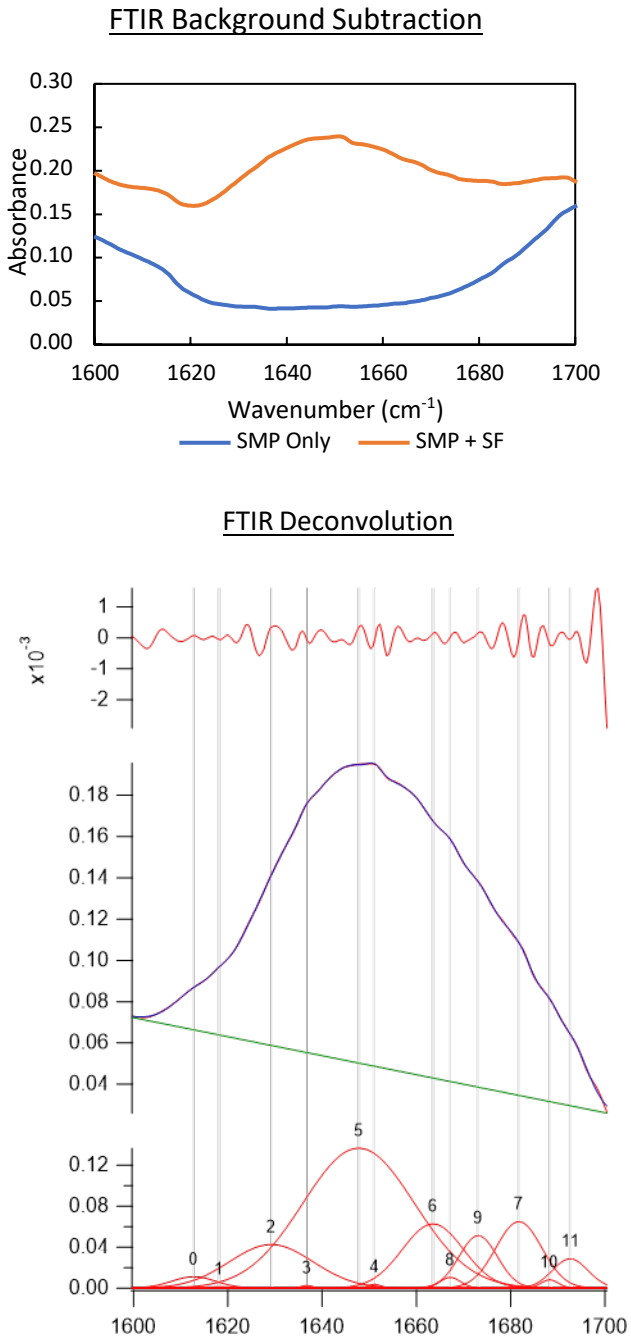


FIGURE S26. Deconvolution of FTIR spectra within the Amide I region (1600-1700 cm^{-1}) following 70 °C heating. (Top panels) Multiple linear regression is used to subtract the contribution of the SMP substrate (blue) from the total signal (SF + unstrained SMP) with no methanol treatment (orange) through decomposition. Three independent replicates were measured for each condition (n=3). (Bottom panels) Deconvolution results of FTIR spectra for SF-SMP with no methanol treatment. In the middle panel, raw data (red) and reproduced spectra (blue) are depicted, with the blue line representing the summation of the deconvoluted peaks. Residual fit errors are depicted in the top panels. The bottom panel shows the results from the deconvolution process; secondary structures are assigned based on peak locations within this panel. The normalized peak areas were used to determine the secondary structures found in the film.

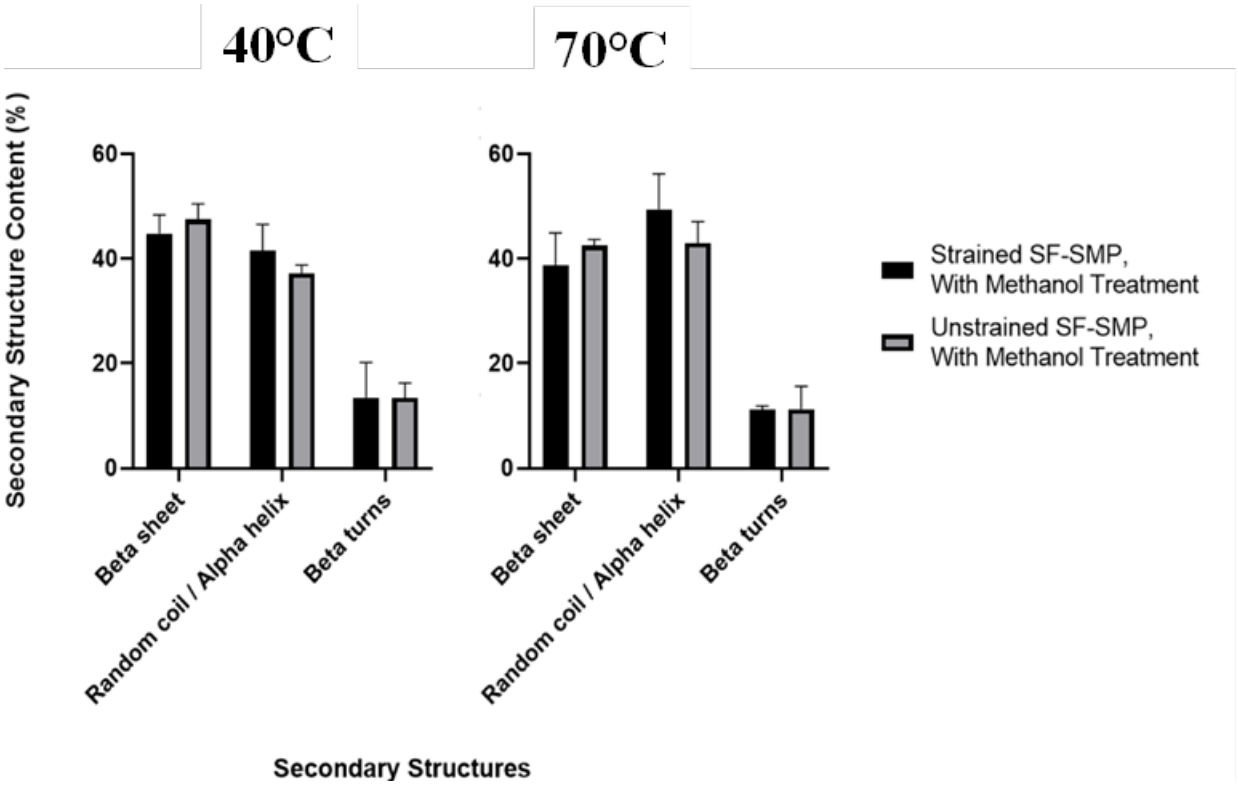
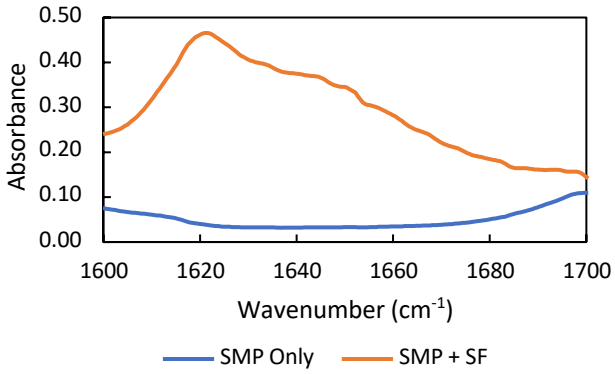


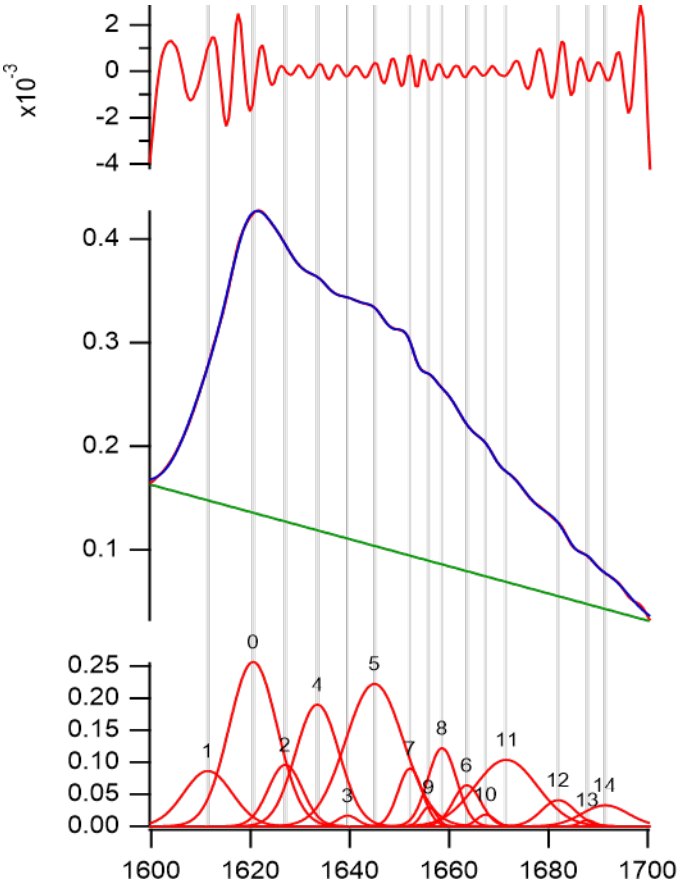
FIGURE S27. No significant differences were observed for secondary structural changes in the silk network between the strained and unstrained SF-SMP with exposure to methanol treatment following 40 °C and 70 °C heating.

Unstrained SF-SMP, With MeOH, 40 °C Trial

FTIR Background Subtraction

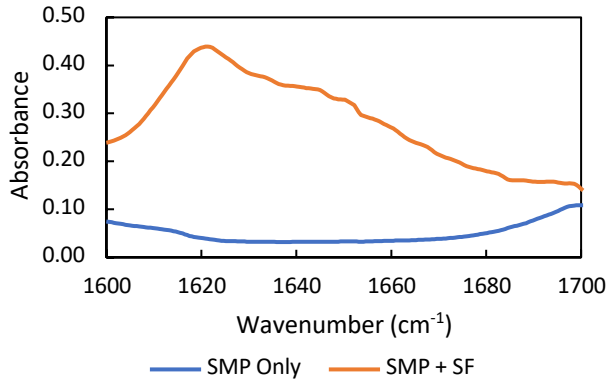


FTIR Deconvolution

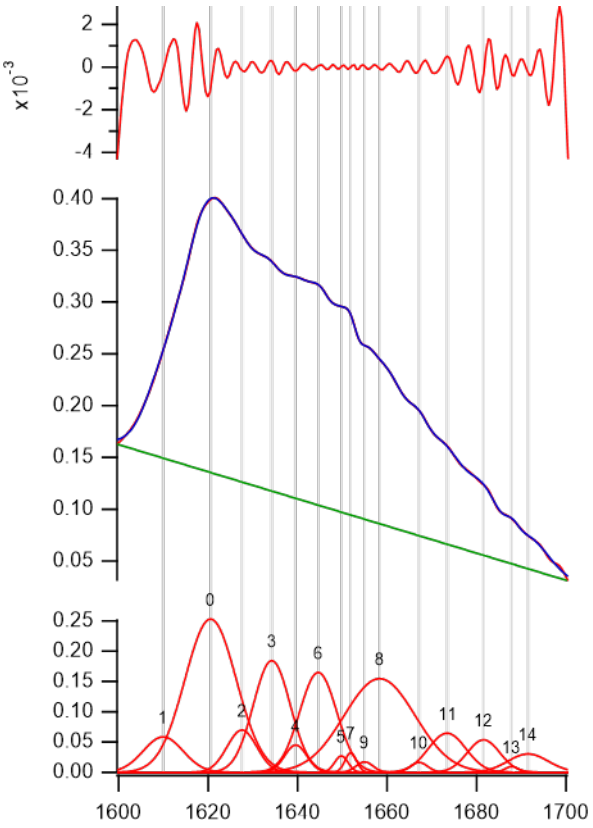


Unstrained SF-SMP, With MeOH, 40 °C Trial

FTIR Background Subtraction



FTIR Deconvolution



Unstrained SF-SMP, With MeOH, 40 °C Trial 3

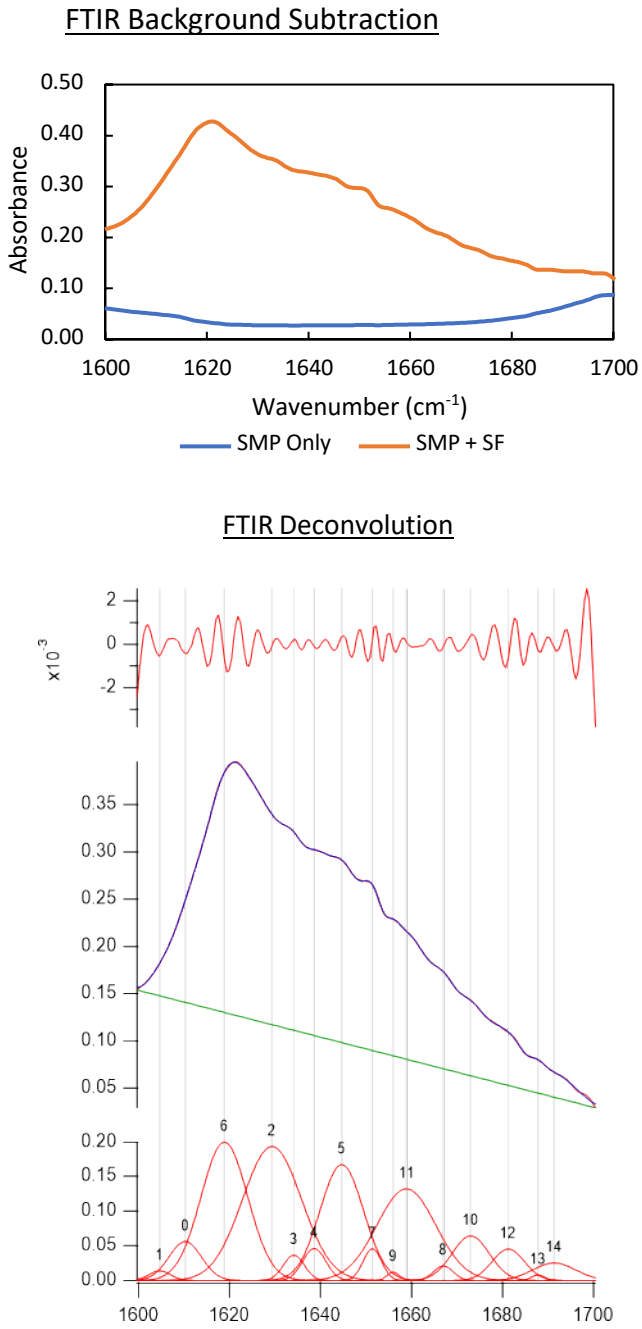
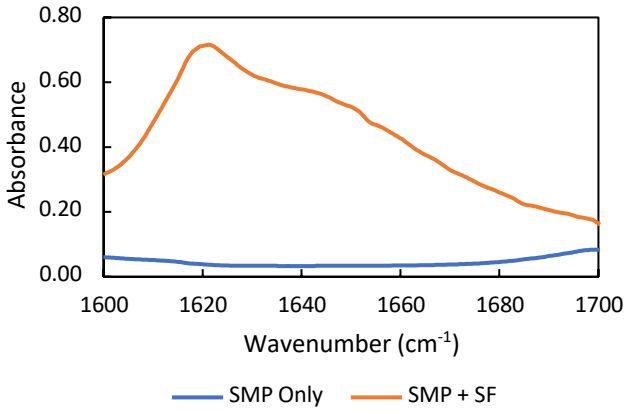


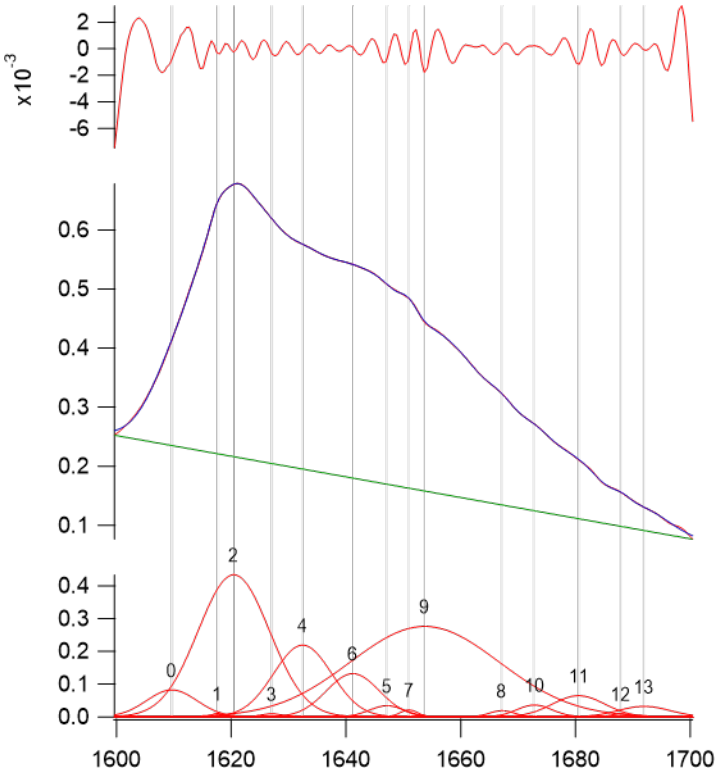
FIGURE S28. Deconvolution of FTIR spectra within the Amide I region (1600-1700 cm^{-1}) following 40 °C heating. (Top panels) Multiple linear regression is used to subtract the contribution of the SMP substrate (blue) from the total signal (SF + unstrained SMP) with methanol treatment (orange) through decomposition. Three independent replicates were measured for each condition (n=3). (Bottom panels) Deconvolution results of FTIR spectra for SF-SMP with methanol treatment. In the middle panel, raw data (red) and reproduced spectra (blue) are depicted, with the blue line representing the summation of the deconvoluted peaks. Residual fit errors are depicted in the top panels. The bottom panel shows the results from the deconvolution process; secondary structures are assigned based on peak locations within this panel. The normalized peak areas were used to determine the secondary structures found in the film.

Unstrained SF-SMP, With MeOH, 70 °C Trial

FTIR Background Subtraction

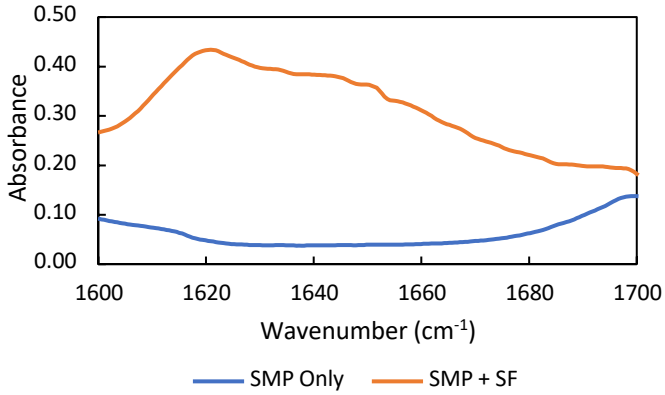


FTIR Deconvolution

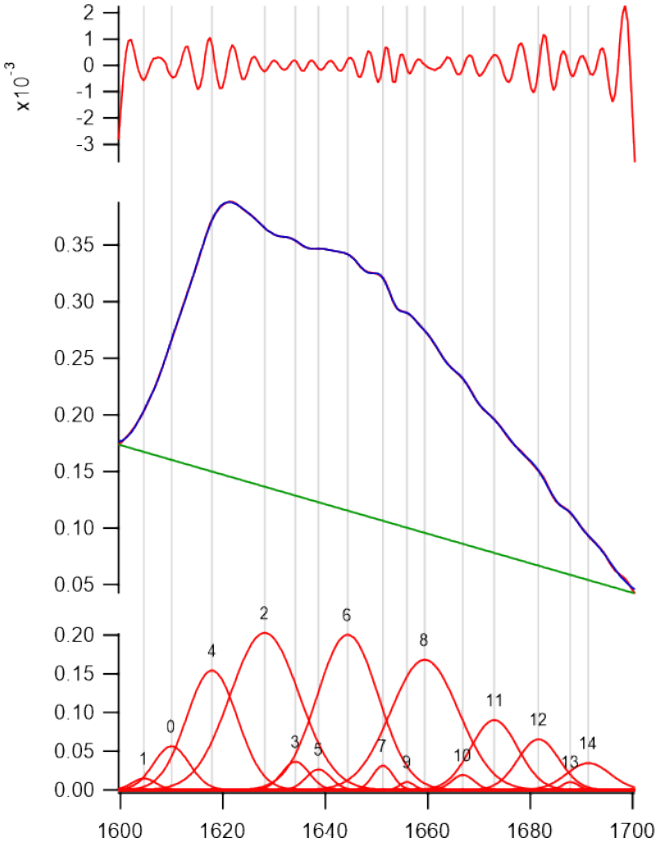


Unstrained SF-SMP, With MeOH, 70 °C Trial

FTIR Background Subtraction



FTIR Deconvolution



Unstrained SF-SMP, With MeOH, 70 °C Trial

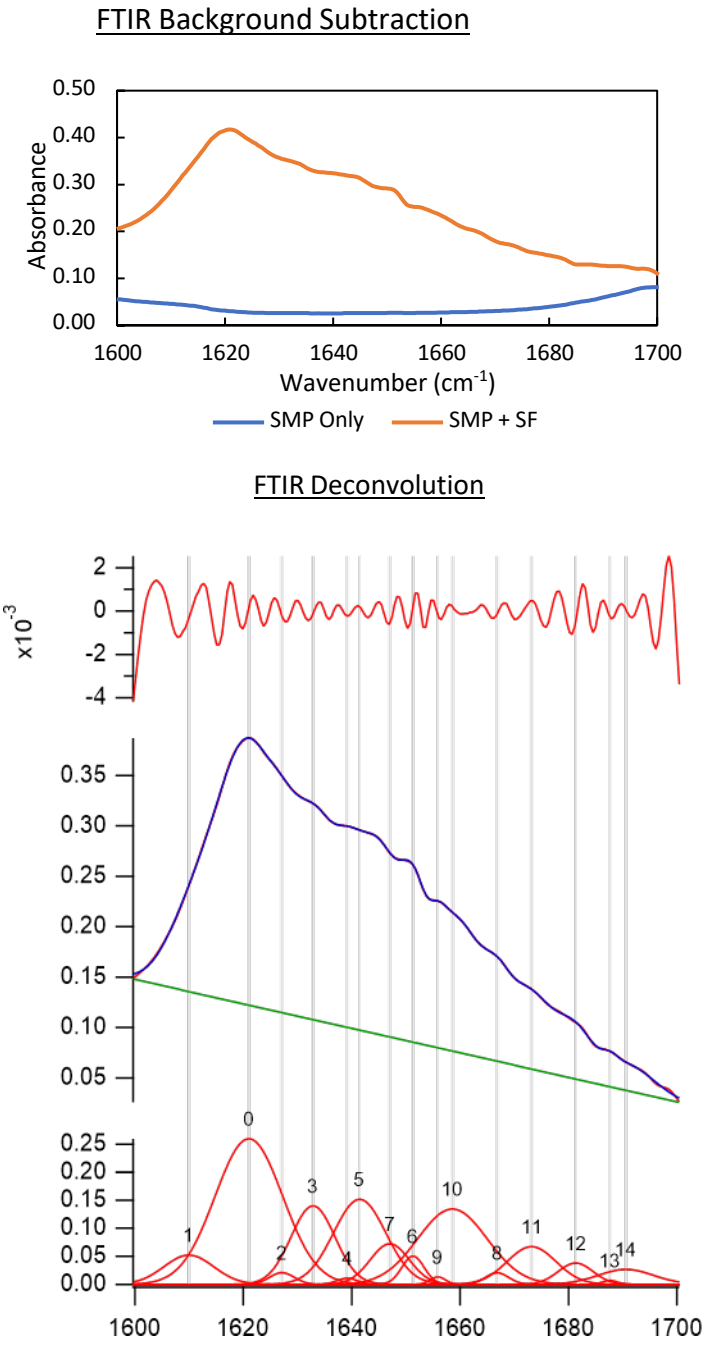


FIGURE S29. Deconvolution of FTIR spectra within the Amide I region (1600-1700 cm^{-1}) following 70 °C heating. (Top panels) Multiple linear regression is used to subtract the contribution of the SMP substrate (blue) from the total signal (SF + unstrained SMP) with methanol treatment (orange) through decomposition. Three independent replicates were measured for each condition (n=3). (Bottom panels) Deconvolution results of FTIR spectra for SF-SMP with methanol treatment. In the middle panel, raw data (red) and reproduced spectra (blue) are depicted, with the blue line representing the summation of the deconvoluted peaks. Residual fit errors are depicted in the top panels. The bottom panel shows the results from the deconvolution process; secondary structures are assigned based on peak locations within this panel. The normalized peak areas were used to determine the secondary structures found in the film.