"Remediation of Perfluorooctane Sulfonic Acid (PFOS) using Supported Lipid Bilayers on Mesoporous-SiO_2"

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Supplementary Information

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Preparation of liposomes

Multilamellar and unilamellar liposomes were made by dissolution of neat DPPC or mixtures of DPPC and PFOS in methanol/chloroform ($MeOH/CHCl_3$ = 1/2 v/v). Lipid concentrations were kept at ~ 1 mg/mL unless otherwise noted. The multilamellar liposomes were prepared by evaporation of the $MeOH/CHCl_3$ and hydrated with 0.1M NaCl. The hydrated liposomes were heated above their order-to-disorder temperature, T_m, and immersed in dry ice/acetone for ~ six freeze-thaw cycles. Unilamellar liposomes were prepared by extrusion of the multilamellar liposomes with 100 nm polycarbonate filters above T_m. Ratios of DPPC/PFOS = 75/1, 50/1, 25/1, 15/1, 10/1, 5/1, 2.5/1, 1/1 and 0.5 were prepared. For incubated multilamellar liposomes/unilamellar liposomes, the DPPC/PFOS ratio indicates the molar ratio of DPPC to PFOS in the solutions, while for the prepared multilamellar liposomes/unilamellar liposomes it is the ratio incorporated into the lipid films before hydration. For the incubation studies, the PFOS concentrations used were well below the critical micelle concentration (CMC)[22], reported as ~ 5.5–7.4 mM for LiPFOS[23] and 0.02 mol/kg for NaPFOS[24] at 25 ^{o}C.

Dynamic Light Scattering (DLS) Data

DLS data for neat unilamellar liposomes and unilamellar liposomes incubated with PFOS in DPPC/PFOS ratios of 25/1, 1/1 and 0.5/1, as a function of time for 90 minutes show that liposomes remain intact.

A table (**Table S1**) of liposome diameters from DLS data indicates that even the 1/1 remains stable for 3 days. After heating above T_m and cooling, some liposome fusion does occur.

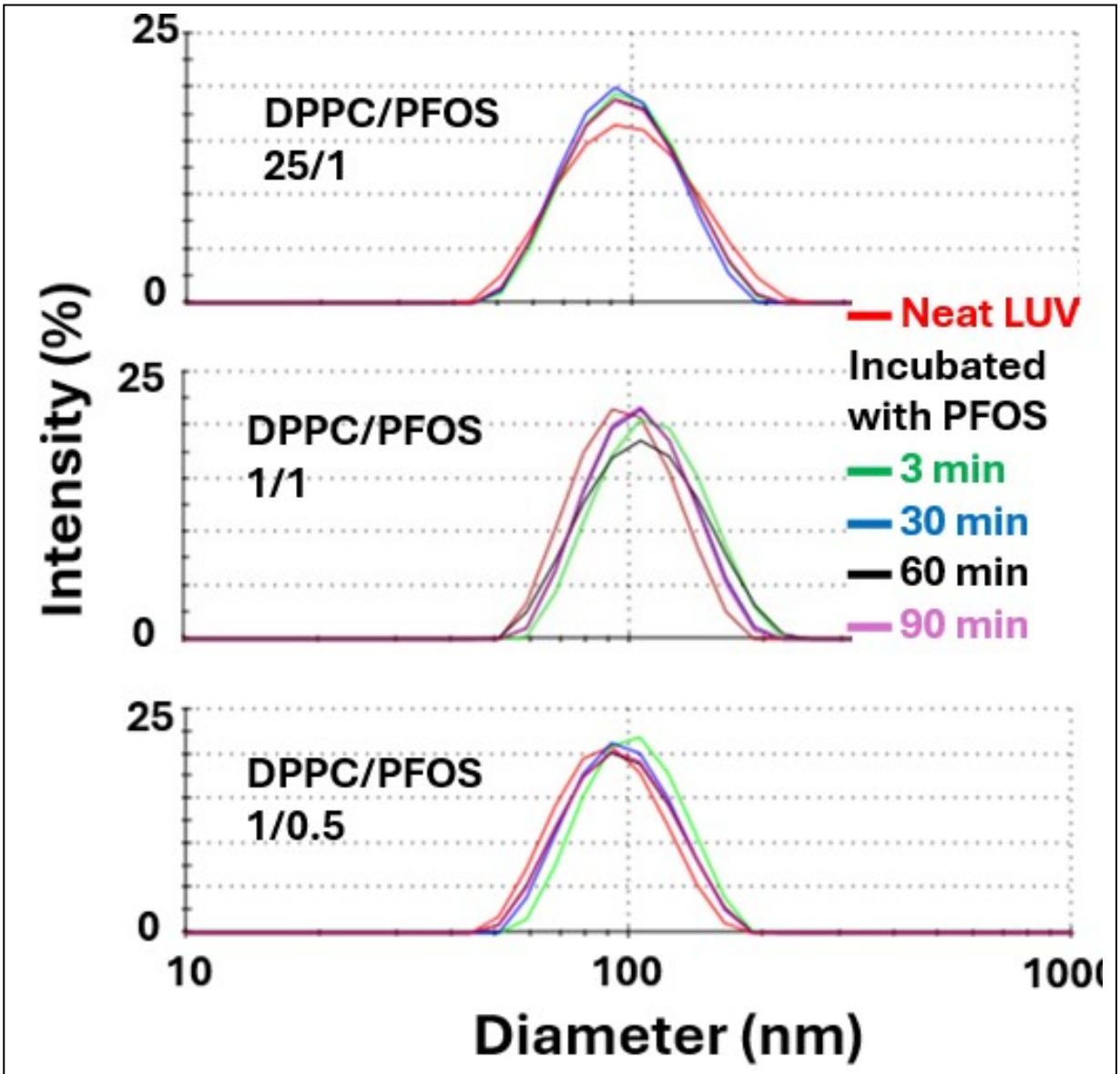


Figure S1. DLS data for neat unilamellar liposomes and unilamellar liposomes incubated with PFOS in DPPC/PFOS ratios of 25/1, 1/1 and 0.5/1, as a function of time for 90 minutes.

Table S1. Dynamic Light Scattering (DLS) data for neat LUVs and LUVs incubated with PFOS				
DPPC/PFOS	Temperature (^{0}C)	Time	Size (Diameter) (nm)	Polydispersity Index (PDI)
100/0	25	0 min	110.8	0.139
	50	15 min	114.8	0.060
	70	1 h	121.0	0.058
	25	10 min	106.4	0.093
7.5/1	25	0 min (no PFOS)	97.14	0.059
	25	3 min	103.6	0.042
	25	1.5 h	103.9	0.046
	50	15 min	116.8	0.056
2.5/1	25	0 min (no PFOS)	103.2	0.037
	25	3 min	108.8	0.022
	50	1 h	116.4	0.058
	25	10 min	106.1	0.123
1/1	25	0 min (no PFOS)	104.9	0.058
	25	3 min	113.4	0.060
	25	3 days	113.8	0.035
	50	1 h	169.4	0.237
	25	10 min	186.6	0.197

Effects of temperature, pH, ionic strength and natural organic matter

Another batch of Syloid®3150 mesoporous SiO_2 was prepared and characterized by TGA for the amount of lipid in the SLBs (**Figure S3**). The same vials containing the SLBs and sorbent solutions were used for both the TGA and ^{19}F NMR experiments. The actual amounts of SLBs and solutions in the vials were adjusted so that there was always 1.02 mg/mL for 10 mg SLBs. Then 750 µL of the supernatant (containing 0.765 mg PFOS) was removed and 65 µL of D_2O was added. Thus, what we measured by ^{19}F NMR was the amount remaining in the supernatant from a solution that originally contained 0.765 mg PFOS. The SLBs were then analyzed by TGA.

The addition of HA to either pristine mesoporous SiO_2 or SLBs on mesoporous SiO_2 did not result in further weight gain (**Figure S4**).

Stability of the SLBs

The removal of the lipids from the SLBs was only possible at the initial stage of lipid addition, when the lipids had been added using MeOH/$CHCl_3$. After the hydration process with 100 mM NaCl + 20 mM $CaCl_2$, it was not possible to remove the SLBs using boiling water for 1 h (**Figure S5**).

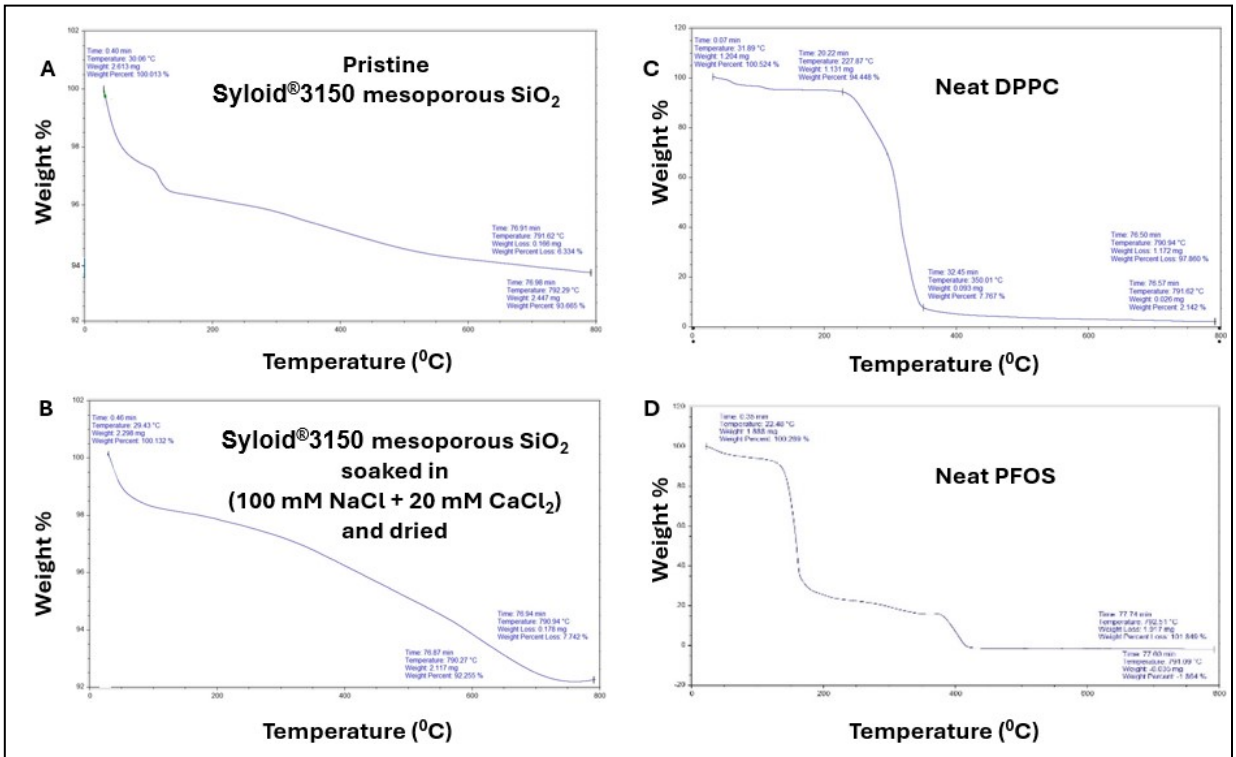


Figure S2. TGA data of: (**A**) prisitine Syloid™3150 mesoporous SiO_2; (**B**) Syloid™3150 mesoporous SiO_2 beads soaked in (100 mM NaCl + 20 mM $CaCl_2$) and dried; (C) neat DPPC; and (**D**) neat PFOS.

TGA Data

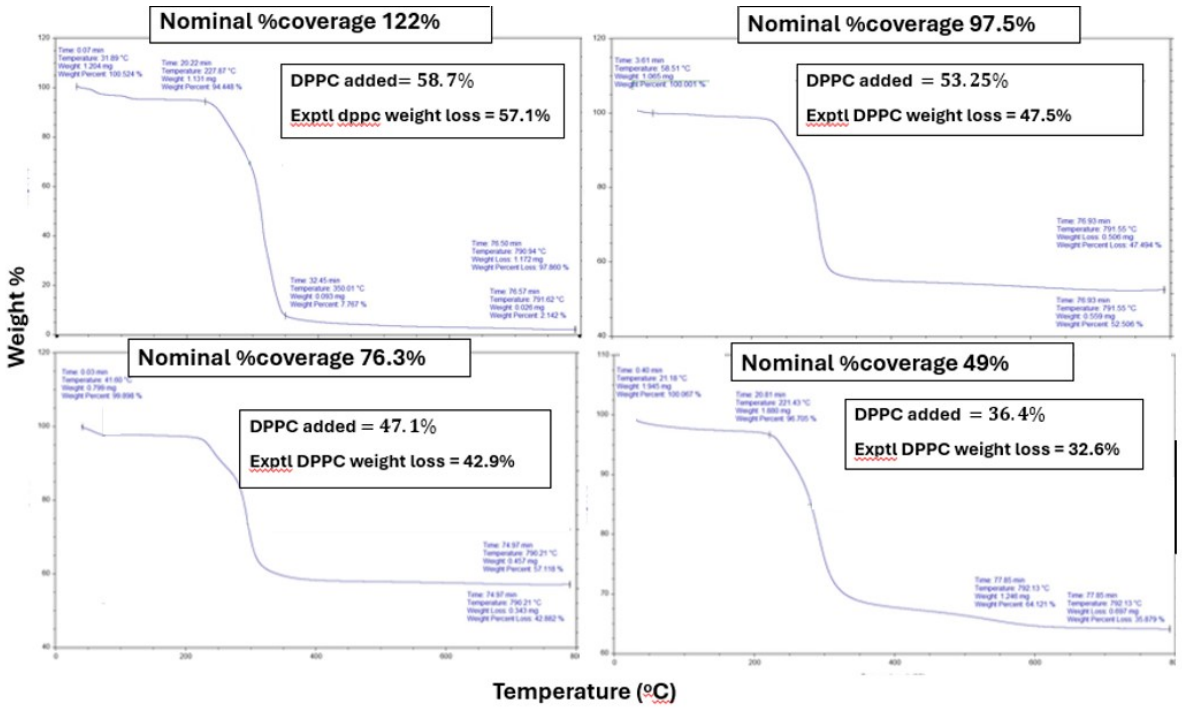


Figure S3. TGA and DTGA data of Syloid®3150 mesoporous SiO_2 after soaking in DPPC/($CHCl_3$/MeOH) and evaporation of the ($CHCl_3$/MeOH) so that there would be nominal 122%, 97.5%, 76.3% and 49% coverage. Comparison with experimental TGA coverage.

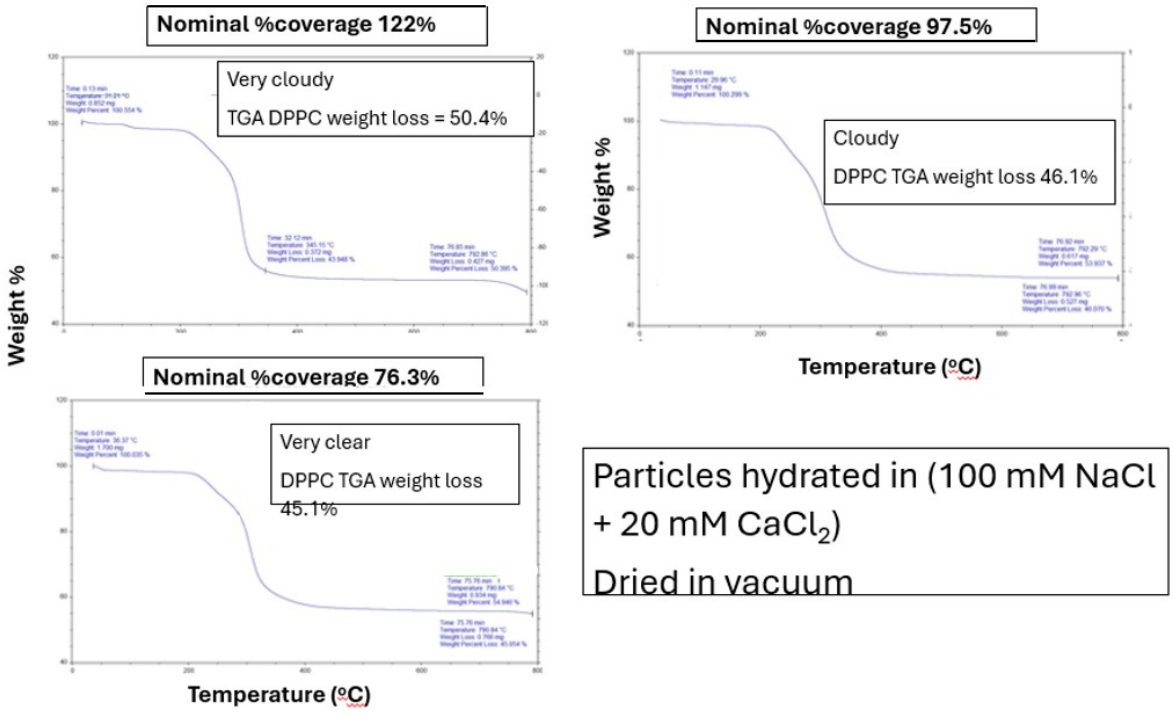
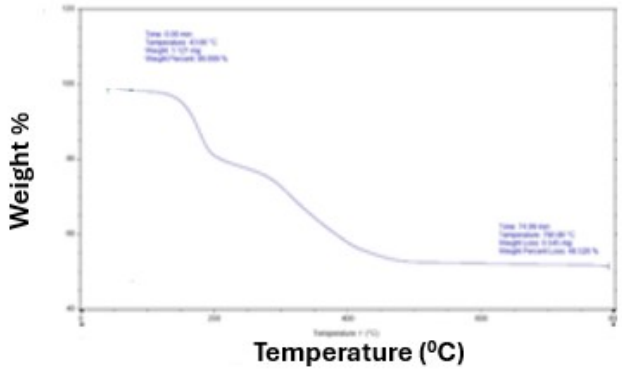
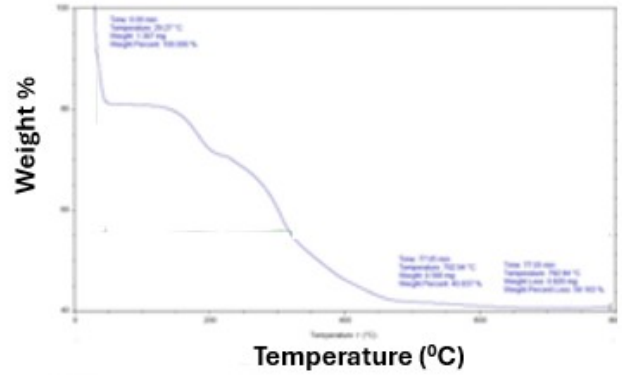


Figure S4. TGA data of Syloid®3150 mesoporous SiO_2 after removal of ($CHCl_3$/MeOH), soaking in aqueous (100 mM NaCl and 20 mM $CaCl_2$) solutions, and undergoing 5-10 freeze/thaw cycles. The mesoporous SiO_2 was allowed to settle, and the supernatant removed. The remaining liquid was dried and TGA obtained.



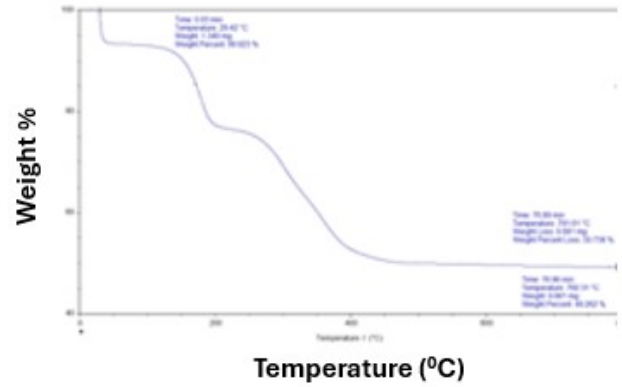
Nominal 122% DPPC coverage
In new vial
SLB= 11.9 mg
PFOS= 2.04 mg (1.2 ml)
Shake with PFOS 1h, centrifuge, remove supernatant and extra solution before TGA.

48.5% DPPC/PFOS



Nominal 97.5% DPPC coverage
In new vial
SLB= 9.3 mg
PFOS= 2.04 mg (1 ml)
Shake with PFOS for 1 h, centrifuge, remove supernatant and extra water before TGA.

59.1% DPPC/PFOS



Nominal 76% DPPC coverage
In new vial
SLB= 7.5 mg
PFOS= 2.04 mg (1 ml)
Shake with PFOS 1h, centrifuge, remove supernatant and extra water before TGA; weight loss

50.7% DPPC/PFOS

Figure S5. TGA data of Syloid®3150 mesoporous SiO_2 from dried aqueous Syloid®3150 mesoporous-SiO_2, incubated with PFOS and particles dried.

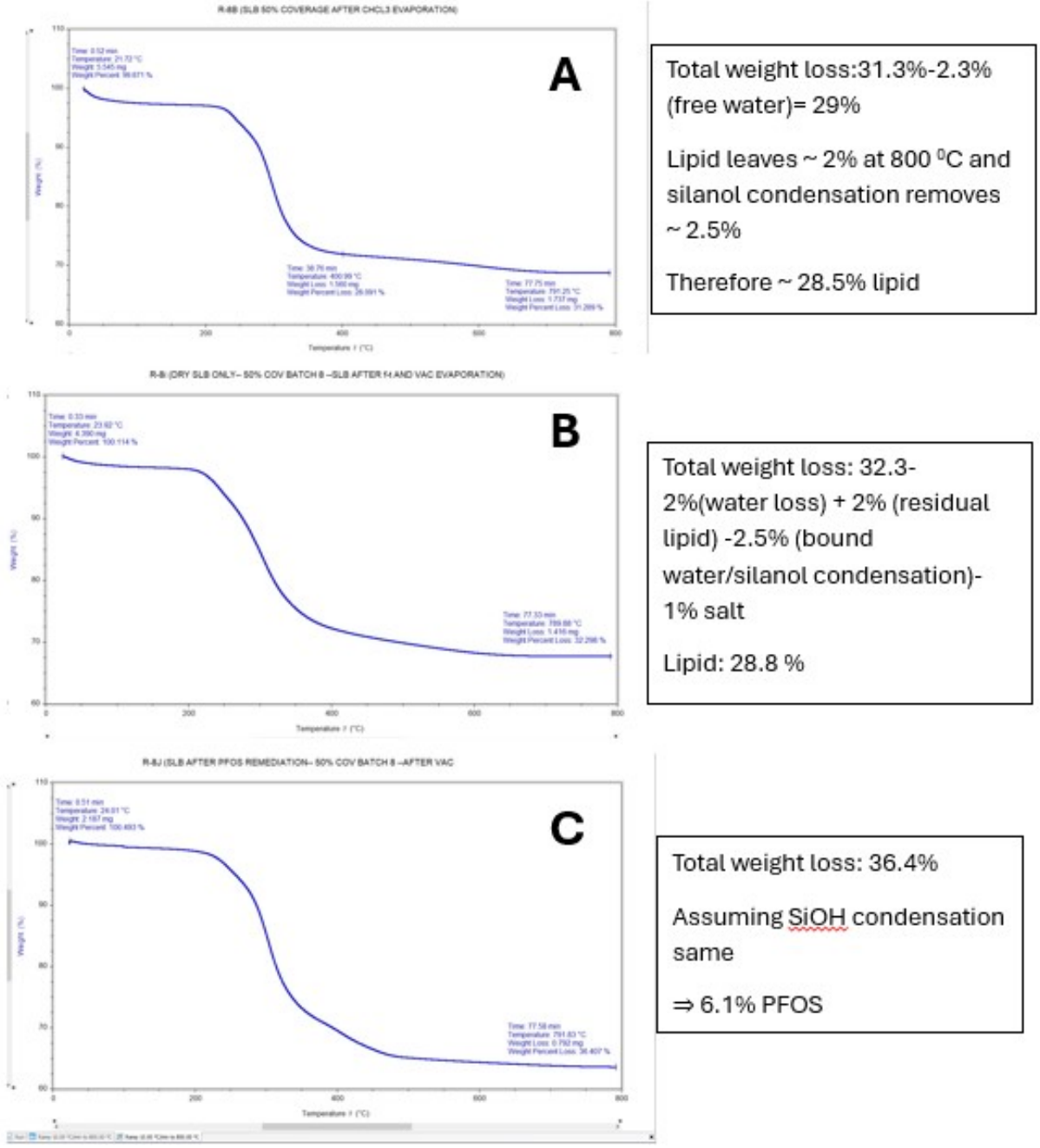


Figure S6. TGA of Syloid®3150 mesoporous SiO_2: (**A**) after addition of DPPC lipid in $CHCl_3$ and evaporation of $CHCl_3$; (**B**) after hydration of the DPPC lipid with 100 mM NaCl + 20 mM $CaCl_2$ to form SLBs and evaporation of water; (**C**) after exposure to PFOS in 0.1 mM NaCl and removal of water.

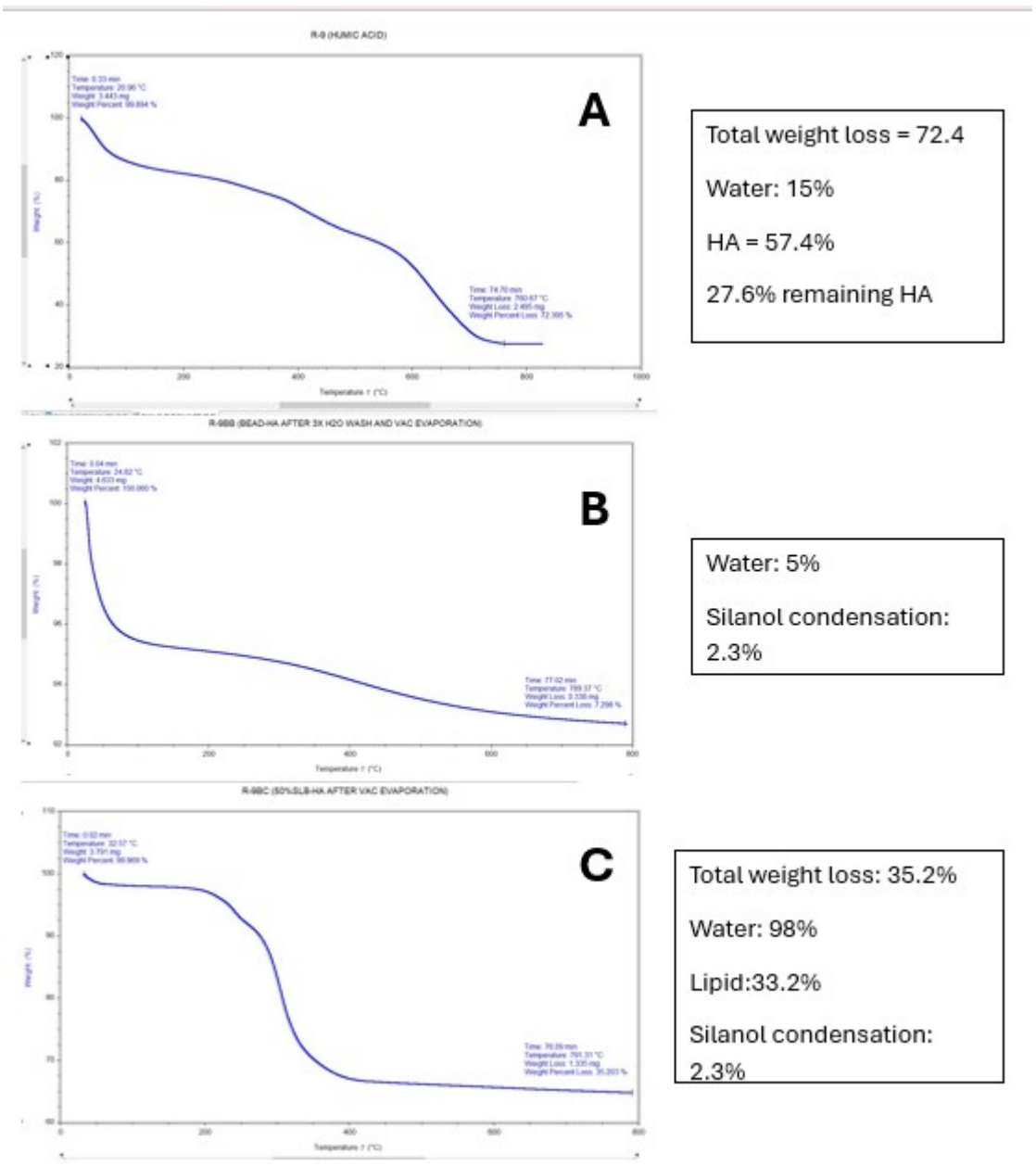


Figure S7. TGA data of: (**A**) pristine humic acid; (**B**) Syloid®3150 mesoporous SiO_2 after exposure to humic acid, rinsed 3x with water and vacuum dried; and (**C**) SLBs with 33% lipid after exposure (15 min) to HA.

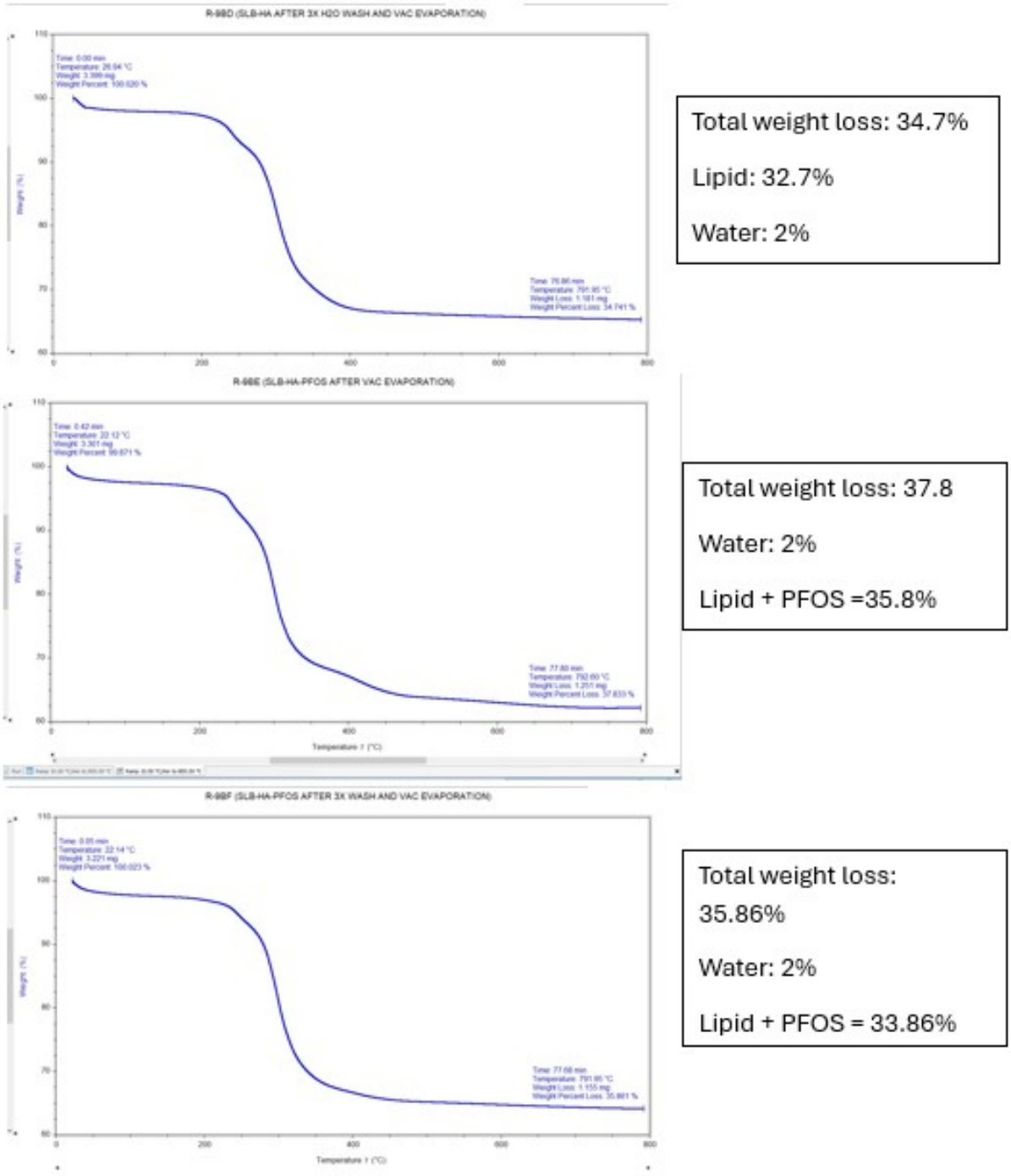


Figure S8. TGA data of (**A**) SLBs exposed to HA solution after rinsing 3x with water and vacuum drying; (**B**) SLBs with PFOS exposed to HA solution after vacuum removal of water; (**C**) same as (**Figure S9B** after rinsing 3x with water and vacuum removal of water.

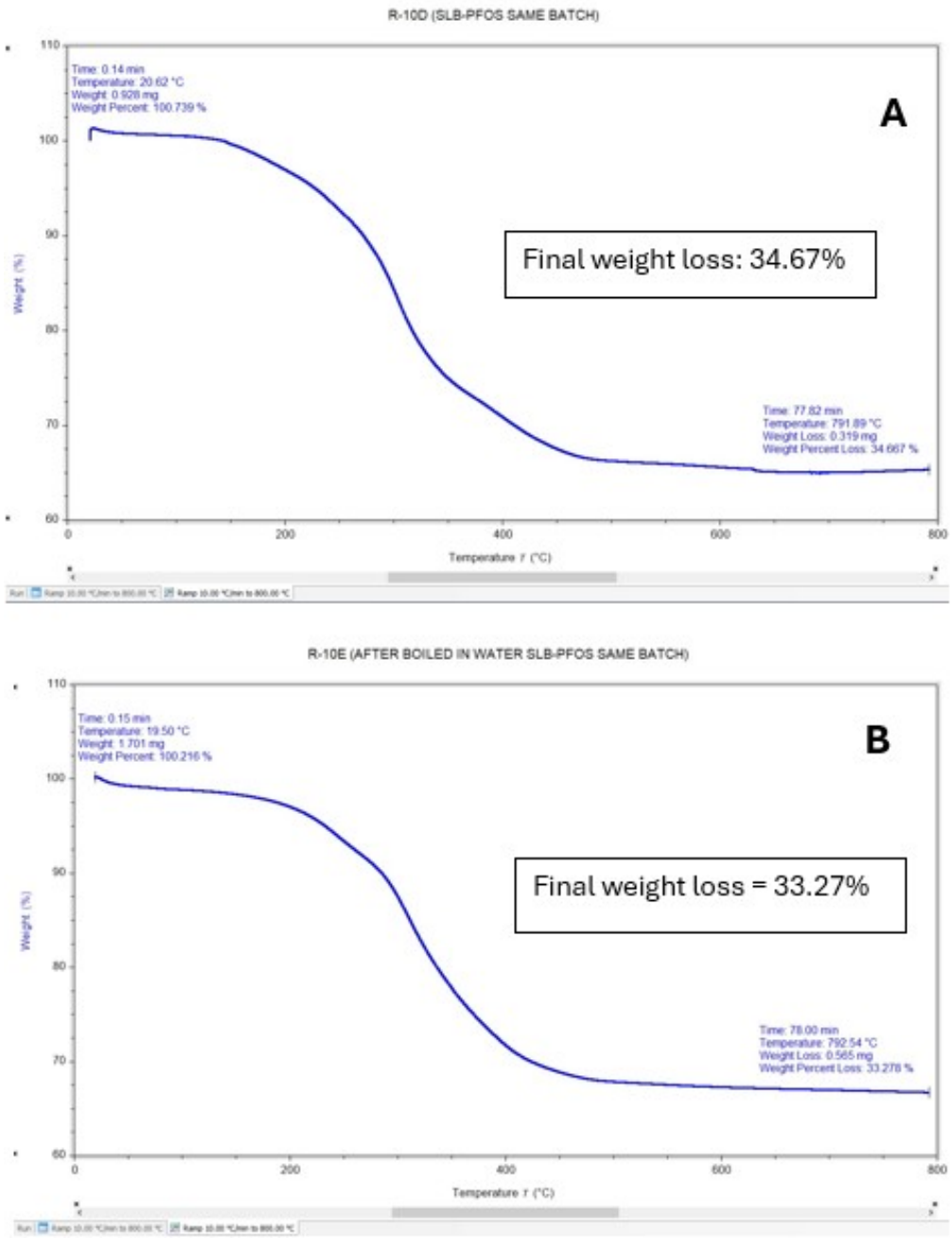


Figure S9. (**A**) TGA data for mesoporous SiO_2-SLBs that were incubated with PFOS; and (**B**) after sample in (**A**) was immersed in boiling water for 1 h and vacuum dried.

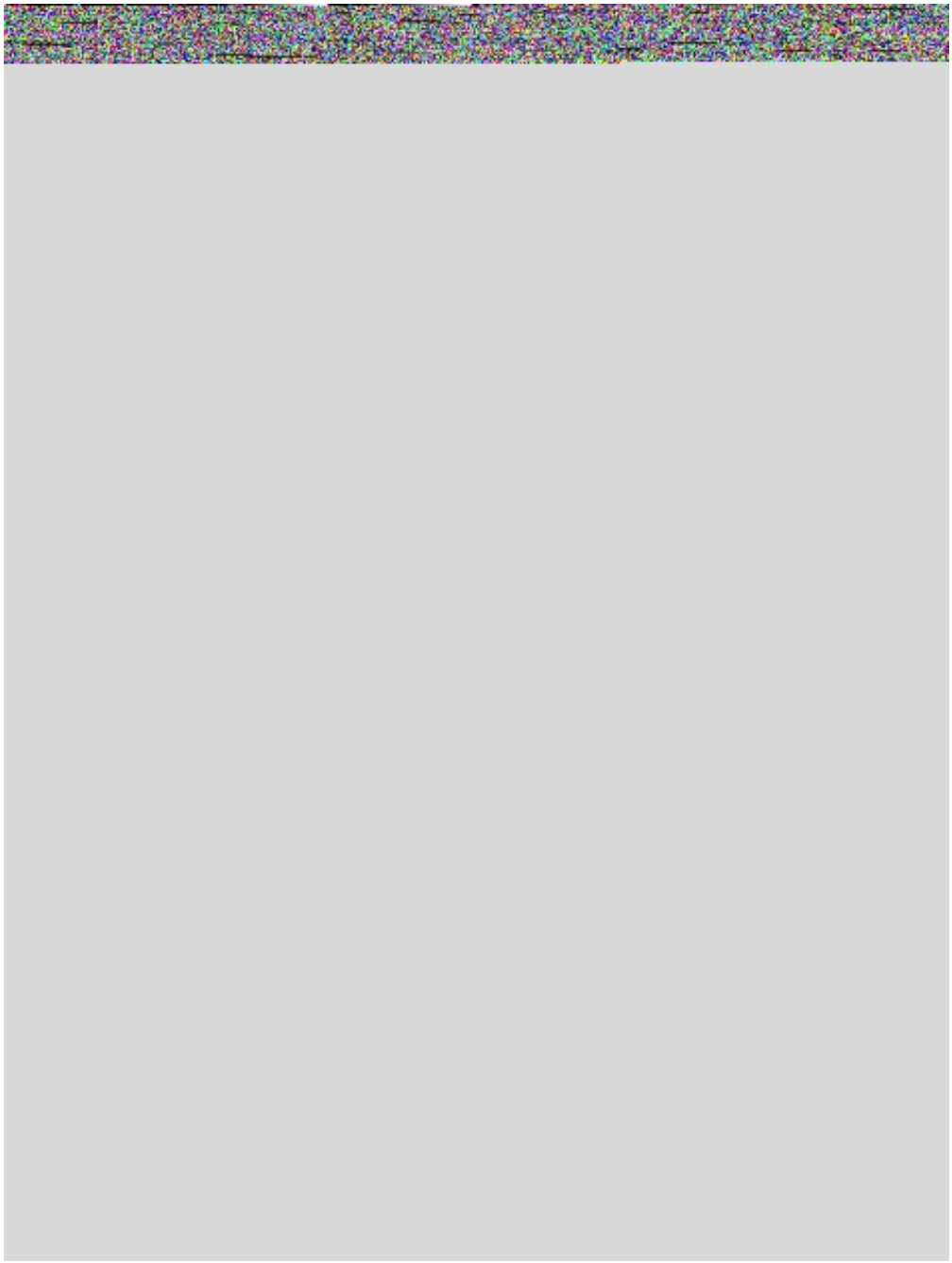
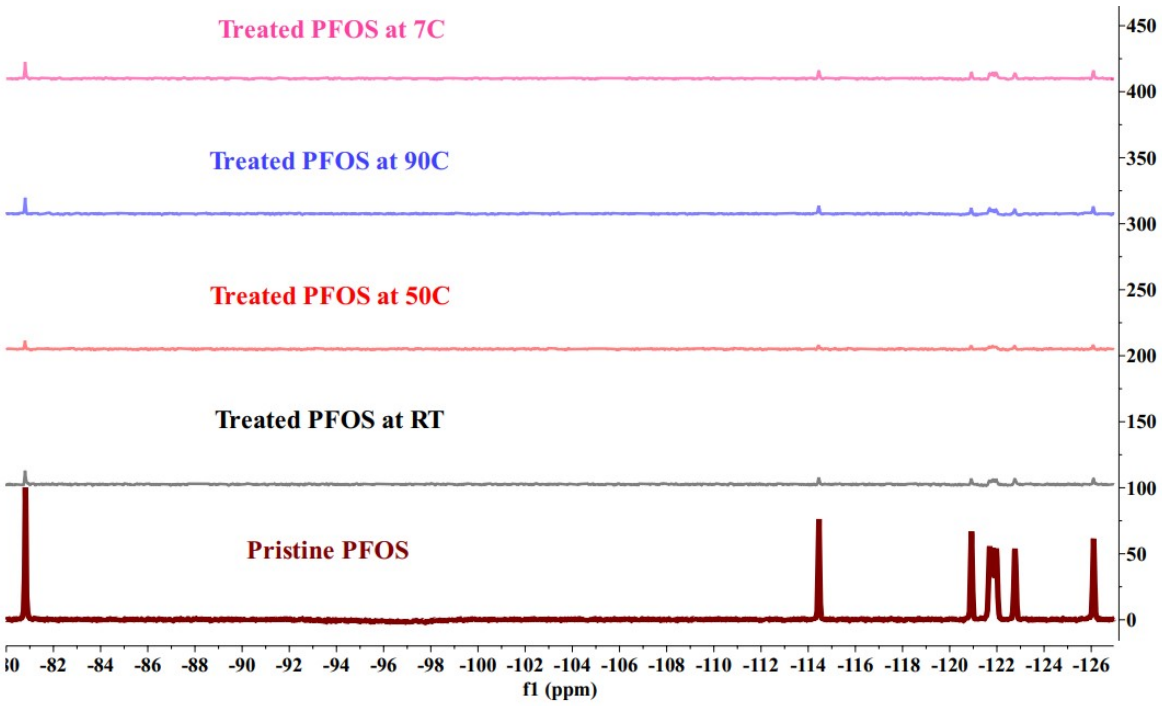


Figure S10. (**A**) TGA data for larger sample of SLBs (from batch of **Figure S7C**) that was incubated with PFOS; and (**B**) after sample in (**A**) was immersed in boiling water for 1 h and vacuum dried.

^{19}F NMR Data

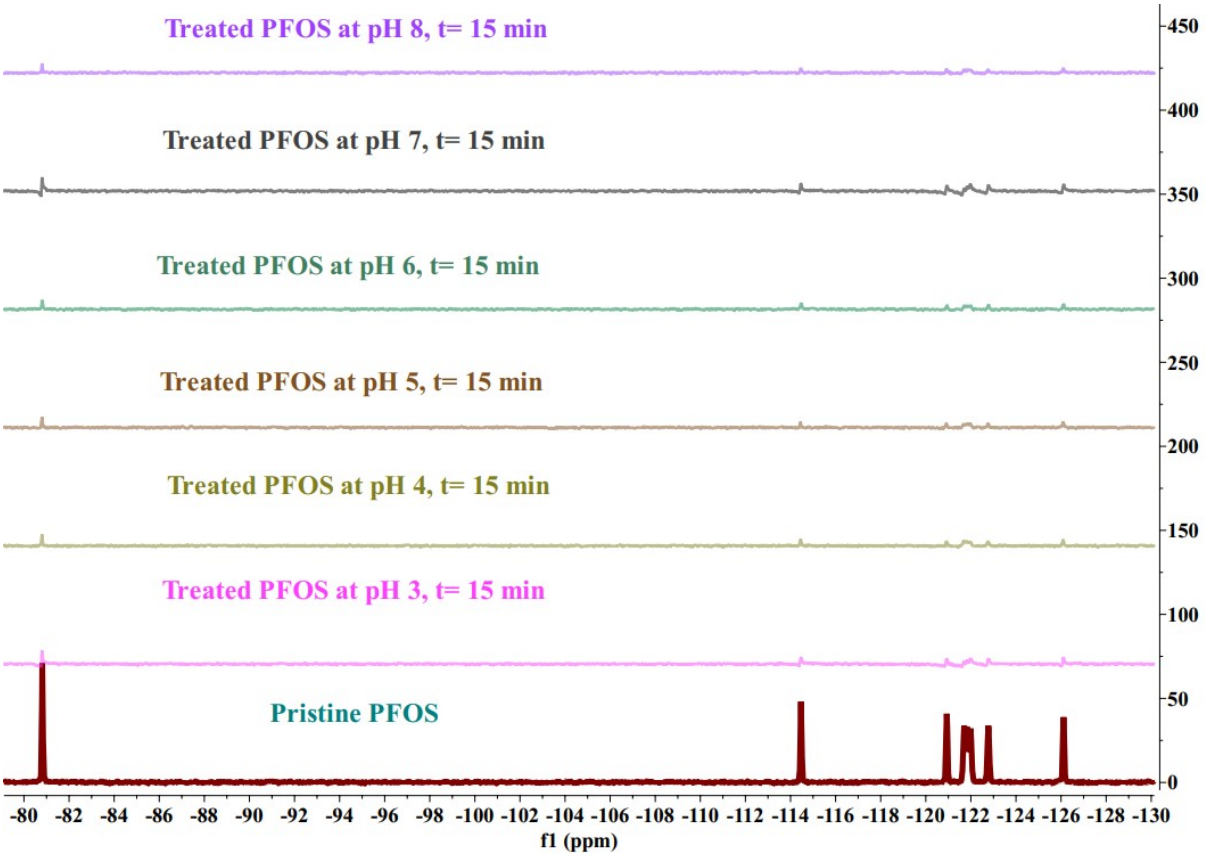
^{19}F NMR Data for the effects of temperature, pH, ionic strength and natural organic matter

For analysis of the NMR data, 65 µL of D_2O was added to 750 µL of the supernatant (containing 0.765 mg PFOS) taken from vials containing approximately 10 mg (9.66±0.4) of the SLBs that were incubated for 15 min with ~1 mL of an aqueous solution. The solution contained 1.02 mg PFOS (1.02 mg/mL PFOS) and NaCl (different molarities) and pH. The actual amounts of SLBs and solution were adjusted so that there was always 1.02 mg/mL for 10 mg SLBs. The ^{19}F NMR signal from -CF_3 was compared with that of the original solution containing 0.765 mg PFOS.



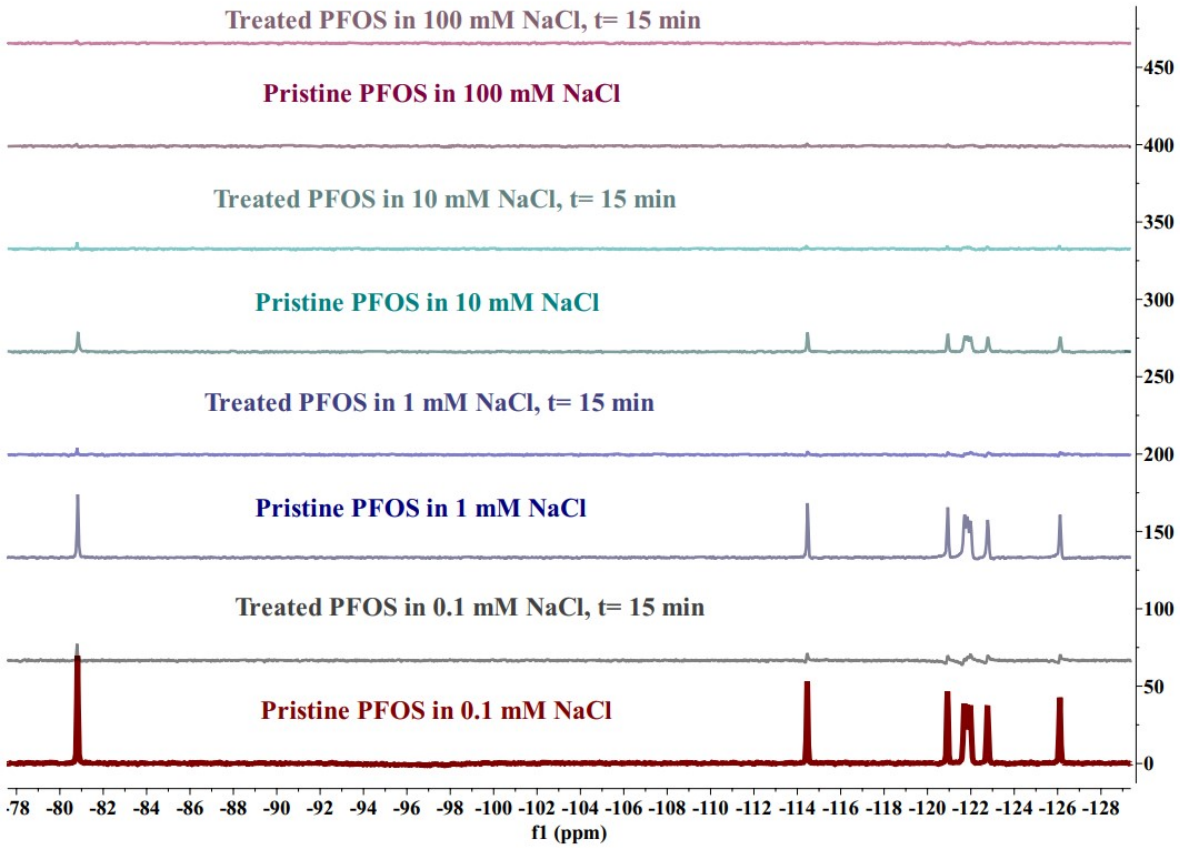
Ionic strength	Pristine	Post-treatment	Area decreased
0.1 mM NaCl	2795.37	311.16	2484.21
1 mM NaCl	1953.8	120.75	1833.05
10 mM NaCl	824.86	120.63	704.23
100 mM NaCl	34.66	70.08	-35.42

Figure S11 ^{19}F NMR of pristine PFOS and supernatant after incubation with SLBs at T = 7 ^{0}C, RT, 50 ^{0}C and 90 ^{0}C. Areas of the -CF_3 resonance at -81 ppm for neat PFOS and after incubation at different temperatures are tabulated.



pH	Pristine	Post-treatment	Area decreased
3	2218.97	318.35	1900.62
4	2496.68	206.84	2289.84
5	2319.68	206.65	2113.03
6	2427.54	211.09	2216.45
7	2795.37	311.16	2484.21
8	2557.57	155.56	2402.01

Figure S12.^{19}F NMR of pristine PFOS at T = 25 ^{0}C, 0.1 mM NaCl at different pH values, and supernatant after incubation with SLBs at T = 25 ^{0}C, 0.1 mM NaCl as a function of pH. Areas of the -CF_3 resonance at -81 ppm for neat PFOS and after incubation at different pH values are tabulated.



Ionic strength	Pristine	Post-treatment	Area decreased
0.1 mM NaCl	2795.37	311.16	2484.21
1 mM NaCl	1953.8	120.75	1833.05
10 mM NaCl	824.86	120.63	704.23
100 mM NaCl	34.66	70.08	-35.42

Figure S13. ^{9}F NMR of pristine PFOS at T = 25 ^{0}C, pH 7 and at different ionic strengths, and the supernatant after incubation with SLBs at T = 25 ^{0}C, pH = 7, as a function of ionic strength. Areas of the -CF_3 resonance at -81 ppm for neat PFOS and after incubation at different ionic strengths values are tabulated.

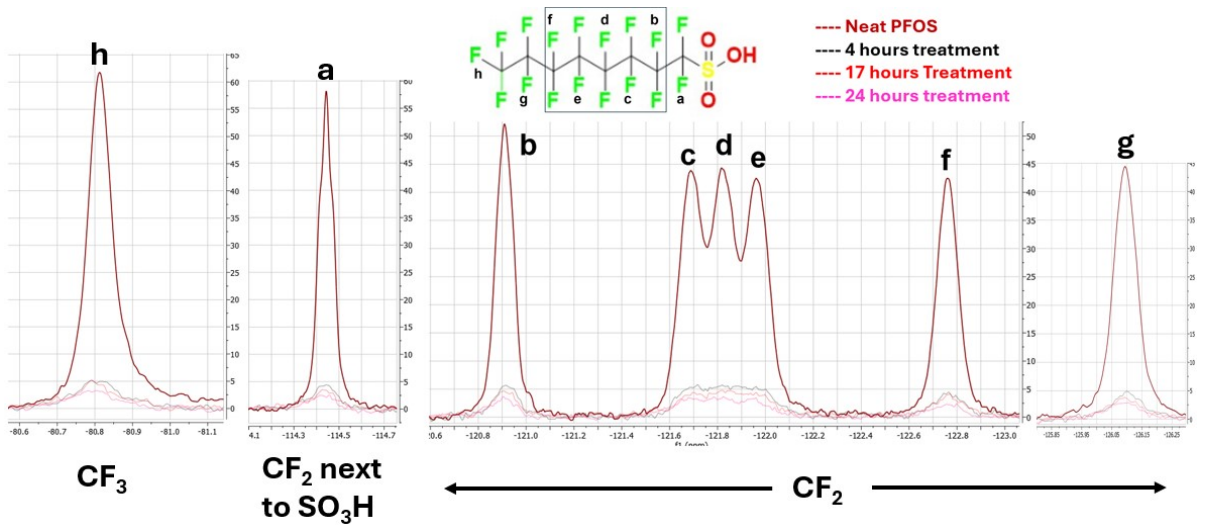


Figure S14. ^{19}F-NMR data of a Syloid®3150 mesoporous SiO_2-SLB with nominal 51% coverage (~35% experimental) hydrated with 100 mM NaCl and 20 mM $CaCl_2$ incubated with PFOS, as a function of time, compared with the pristine PFOS solution.

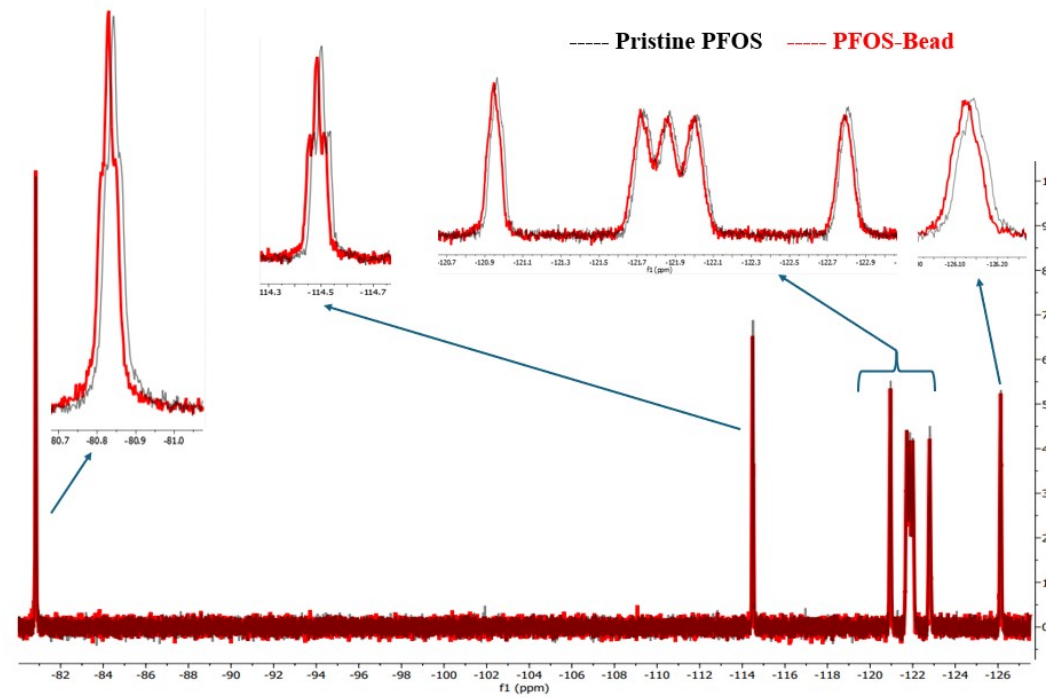


Figure S15. Control study.^{19}F-NMR data of a pristine Syloid®3150 mesoporous SiO_2 (15.4 mg) incubated in 2.5 mL 1.02 mg/mL PFOS for 1 h and centrifuged (15 min). Supernatant (750uL PFOS solution+ 80 uL D_2O) compared with (750uL PFOS solution+ 80 uL D_2O) of original PFOS solution.

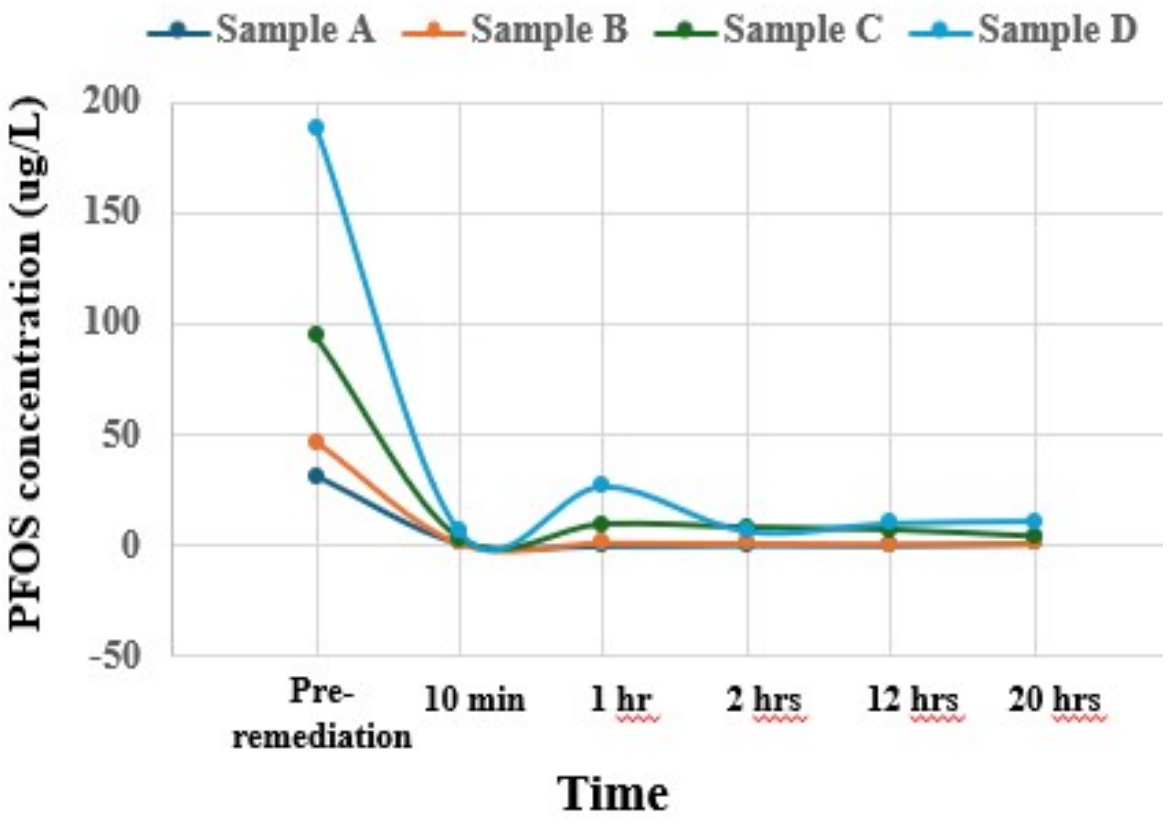


Figure S16. LC-TQOF-MS data on ug/L PFOS solutions as a function of time, showing PFOS removed < 10 min

Calculations

Lipid Coverage in the pores of the mesoporous SiO_2

When the mesoporous SiO_2 beads are immersed in $CHCl_3$ solutions of lipids, all the lipid may not be incorporated into the pore interior since:

1. Not all the pores may be accessible (e.g. too small)
2. Lipid films can also form on the exterior of the beads, and form MLVs during the hydration step.

To ensure that sorption capacities were measured for mesoporous SiO_2 beads when there were *SLBs only in the pores*, the solutions that were clear (nominal 76.3% and 49%) were used for capacity measurements. However, sorption capacities were also calculated for nominal coverages (122%, and 97.5%) after MLVs were removed from the solution, by removing the mesoporous SiO_2 and placing it in a new aqueous solution; in the latter case the capacities reflected PFOS removal from both the SLBs and the MLVs. However, since the capacities are similar to the nominal 76.3% and 49% solutions, it is likely that most of the MLVs were removed.

1. **Nominal Coverage**: The weight of lipids necessary to form SLBs/weight beads, assuming all the pores are accessible.
2. Lipid solutions above and below this calculated amount were prepared.
3. Known weights of mesoporous SiO_2 beads were immersed in the lipid solutions.
4. After removing the $CHCl_3$, the mesoporous SiO_2 beads were immersed in water and salt solutions and were freeze dried to form SLBs (expected in the pores) or MLVs (expected from lipids forming films on the exterior of the particles).
5. **Experimental Coverage**: Experimental lipid coverage for solutions after removal of $CHCl_3$ and water (with 100 mM NaCl + 20 mM $CaCl_2$) were measured by TGA.
6. **Sorption Capacity**:
    a. When MLVs were formed, the solutions were cloudy.
    b. The SLBs were removed from the cloudy solutions and rinsed.
    c. Sorption capacities were measured for the solutions not originally cloudy and for the rinsed beads (that could still contain some MLVs).

Nominal (100%) coverage: assume all pores accessible, i.e. use specific surface area (SSA) provided by manufacturer

Calculation of lipid to form a single SLB in all the pores, assuming all pores accessible, i.e. 100% lipid coverage (nominal coverage):

1. Need nominal specific surface area (SSA)- given by manufacturer. For Syloid®3150, SSA= $\frac{320 m^2 SiO_2}{g\ SiO_2}$
2. Assume area/DPPC is $\frac{0.67 nm^2}{lipid}$ and 2 lipids/bilayer
3. Calculate # DPPC molecules needed to form 100% coverage and convert SIO_2 and DPPC to weights:

For 100% coverage: $\frac{wt\ (mgDPPC)}{wt\ (x\ mgSiO_2)} = 1.168$

4. Compare the calculated (nominal) 100% coverage to actual (experimental) coverage measured by TGA to obtain Experimental % coverage.

Experimental Coverages

Experimental $\%\ coverage = \frac{\frac{\%\ DPPC\ measured\ by\ TGA}{\%\ SiO2\ maeasured\ by\ TGA}}{1.168} x100$

Nominal and Experimental DPPC coverages of mesoporous SiO_2 beads and their capacities presented in **Table 2.**