Supporting Information

Interaction of human plasma proteins with thin gelatin-based hydrogel films: a QCM-D and ToF- SIMS study

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Surface analysis of poly(*p*-xylylene) and poly(4-aminomethyl-*p*-xylylene-*co*-*p*-xylylene) *by* ToF-SIMS

Poly(*p*-xylylene) and poly(4-aminomethyl-*p*-xylylene-*co*-*p*-xylylene), non-functionalized and amino-functionalized parylenes were analyzed by ToF-SIMS. Based on these analyses, the presence of amine groups on the amino-functionalized CVD polymer was proven. As shown in Figure SI-1, the amino-functionalized CVD polymer is characterized by several C_xNH_y peaks in the positive polarity secondary ions spectrum.

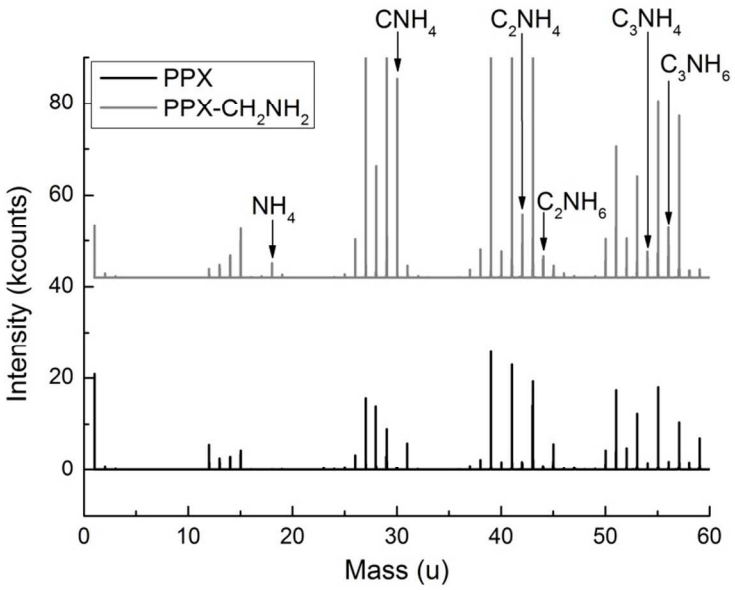


Figure SI-1. Positive polarity ToF-SIMS spectra of amino-functionalized parylene (up, grey) and non-functionalized parylene (down, black). Fragments indicating the presence of amine groups are marked with chemical assignments.

Characterization of thin hydrogel films

Table SI-1. Tentative assignment of some bands frequently found in IRRAS spectra of biological materials[1] and measured bands.

Label	Wavenumber measured bands [cm^{-1}]	Assignment[1]
A	3300-3600	O-H str. of hydroxyl groups
B	3200-3300	N-H str. of amide groups
C	2800-2950	C-H str. (sym) of $–CH_3$
D	1600-1700	Amide I band
E	1400-1550	Amide II band
F	1300-1400	Amide III band

Quartz Crystal Microbalance with Dissipation monitoring - Viscoelastic model

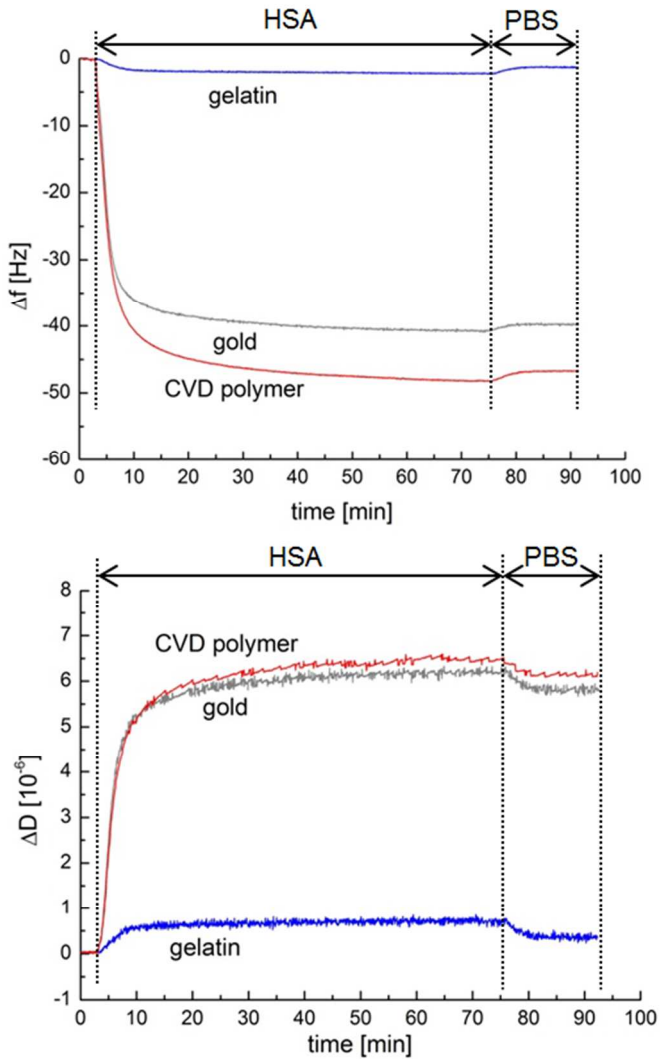


Figure SI-2. Frequency shifts (a) and dissipation shifts (b) during HSA adsorption on CVD coating (red), bare gold (grey) and gelatin-based hydrogel (blue) at third overtone (f = 15 MHz) at 37 °C.

Slope in $\Delta D/(-\Delta f)$-Plot

The ratio $\frac{\Delta\Gamma}{-\Delta f}$ measured for a thin soft film on a QCM-D sensor in a liquid is derived[2] to

$\frac{\Delta\Gamma_n}{-\Delta f_n} = 2\pi n f_0 \eta J'_f$, (eq.1).

where f denotes the resonance frequency, Γ the imaginary part of the resonance frequency, n is the harmonic order, f_0 is the fundamental frequency, η is the viscosity of the liquid and J'_f is the real part of the shear compliance. The Δ indicates the difference to the reference state (i.e. initial values).

By using the relation between the dissipation D and Γ (i.e. $D=2\Gamma/f$) and knowing that $f_n \approx n f_0$, we can rewrite eq.1 as

$J'_f = \frac{1}{4\pi\eta}\frac{\Delta D_n}{-\Delta f_n}$ (eq.2).

The shear compliance is defined as $J=1/G$, with G denoting the shear modulus. If the film is much more rigid than the liquid (which is the case here), the imaginary part of the compliance J'_f is much smaller than J'^2_f and G'=1/J' follows. This means the shear modulus of the thin film can be determined by

$G_f = \frac{4\pi\eta}{\Delta D_n/-\Delta f_n}$ (eq.3).

***ΔD*$_n$*/(-Δf*$_n$*)* values during the protein adsorption process**

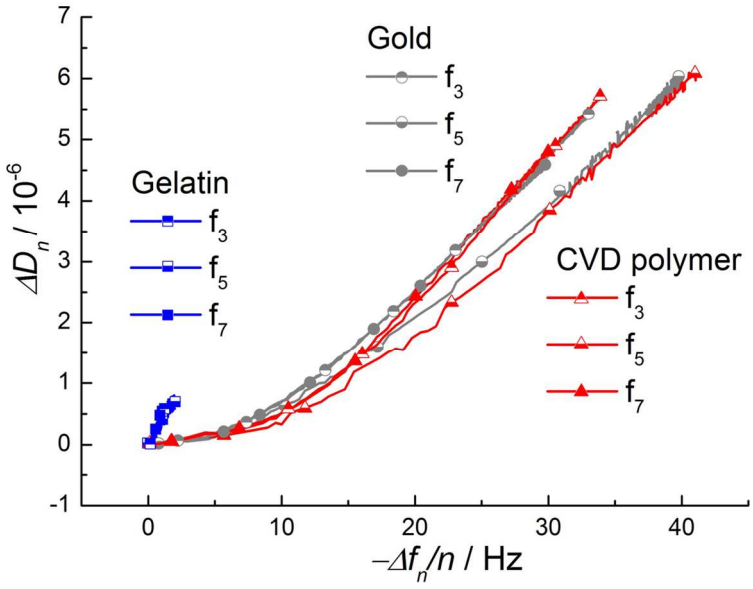
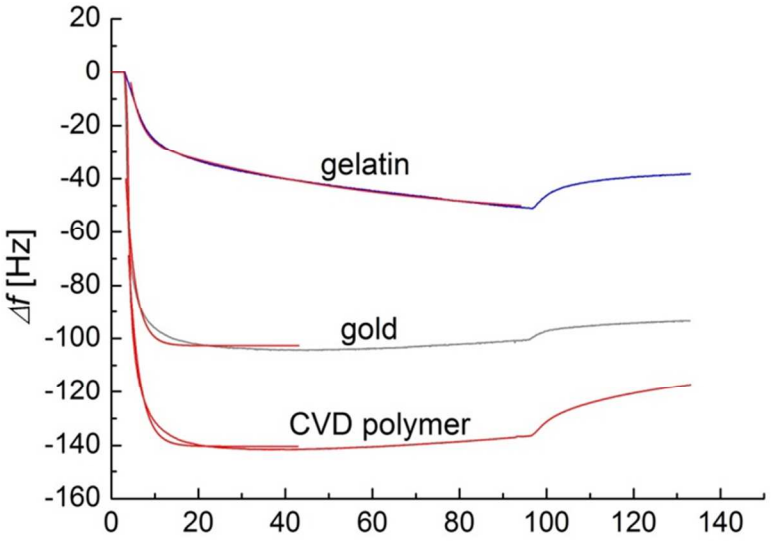


Figure SI-3. *ΔD*$_n$*/(-Δf*$_n$*)* values during the adsorption process of HSA on the gelatin-based hydrogel (blue shades; squares), on bare gold (grey shades; circles) and on the CVD polymer (magenta shades; triangles). For sake of clarity, only the third, the fifth and the seventh resonance frequencies are shown.

Detailed characterization of the adsorption kinetic by exponential fitting

Assuming identical time constants for the initial protein adsorption t1 = 150 s (determined by exponential fitting of recorded -Δf curve from gold, t1 = 134 s, respectively CVD, t1 = 149 s), the measured -Δf curve of the protein adsorption on the gelatin hydrogel can be fitted nicely by using an exponential fit.

To suppose a two-step process with a time constant t1 = 150 s (protein accumulation on top) and t2 = 3300 s (protein diffusion in the gel), the calculated values A1 = 126,5 and A2 = 33,2 leads to a ratio 4:1 of adsorbed and diffused fibrinogen on, respectively in the hydrogel.



Model	ExpDec2		
Equation	y = A1*exp(-x/t1) + A2*exp(-x/t2) + y0		
Reduced Chi-Sqr	0,19175		
Adj. R-Square	0,99734		
		Value	Standard Error
f3	y0	-55,97114	0,15473
	A1	126,48822	0,8924
	t1	150	0
	A2	33,18205	0,1036
	t2	3296,67321	36,33738

Figure SI-4: Exponential fits of the -Δf values from fibrinogen adsorption on gelatin-based hydrogel, gold and CVD polymer.

***ΔD/(-Δf)* ratios and calculated shear moduli**

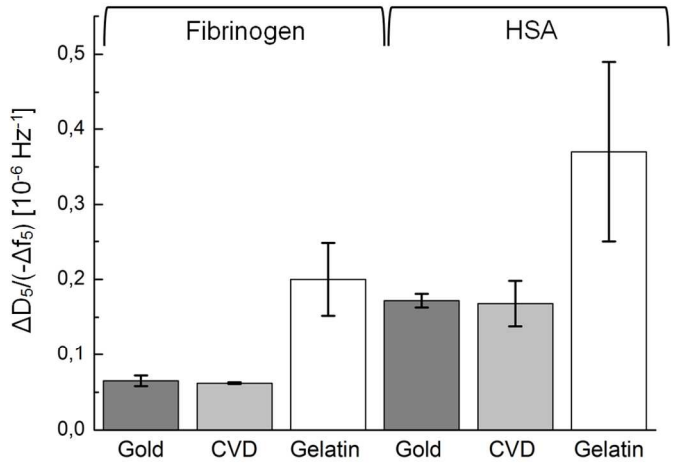


Figure SI-5: *ΔD/(-Δf)* ratio of the adsorbed wetted protein mass on the gelatin-based hydrogel, gold and the CVD functionalized substrate at fifth overtone.

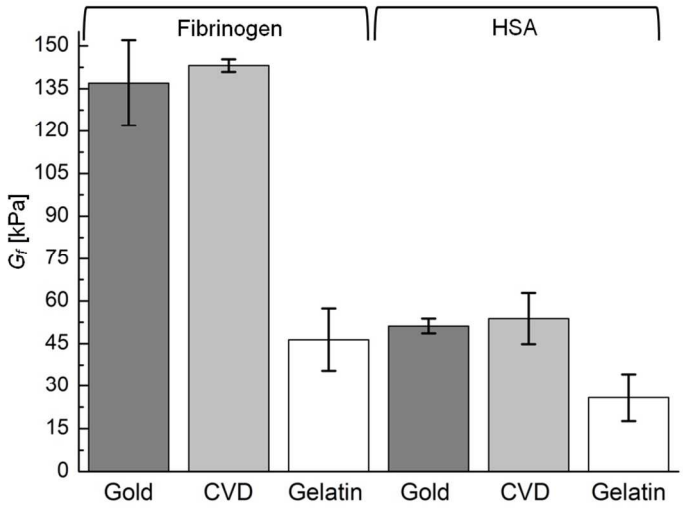


Figure SI-6: Shear moduli from the adsorbed wetted protein mass on the gelatin-based hydrogel, gold and the CVD functionalized substrate at fifth overtone calculated by eq. 3 (see Supporting Information, page 5).

Additional characterization of the gelatin-based hydrogel film with and without adsorbed fibrinogen respectively HSA using ToF-SIMS and principal component analysis

Table SI-2. Principal component analysis of gelatin and fibrinogen respectively HSA. Determined mass [amu], loadings for PC1 and PC2, chemical assignment of the fragment (positive polarity) and amino acid origin (see also Figure 7 main text).

Mass	PC1	PC2	Assignment	Amino Acid (Code)
30.04	-0.46	-0.21	CH_4N	Glycine (Gly, G)
43.03	0.16	-0.23	CH_3N_2	Arginine (Arg, R)
44.05	-0.13	-0.31	C_2H_6N	Alanine (Ala, A)
44.98	0.01	-0.03	CHS	Cysteine (Cys, C)
60.05	-0.01	-0.01	C_2H_6NO	Serine (Ser, S)
61.01	0.02	-0.09	C_2H_5S	Methionine (Met, M)
68.05	-0.33	-0.08	C_4H_6N	Proline (Pro, P)
69.03	-0.07	-0.05	C_4H_5O	Threonine (Thr, T)
70.03	-0.10	0.03	C_3H_4NO	Asparagine (Asn, N)
70.07	-0.44	-0.06	C_4H_8N	Proline (Pro, P)
71.01	0.00	0.02	$C_3H_3O_2$	Serine (Ser, S)
72.08	0.03	-0.07	$C_4H_{10}N$	Valine (Val, V)
73.07	0.24	-0.43	$C_2H_7N_3$	Arginine (Arg, R)
74.06	0.07	-0.04	C_3H_8NO	Threonine (Thr, T)
81.04	0.01	-0.07	$C_4H_5N_2$	Histidine (His, H)

82.06	-0.17	-0.04	$C_4H_6N_2$	Histidine (His, H)
83.05	-0.02	-0.01	C_5H_7O	Valine (Val, V)
84.05	-0.14	-0.05	C_4H_6NO	Glutamic Acid (Glu, E)
84.09	-0.46	0.09	$C_5H_{10}N$	Lysine (Lys, K)
86.10	0.02	-0.41	$C_5H_{12}N$	Leucine (Leu, L)*
87.05	0.10	0.26	$C_3H_7N_2O$	Asparagine (Asn, N)
88.04	-0.02	0.04	$C_3H_6NO_2$	Aspartic Acid (Asp, D)
98.02	-0.04	0.09	$C_4H_4NO_2$	Asparagine (Asn, N)
100.09	0.11	-0.36	$C_4H_{10}N_3$	Arginine (Arg, R)
101.1	0.07	-0.14	$C_4H_{11}N_3$	Arginine (Arg, R)
102.06	-0.09	0.04	$C_4H_8NO_2$	Glutamic Acid (Glu, E)
107.05	0.06	0.03	C_7H_7O	Tyrosine (Tyr, Y)
110.08	0.04	-0.27	$C_5H_8N_3$	Histidine (His,H)
120.08	-0.07	-0.22	$C_8H_{10}N$	Phenylalanine (Phe, F)
127.10	-0.12	-0.12	$C_5H_{11}N_4$	Arginine (Arg, R)
130.07	0.16	0.16	C_9H_8N	Tryptophan (Trp, W)
131.05	-0.01	-0.03	C_9H_7O	Phenylalanine (Phe, F)
132.05	-0.00	-0.00	C_9H_8O	Phenylalanine (Phe, F)
136.09	0.13	-0.14	$C_8H_{10}NO$	Tyrosine (Tyr, Y)
170.07	0.04	0.04	$C_{11}H_8NO$	Tryptophan (Trp, W)

For the principal component analysis of the secondary ion mass spectra of adsorbed protein, as shown in Figure 7 and Table SI-2, peaks in positive polarity spectra were selected based on conventional datasets[3, 4] of canonical poly(amino acids). Integration intervals were defined from the peak shapes in high mass resolution spectra directly (in other studies several signals with the same nominal mass are combined). The fragment CH_4N is detectable also for other amino acids beside glycine. Leucine and isoleucine both yield $C_5H_{12}N$.

Amino acid compositions of the studied proteins

Table SI-3. Amino acid composition of HSA[5], human Fbn[5] and porcine gelatin[6].

Amino acid	Composition HSA [mol %]	Composition human Fbn [mol %]	Composition gelatin [mol %]
Ala (A)	10.34	4.86	11.14
Arg (R)	4.43	5.19	4.85
Asn (N)	2.79	5.58	0.0
Asp (D)	5.91	6.19	4.67
Cys (C)	5.75	1.99	0.0
Gln (Q)	3.28	4.42	n.d.
Glu (E)	10.18	6.63	7.13
Gly (G)	2.13	9.56	32.61
His (H)	2.63	2.21	0.60
Ile (I)	1.48	3.76	0.96
Leu (L)	10.51	6.57	2.36
Lys (K)	9.85	6.24	2.63
Met (M)	1.15	2.32	0.55
Phe (F)	5.75	3.37	1.44
Pro (P)	3.94	4.36	13.06
Ser (S)	4.60	9.45	3.65
Thr (T)	4.76	6.19	1.71
Trp (W)	0.33	2.43	0.0
Tyr (Y)	3.12	3.81	0.31
Val (V)	7.06	4.86	2.19
Pyl (O)	0.0	0.0	0.0
Hyp	0.0	0.0	9.55
Hyl	0.0	0.0	0.6

References

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