

Supplementary Information

Tuning Protein Adsorption on Graphene Surfaces via Laser-Induced Oxidation

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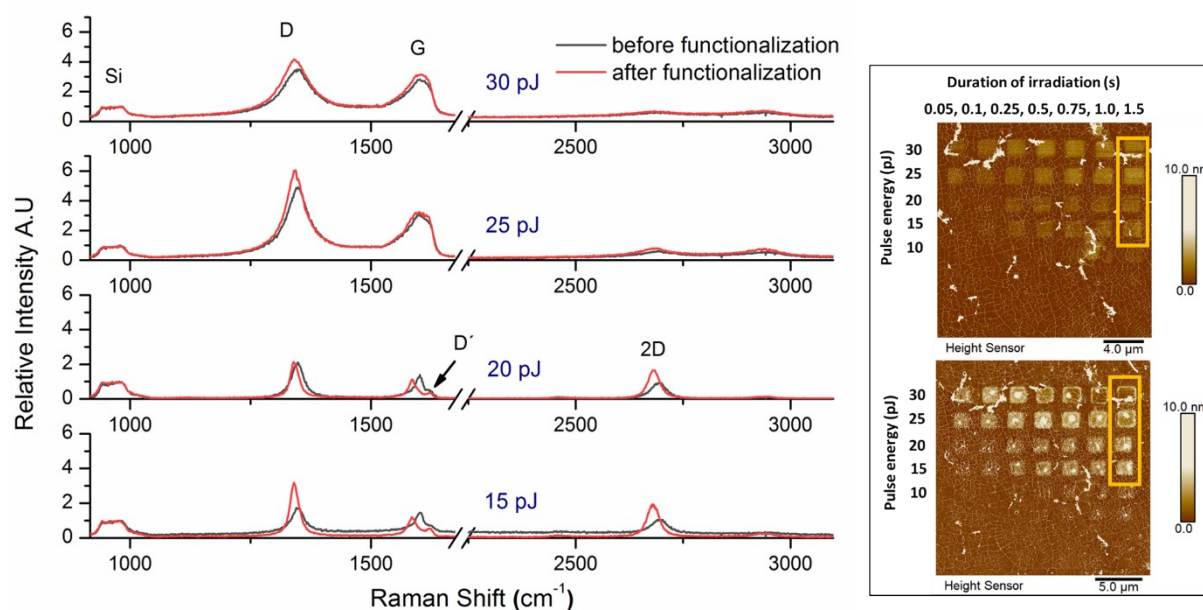


Figure S1. Raman spectra before and after HRP adhesion. Increased pulse energy (15 to 30 pJ) for a constant irradiation time of 1.5 s. Insert depicts the location of the corresponding squares, before (top) and after (bottom) HRP adhesion. All spectra are normalized with respect to the Si band intensities.

Table S1. Raman shifts (cm^{-1}), quantified by the Lorentzian function fitting procedure, corresponding to the spectra depicted in **Figure S1**. Shifts are given in black colour font (before HRP adhesion) and red colour font (after HRP adhesion).

Pulse energy (pJ)	D	G	2D	D+D'
30	1347.8	1596.6	2692.5	2933.5
	1343.6	1595.8	2683.7	2930.5
25	1348.5	1596.5	2693.3	2933.9
	1343.2	1595.5	2683.6	2931.4
20	1347.4	1599.8	2693.0	2939.8
	1341.3	1586.2	2681.8	---
15	1348.3	1600.7	2694.8	---
	1340.9	1586.7	2680.4	---

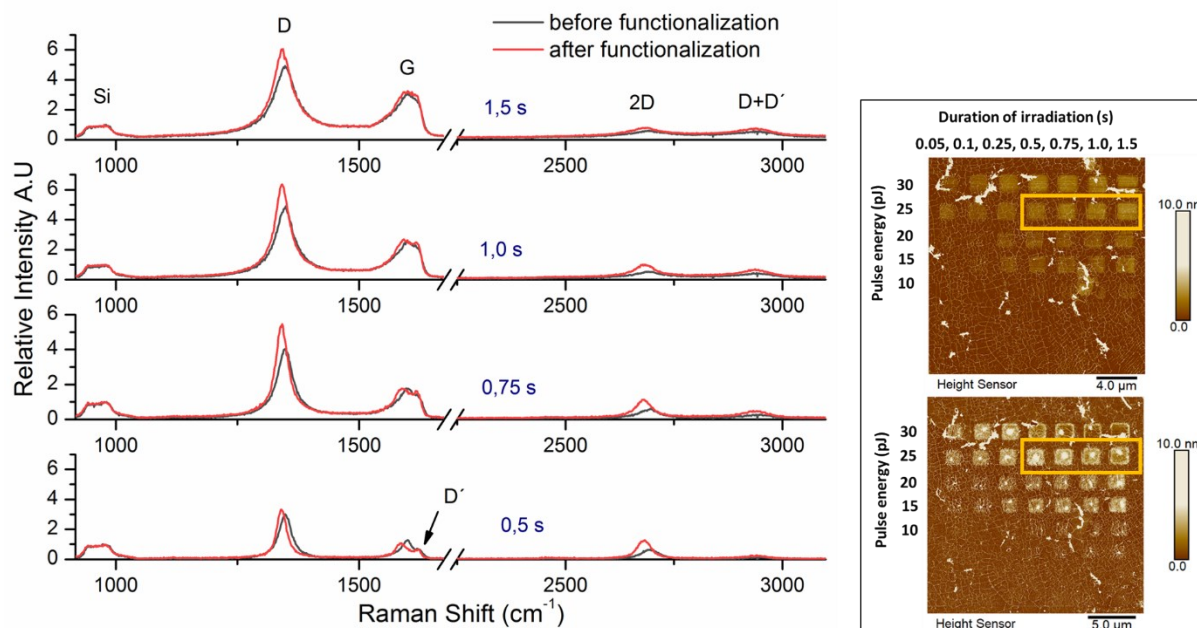


Figure S2. Raman spectra before and after HRP adhesion. Increased irradiation time at a constant pulse energy of 25 pJ. Insert depicts the location of the corresponding squares, before (top) and after (bottom) HRP adhesion. All spectra are normalized with respect to the Si band intensities.

Table S2. Raman shifts (cm^{-1}), quantified by the Lorentzian function fitting procedure, corresponding to the spectra depicted in **Figure S2**. Shifts are given in black colour font (before HRP adhesion) and red colour font (after HRP adhesion).

Duration of irradiation (s)	D	G	2D	D+D'
1.5	1347.3	1600.6	2689.2	2938.1
	1342.6	1599.5	2684.9	2931.7
1.0	1347.1	1600.9	2690.3	2941.2
	1341.9	1598.6	2682.0	2934.8
0.75	1347.6	1601.0	2692.6	2943.7
	1341.1	1596.1	2679.8	2937.4
0.5	1348.3	1599.7	2694.5	2949.9
	1340.9	1586.9	2681.0	2938.6

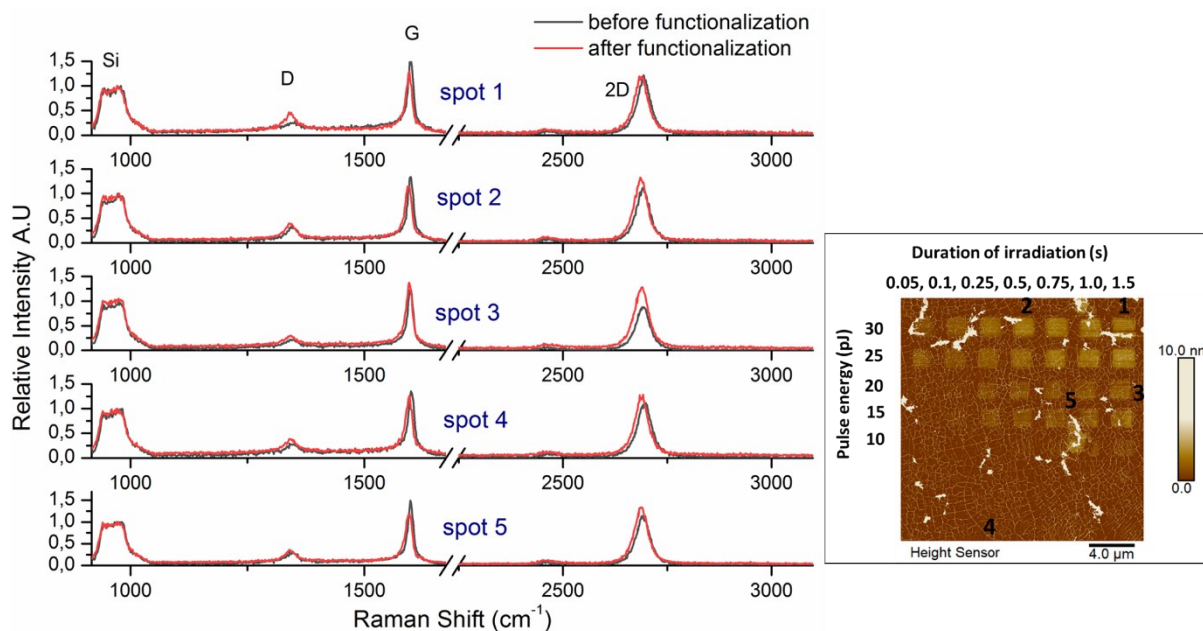


Figure S3. Raman spectra before and after HRP adhesion. Non-irradiated graphene spots. Insert depicts the location of the corresponding spots. All spectra are normalized with respect to the Si band intensities.

Table S3. Raman shifts (cm^{-1}), quantified by the Lorentzian function fitting procedure, corresponding to the spectra depicted in **Figure S3**. Shifts are given in black colour font (before HRP adhesion) and red colour font (after HRP adhesion).

Non-irradiated spots	D	G	G*	2D
spot 1	1347.4 1343.2	1600.3 1596.6	2465.7 ---	2695.1 2688.6
spot 2	1347.6 1343.0	1599.6 1595.4	2464.2 ---	2693.1 2687.9
spot 3	1345.9 1342.7	1599.7 1597.3	--- ---	2692.4 2689.6
spot 4	1347.8 1344.1	1601.6 1596.6	--- ---	2696.8 2689.6
spot 5	1345.6 1343.1	1600.2 1596.3	--- ---	2693.1 2688.8

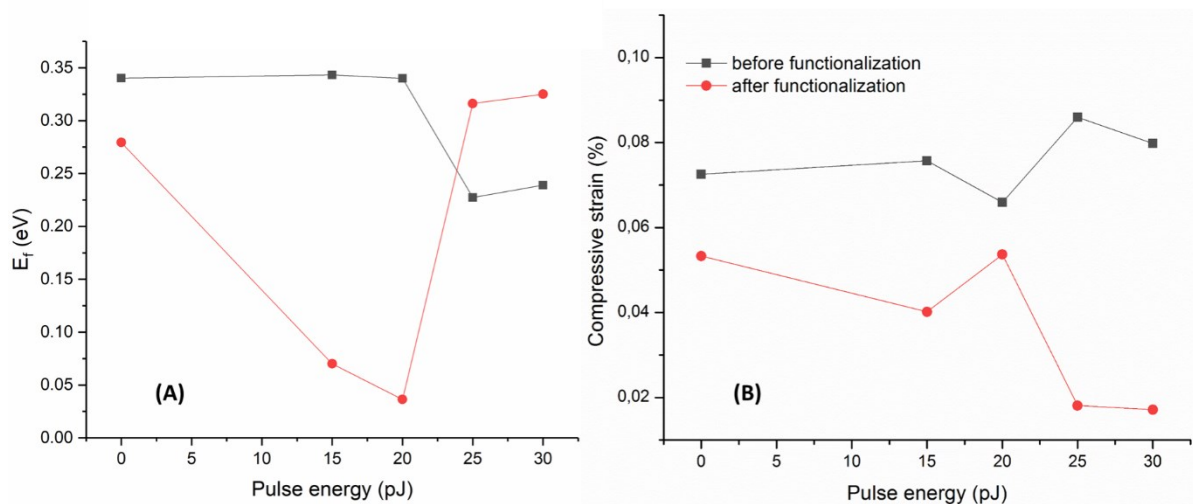


Figure S4. Fermi level (A) and compressive strain (B) plots calculated from the obtained Raman data before and after HRP adhesion. The values correspond to graphene squares irradiated at a constant duration of 1.5 s over a range of pulse energies (corresponding Raman spectra are given in **Figure S1**).

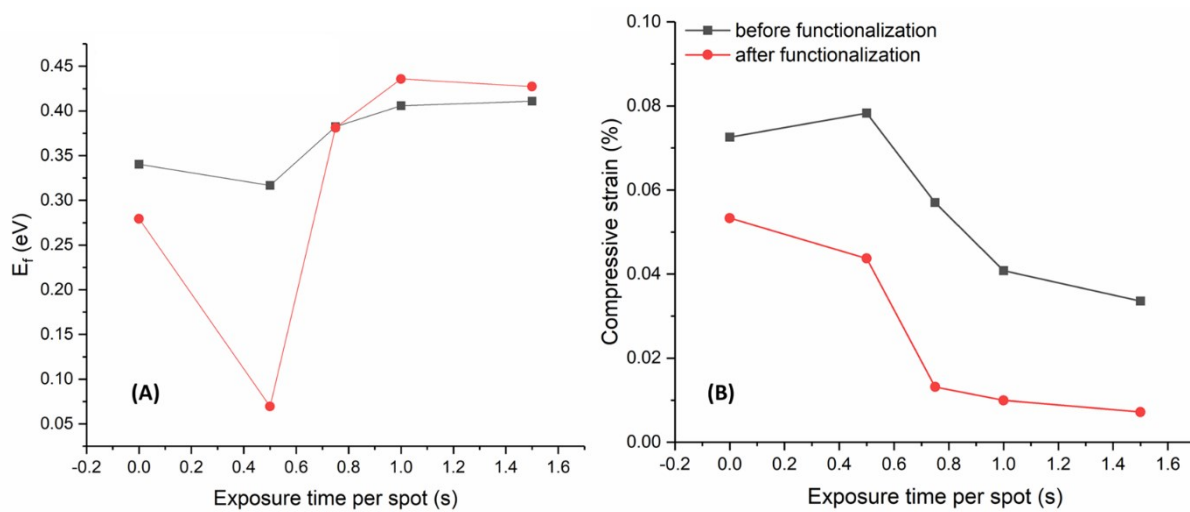


Figure S5. Fermi level (A) and compressive strain (B) plots calculated from the obtained Raman data before and after HRP adhesion. The values correspond to graphene squares irradiated at a constant pulse energy of 25 pJ over a range of time durations. (corresponding Raman spectra are given in **Figure S2**).

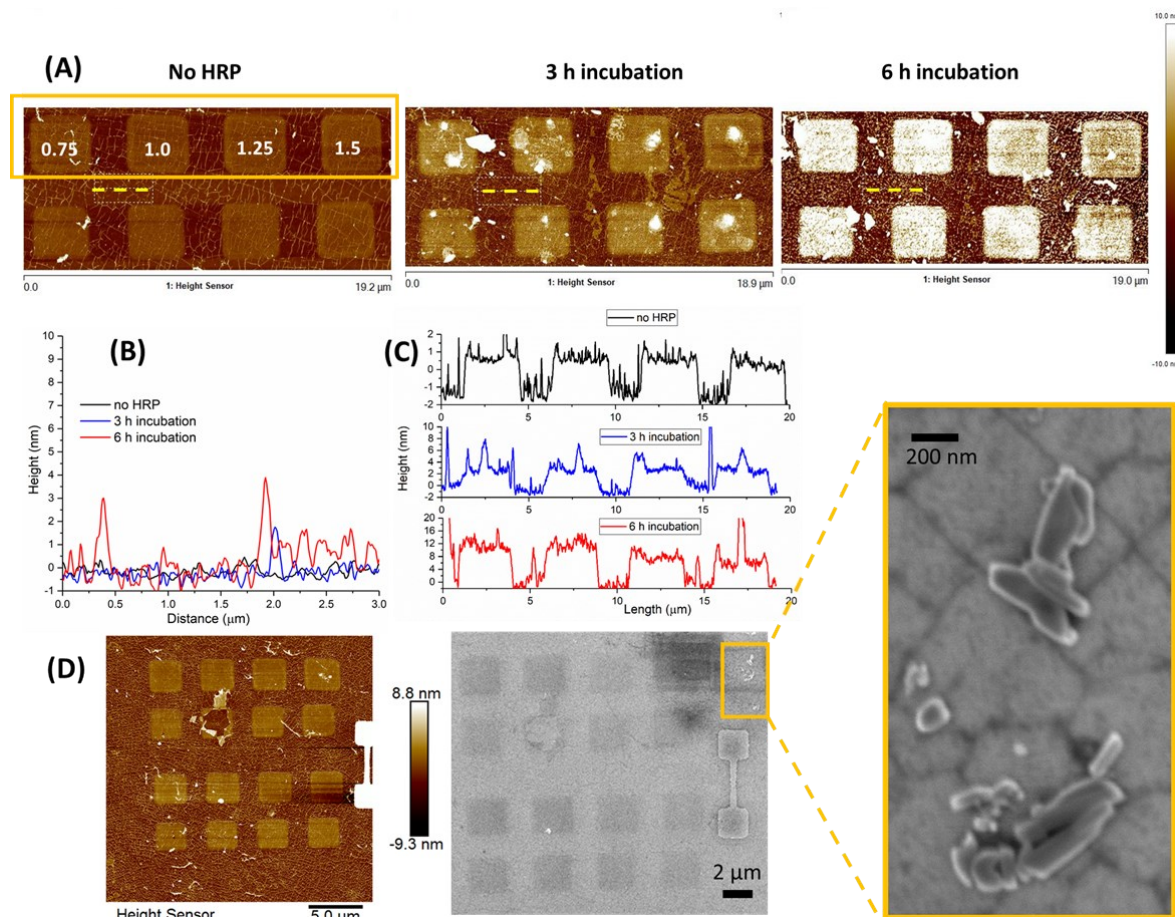


Figure S6. Control measurements of protein adhesion based on topographic height profiles. (A) AFM height sensor images of a selected area at different cycles of incubation. Graphene was irradiated by laser pulse energy of 15 pJ at different time durations (orange frame); (B) Topographic height profile of non-irradiated graphene (yellow dashed line) at different cycles of incubation; (C) Topographic height profile of irradiated graphene (orange frame) at different cycles of incubation; (D) Comparison of the microchip's AFM height sensor image with the corresponding SEM image. Crystals of the PBS solutes (magnification insert) are distinct in shape compared to the immobilized HRP upon the irradiated squares. Images were captured after incubating the microchip in HRP solution for 1 h.

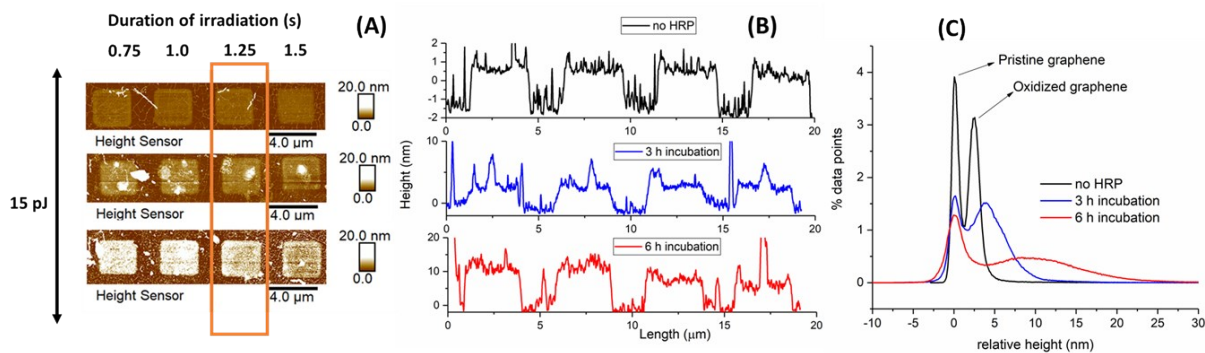


Figure S7. Incubation studies in HRP solution. (A) AFM height sensor images of a selected area at different cycles of incubation. Graphene was irradiated by laser pulse energy of 15 pJ at different time

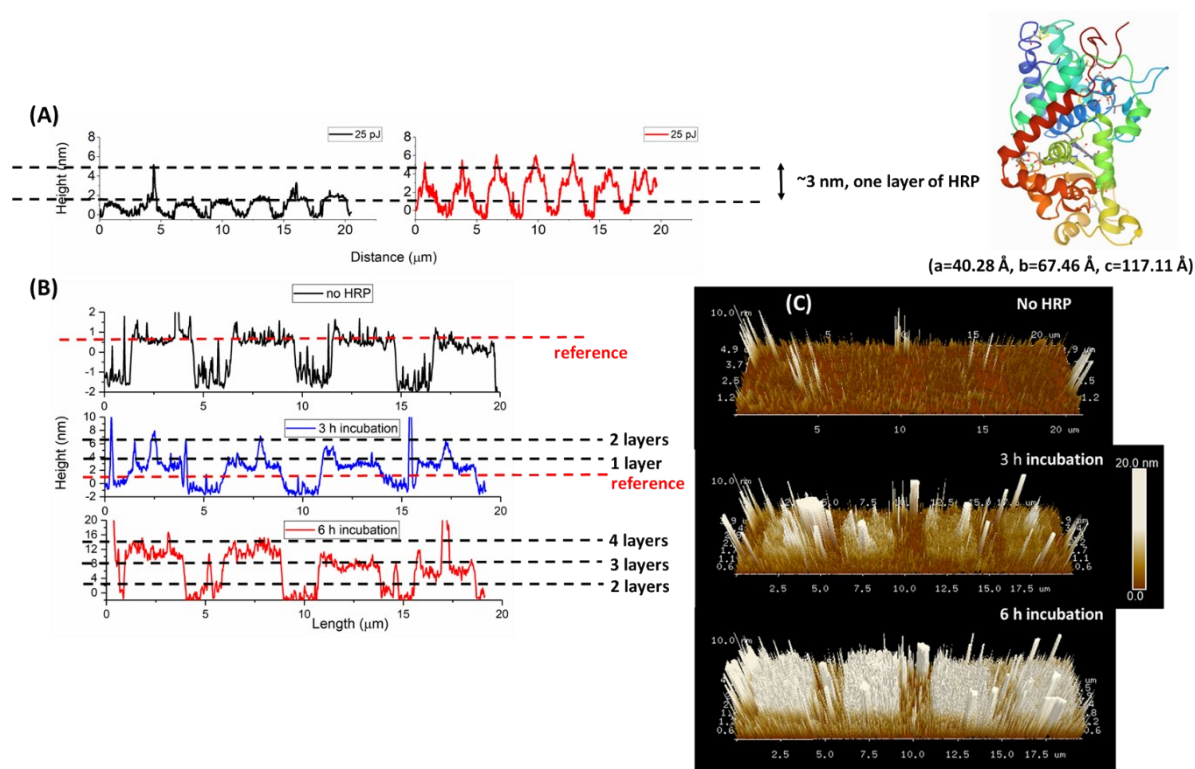


Figure S8. Layer deposition of HRP enzyme. (A) Topographic height profile of irradiated graphene (25 pJ for a range of time durations) before (black) and after (red) incubation in HRP solution for 1 h. A 3 nm increase in height suggests the deposition of a single layer of HRP. The insert depicts the structure of HRP enzyme and given dimensions (protein data bank ID: 1HCH)¹; (B) Topographic height profile of irradiated graphene (15 pJ for a range of time durations) at different incubation cycles. After 3 h of incubation 2 layers of HRP appear to be immobilized while after 6 h almost 4 layers of HRP have been deposited; (C) Three-dimensional images of the corresponding microchip area at different incubation cycles showing the immobilization of HRP upon irradiated graphene.

References

1. G. I. Berglund, G. H. Carlsson, A. T. Smith, H. Szöke, A. Henriksen, J. Hajdu, *Nature*, 2002, **417**, 463-468.