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Supporting Information

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Chemically Exfoliated MoS_2 as Near-Infrared Photothermal Agents**

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As referred to in the manuscript, this document contains detailed experiment procedures and supporting data and figures.

ceMoS_2 Synthesis (reiterated): All materials were obtained from Sigma Aldrich, unless otherwise indicated. Procedures for were adapted from Joensen, et al.[1] In a nitrogen environment, 300 mg of MoS_2 powder was immersed in 3 ml of n-butyllithium (1.6 M in hexane). Solution was stirred for 48 hr at room temperature. Mixture was then filtered over Whatman #41 filter and rinsed with 100 ml of hexane. Mixture was collected and then removed from the nitrogen environment. To the semi-dry mixture, 300 ml of di-H_2O (18.2 MΩ•cm at 25 °C) was added. Mixture was sonicated for 1 hr to achieve exfoliation. The MoS2 nanosheets were then centrifuged and washed with di-H_2O 3 times. It was then collected and dialyzed against di-H_2O for 5 days. Concentration was determined by ICP-MS.

Atomic Force Microscopy. AFM samples were prepared by either drop casting or dip coating MoS_2 solution, diluted to the point of no visible color, on to a silicon chip with 300 nm SiO_2. Samples were placed in vacuum chamber and allowed to dry for 2 hours. Images were acquired using a Park XE-100 AFM operating in tapping mode with a silicon nitride cantilever.

Inductively coupled plasma mass spectrometry (ICP-MS): ICP-MS specimens were prepared by adding fixed aliquots (2.5 to 50 µl) of ceMoS_2 solution to 100µl of 1:1 HCL:HNO_3 (trace grade). Mixture was then placed in 75 ^{o}C water bath for digestion (6 hours). 25 µl of ICP-MS internal standard solution was then added, and solution was diluted to 5 ml using di-H_2O. External standards were prepared accordingly using Mo standard solution from Sigma Aldrich. Each external standard is made with varying quantities of Mo standard, 100 µl of 1:1 HC:HNO_3 (trace grade) and 25 µl of internal standard. Each external standard solution was then diluted to 5 ml, as with the case of specimens. Accuracy of external standard preparation was checked by linear regression of ICP-MS signal (R^2 > 99.999). Mo concentration of undiluted specimen was then determined by using dilution factor described above. Mass of MoS_2 in solution was estimated by taking formula weight ratios of MoS_2 versus Mo.

NIR laser heating of MoS_2 in water: The output of a titanium:sapphire laser (Tsunami, Spectra Physics) centered at 800 nm and operating in continuous wave mode was directed into a 7 mm diameter circular well containing 100 µl of solution. The ~ 3 mm beam spot was centered in the well and measured 0.8-0.82 W at the sample. A thermocouple was positioned in the solution at the edge of the well to avoid interaction with the laser. All solutions were equilibrated to room temperature prior to laser heating.

X-Ray Photoelectron Spectrascopy (XPS). XPS samples were prepared by drop casting MoS_2 solution on to a silicon chip with 300 nm SiO_2. Samples were placed in vacuum chamber and allowed to dry overnight. Spectra were acquired using a Thermo Scientific ESCALAB 250 Xi.

Time-Domain Thermotransmission (TDTT) measurements: TDTT was used to measure the thermal boundary conductance from the ceMoS_2 nanosheets into the water. In short, TDTT is a pump-probe technique in which the change in transmission of a probe beam is monitored as a function of time delay of the pump-beam, which acts as a modulated heating event. Our TDTT system is centered around

pulsed laser heating from a Ti:Sapphire oscillator with a fundamental output of 90 fs pulses at 80 MHz (12.5 ns between laser pulses) and 800 nm. The output of the laser system is split into two paths – a "pump" path and a time delayed "probe" path. The pump pulses are then modulated at with an electro-optic modulator to create a modulated heating of 11.39 MHz in the ceMoS$_2$ sheets. Before focusing the pump-pulses down into the MoS$_2$ suspension, the pulses are passed through a BiBO crystal to frequency double the heating event to 400 nm. Then, these 400 nm pump pulses are passed through a 5X objective to creating the modulated heating event in the ceMoS$_2$ from a Gaussian pump spot with a 1/e^2 radii of 25 μm. The pump pulse are time delayed via a mechanical delay stage and focused collinearly via the same 5X objective to probe the middle of the pump heating event with a 1/e^2 radii of 6 μm. The transmitted probe beam through the ceMoS$_2$/water suspension is focused into a photodiode, the output voltage of which is filtered by a lock in amplifier to monitor the change in voltage (transmission) at the modulation frequency of the pump pulses as a function of pump-probe delay time. This transient transmission, which is directly related to the transient absorption, is related to the temperature change from the MoS$_2$ to the water over ~1 ns of delay time. As the ceMoS$_2$ sheets are essentially monolayers, the heat transfer can be assumed as purely one dimensional from the planar, absorbing surface of the nanosheets. We therefore model the heat transfer with a one-dimensional thermal model described elsewhere;[2] this particular model is based on the solution to the one-dimensional heat equation in the frequency domain for a two layer system: an instantaneously thermalized ceMoS$_2$ sheet and a semi-infinite water heat sink (this semi-infinite assumption is valid due to the modulation frequency of the pump pulses and pump-probe time delay in the experiments[3]). The temperature of the absorbing MoS$_2$ nanosheets are then related to the output of the lock-in amplifier through the lock-in transfer function. For these data, we monitor in-phase voltage of the lock-in amplifier, which is related to the temperature change via[4]

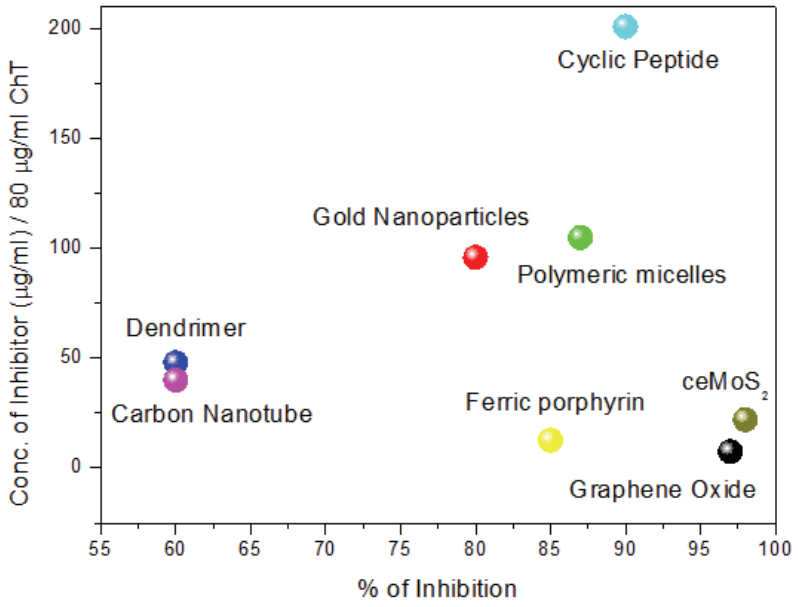
$$V_{in}\left(f_{pump}\right) = A\sum_{M=-\infty}^{\infty}\left(\theta\left(f_{pump}+Mf\right)+\theta\left(f_{pump}-Mf\right)\right)\exp\left[iMf\tau\right], \quad (1)$$

where θ is the temperature rise in the MoS$_2$ calculated with the one-dimensional thermal model, f_{pump} is the modulation frequency of the pump beams via the electro-optic modulator, f is the Ti:Sapphire repetition rate (80 MHz), τ is the pump-probe delay time, and A is a constant. We fit Eq. (1) to our data by scaling A to the data at ~50 ps of pump probe delay time, and then iterating the thermal boundary conductance between the MoS$_2$ and water to achieve agreement between the model and data over ~1.5 ns of pump-probe delay time. For our thermal model, we assume literature values for the heat capacity and thermal conductivity of the water;[5] we are completely insensitive to the thermal conductivity of the MoS$_2$ sheets since they are so thin that the temperature is instantaneous homogeneous. We take the heat capacity of MoS$_2$ as 1.81 MJ m^{-3} K^{-1}. (ref: Mclaren, R. C., "thermal conductivity anisotropy of molybdenum disulfide thin films", Masters Thesis, University of Illinois at Urbana-Champaign, 2009)

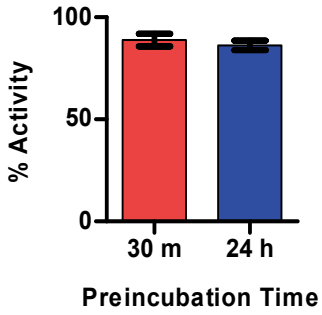
Enzymatic Activity Assay: All experiments were performed in independent triplicates or better. 20 μg/ml of α-chymotrypsin (ChT) was incubated with varying concentrations of ceMoS$_2$ in 5 mM sodium phosphate buffer (pH 7.4) for 30 min, at a total volume of 184 μl/well. SPNA substrate (16 μl) was then added to reach a final SPNA concentration of 2 mM. The total volume is 200 μl/well. Enzymatic activity

was monitored every by measuring p-nitroaniline formation (λ = 405 nm) using a microplate reader (Synergy Mx, Bio-Tek Instruments, Winooski, VT). Control experiment showed that the auto-hydrolysis of SPNA was negligible. Activity rates are reported as change over 60 min time period.

Enzymatic Loading comparison: Because ChT is a well characterized enzyme, its inhibition by colloids has often been used to indicate protein loading capacities.[6] A comparison of ceMoS$_2$'s loading capacity versus previous published results can thus be adapted. It can be seen that ceMoS$_2$ compares favorably to most nanomaterials, and is the only material to achieve comparable inhibition to Graphene Oxide. Figure is adapted from reference [6].



ChT Activity restoration: 40 µg/ml ChT was incubated with 11 ppm of ceMoS$_2$. NaCl was added either before the addition of ceMoS$_2$ or after the 30 min incubation. An additional experiment extending the incubation time to 24 hr was also performed (see below). The NaCl concentration was varied from 0 to 1000 mM in 5 mM sodium phosphate buffer (pH 7.4), reaching a final concentration of 184 µL. SPNA stock solution was then added (16 µL) to reach a final SPNA concentration of 2 mM. All activity was normalized by measuring the absorbance of control experiment. The assays were performed in triplicates, and the averages are reported at 60 min time period.



Circular Dichroism (CD) Spectra: Far-UV CD spectra of ChT (20 μg/ml) were measured on a JASCO Circular Dichroism Spectrophotometer J-815 with quartz cuvettes of 1 mm path length at 25 °C. The spectra were recorded from 190 to 250 nm. For the GO-ChT complex the concentration of GO is 11 ppm. The CD spectra of GO and buffer were subtracted from complex spectra to eliminate any background effects.

Fluorescence Spectroscopy: For the denaturation study, fluorescence spectra were measured on a ISS PC1 photon counting steady state spectrofluorimeter at room temperature (ca. 20 °C). The samples were excitated at 295 nm, and the emission spectra were recorded from 300 to 450 nm. Sample concentrations are the same as the CD study.

Cell Viability Assay: All products were purchased from Invitrogen/Gibco. Hela cells were grown in Minimum Essential Medium (MEM) + 10% Fetal Bovine Serum (FBS) with non-essential amino acid and sodium pyruvate supplements ("media"). After trypsinizing (TrypLE), cells were collected at 100,000/ml concentration in media, and then subjected to various treatments described in the main text. After treatments, cells were washed thrice using PBS and centrifugation (200 rcf, 2 minutes). Cells were then plated in 96 well plates at concentrations of 10,000/well. Media was added to reach a final volume of 200 μl/well. Surplus samples were plated in 24 well plates for optical imaging (50,000/well, diluted to 1 ml). After 24 hr incubation, cell viabilities in the 96 well plates were measured using the CellTiter 96 Assay (CT96A). To each well, 20 ul of CellTiter 96 Assay solution was added. Initial absorbance at λ = 495 nm is measured, and cell viabilities were monitored by changes in absorbance at λ = 495 nm. Viability data is normalized to control specimens. Experiments were performed in triplicates.

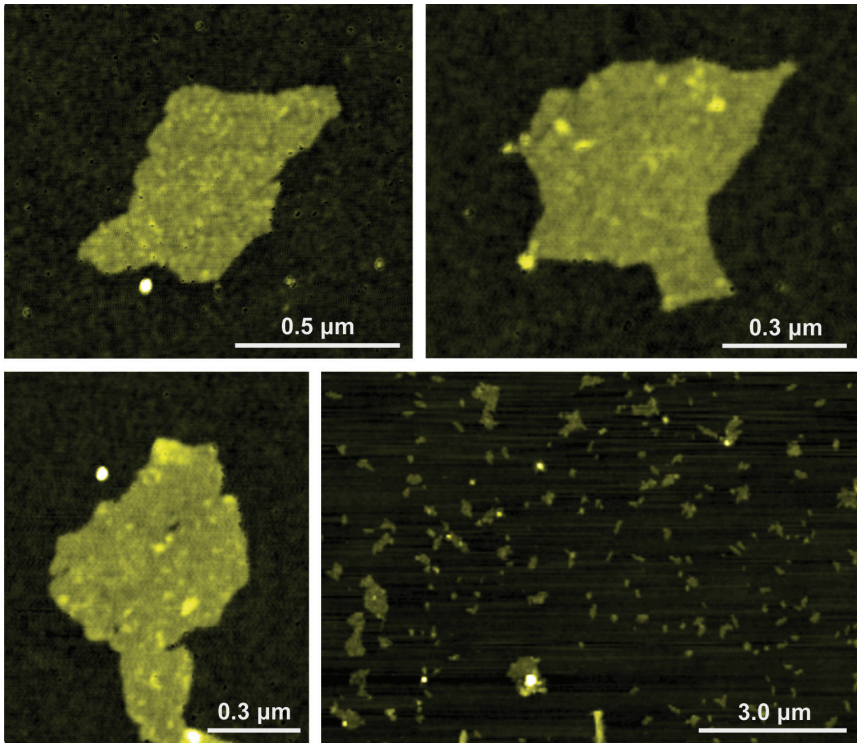


Figure S1. Representative AFM images and a large area AFM scan of individual ceMoS_2 monolayer sheets (bottom right).

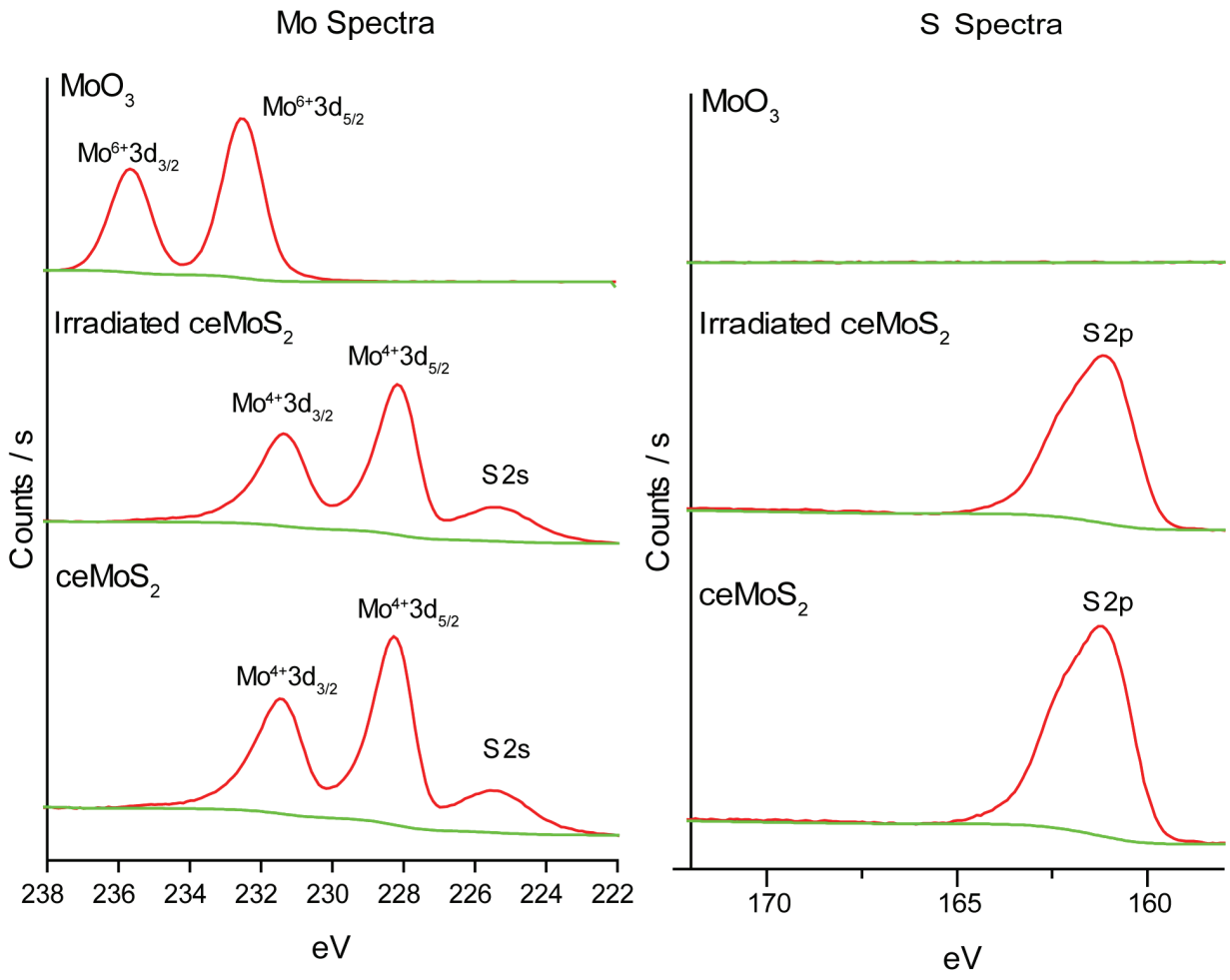


Figure S2. XPS spectra of MoO_3, irradiated $ceMoS_2$ and $ceMoS_2$. The absence of Mo^{6+} peaks at ~232 and 236 eV in $ceMoS_2$'s spectra indicate no significant oxidation before and after NIR irradiation (λ = 800 nm, 15 m). Similarly, the lack of a prominent peak at 168-170 eV for S spectra indicates no significant oxidation for S before or after irradiation. The spectra of MoO_3 is provided as a reference.

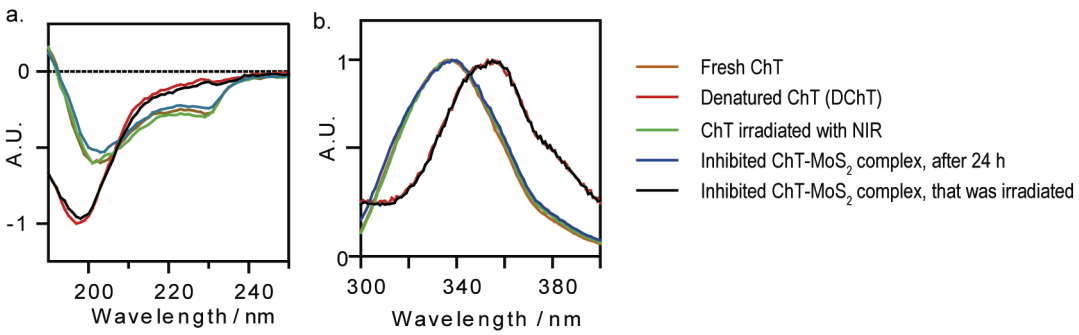


Figure S3. (a) CD and (b) fluorescent spectra of ChT after various treatments. Spectra of denatured Cht (DChT) obtained by heating solution to 50 ^{o}C via conventional oven is provided as a control reference. Free ChT irradiated with NIR (λ = 800 nm) in absence of MoS2 complexation is also provided as a reference.

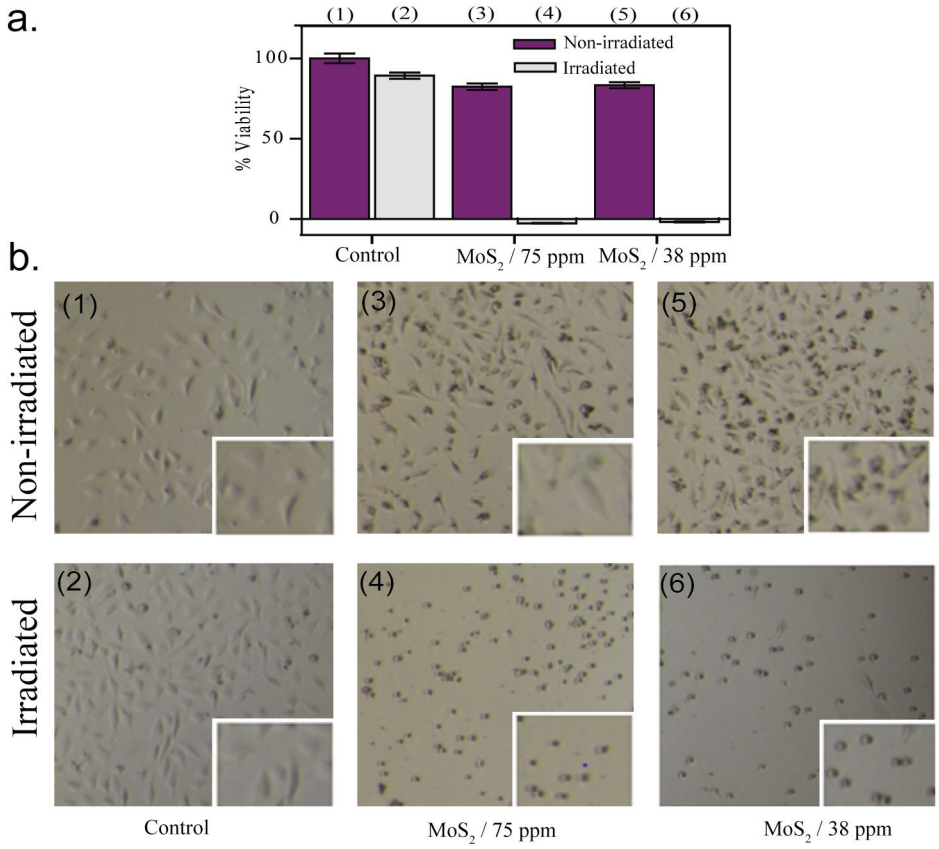


Figure S4. (a) Hela cell viabilities after various treatments. (b) Corresponding optical micrographs of cells after treatment.

References

[1] P. Joensen, R. F. Frindt, S. R. Morrison, *Mater. Res. Bull.* **1986**, *21*, 457-461.

[2] P. E. Hopkins, J. R. Serrano, L. M. Phinney, S. P. Kearney, T. W. Grasser, C. T. Harris, *Journal of Heat Transfer* **2010**, *132*, 081302.

[3] P. E. Hopkins, J. R. Serrano, L. M. Phinney, *International Journal of Thermophysics* **2010**, *31*, 2380-2393.

[4] O. M. Wilson, X. Hu, D. G. Cahill, P. V. Braun, *Physical Review B* **2002**, *66*, 224301.

[5] D. R. Lide, *CRC Handbook for Chemistry and Physics*, 89th ed., Internet Version, CRC Press/Taylor and Francis, Boca Raton, FL, **2008**.

[6] M. De, S. S. Chou, V. P. Dravid, *J. Am. Chem. Soc.* **2011**, *133*, 17524-17527.