Electronic Supplementary Material (ESI) for Journal of Materials Chemistry B.
This journal is © The Royal Society of Chemistry 2019

Combinational photochemotherapy of organoplatinum(II) metallacage based nanoparticles on liver cancer stem cells

Beng Yang[1], Hua Liu[1], Huang Yang[4], Wei Chen[2], Jingban Wu[1], Xiaode Feng[1], Rongliang Tong[1], Hanxi Yu[1], Yunhao Chen[1], Zhen Lv[1], Weijian Sun[3], Bin He[1], Jian Wu[1], Guocan Yu[5*], Zhengwei Mao[4*], Shusen Zheng[1*]

[1] Division of Hepatobiliary and Pancreatic Surgery, Department of Surgery, First Affiliated Hospital, School of Medicine, Zhejiang University. NHFPC Key Laboratory of Combined Multi-organ Transplantation. Key Laboratory of the diagnosis and treatment of organ Transplantation, CAMS. Key Laboratory of Organ Transplantation, Collaborative Innovation Centre for Diagnosis Treatment of Infectious Diseases, Zhejiang Province, Hangzhou, 310003, China.

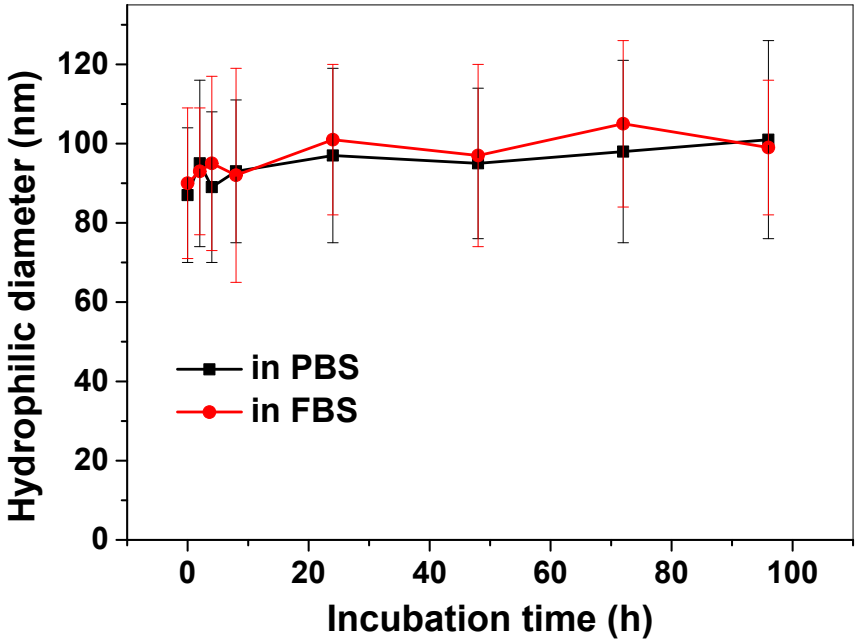
[2] Department of general medicine, Second Affiliated Hospital of Zhejiang Chinese medical university, Hangzhou, 310003, China.

[3] Department of Surgery, The Second Affiliated Hospital and Yuying Children's Hospital of Wenzhou Medical University, School of Medicine, Wenzhou 325027, China.

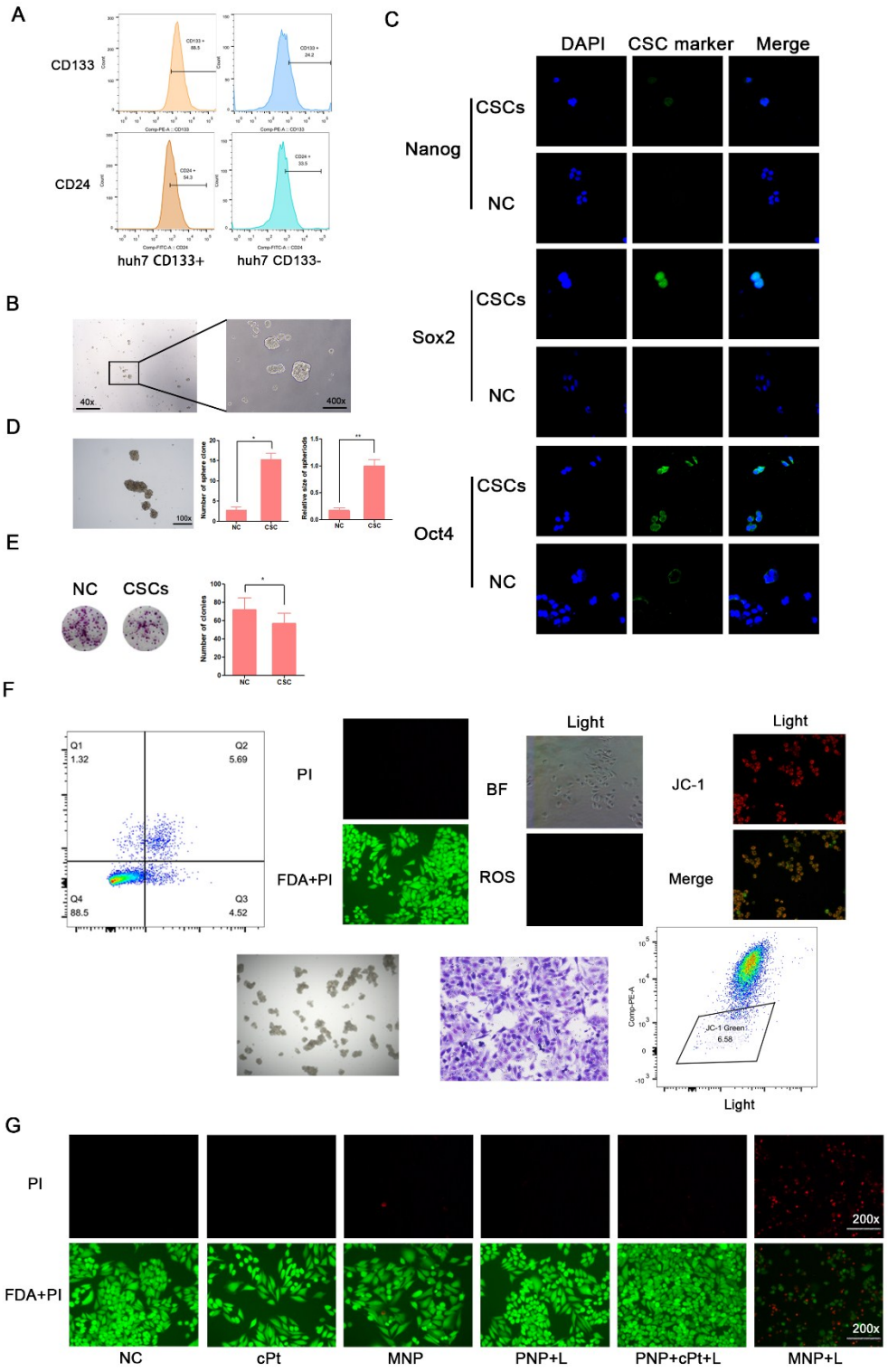
[4] MOE Key Laboratory of Macromolecular Synthesis and Functionalization, Department of Polymer Science and Engineering, Zhejiang University, Hangzhou 310027, China

[5] Laboratory of Molecular Imaging and Nanomedicine, National Institute of Biomedical Imaging and Bioengineering, National Institutes of Health, Bethesda, Maryland 20892, United States.

[*] Corresponding authors: shusenzheng@zju.edu.cn (S. Z.), zwmao@zju.edu.cn (Z. M.) and guocan.yu@nih.gov (G. Y.).

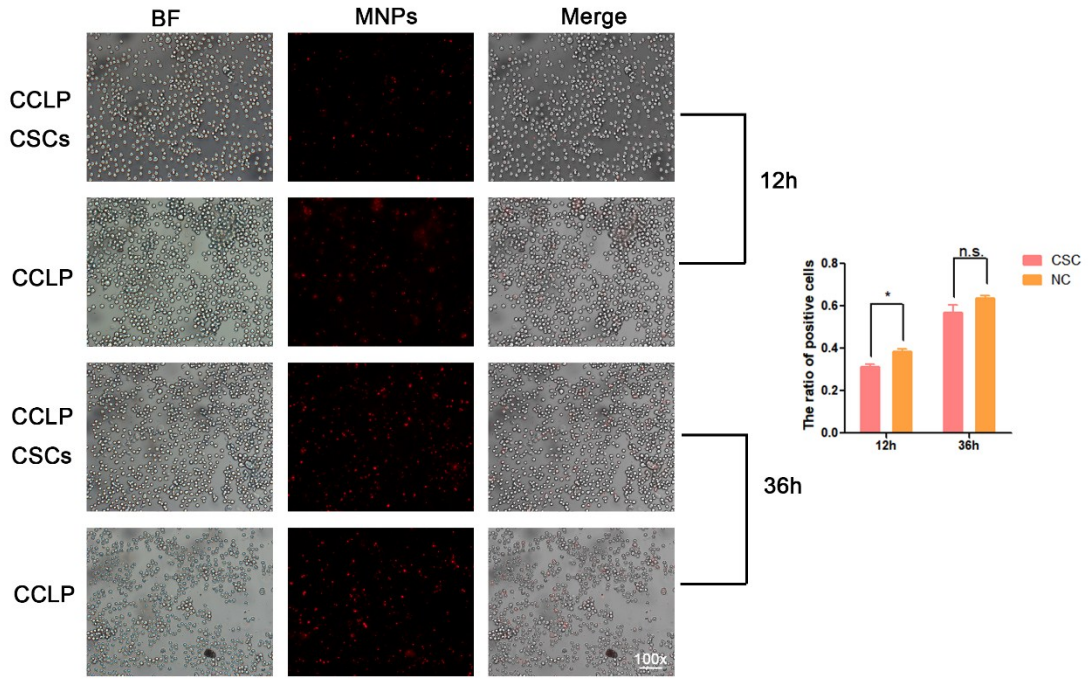


Supplementary Figure 1. The average hydrophilic diameters of **MNPs** in PBS and FBS after different time periods of incubation.

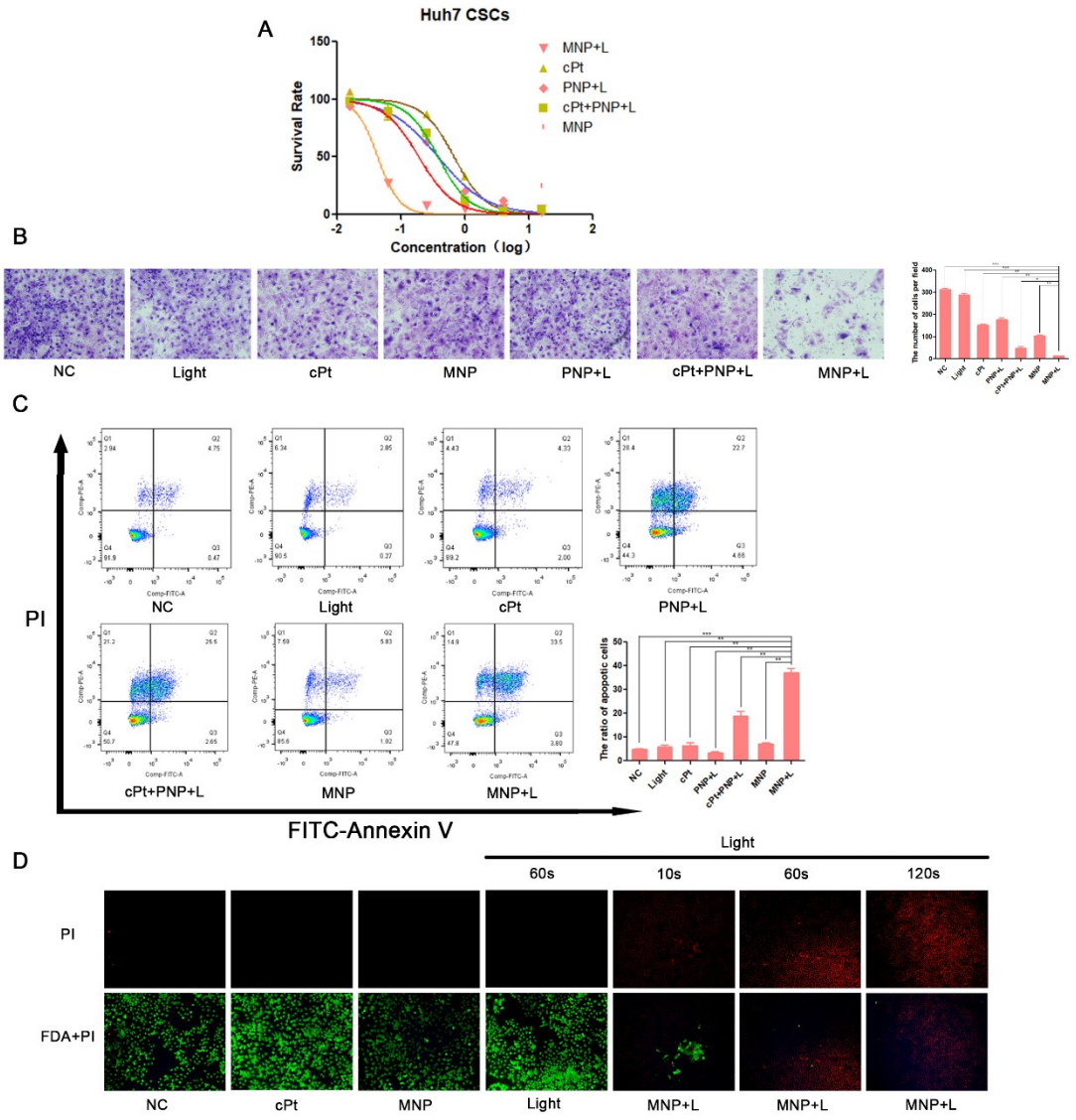


Supplementary Figure 2. (A) The ratio of CD133+ subset and CD44+ subset of CD133- cells and CD133+ Huh7 CSCs after MACS sorting. (B) The image of Huh7 CSCs after MACS sorting and followed 3D non-adherent culture with a magnification of 100 times. (C) Fluorescence imaging of Huh7 cells and Huh7 CSCs stained with Nanog, Sox2, Oct4 (green), respectively, to show the expression of stem cell markers.

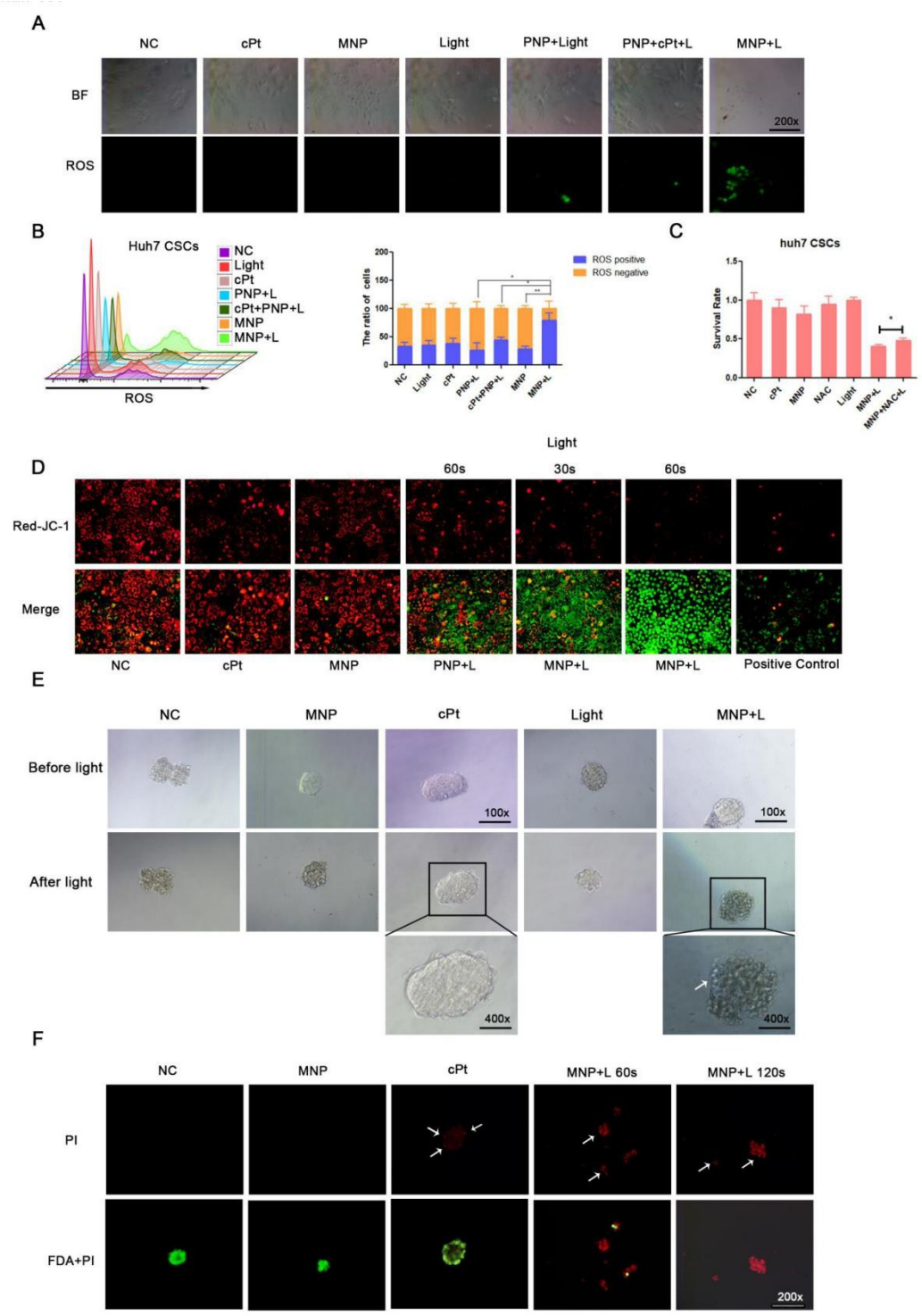
The cell nuclei were stained by DAPI (blue). The images were magnified 400 times. (D) Comparison of spheroid formation and (E) clone formation between Huh7 cells and Huh7 CSCs. * $p<0.05$. The image of tumor sphere was magnified 100 times. (F) The apoptosis, migration, living status, ROS production and mitochondria injury of CCLP-1 CSCs received only light irradiation (0.5 W/cm^2, 1 min). (G) Fluorescence images of CCLP-1 CSCs received different treatment. Dead cells were stained with red color (PI) while alive cells were stained with green color (fluorescein diacetate, FDA).



Supplementary Figure 3. The cell uptake investigation of MNPs in CCLP and CCLP CSCs. Representative photograph of positive cells in CCLP CSCs and CCLP NC groups after incubation with MNPs for 12 and 36 hours. (BF = bright field, the images and Fluorescence images were magnified 100 times).

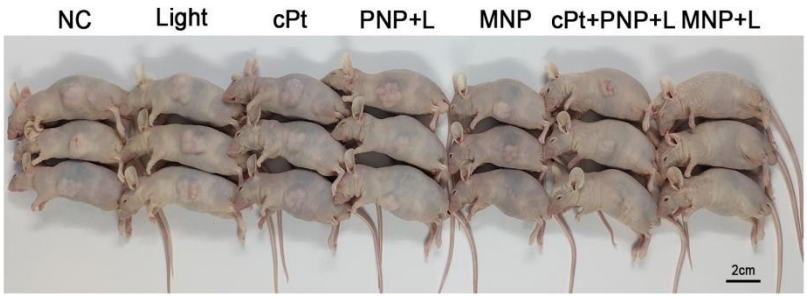


Supplementary Figure 4. (A) Cytotoxicity comparison of Huh7 CSCs incubated with PNP, cPt or MNP with or without irradiation of 638 nm light (0.5 W/cm^2, 1 min), respectively. (B) The migration and (C) apoptosis of Huh7 CSCs treated with cPt, PNP, PNP + cPt, MNP with or without irradiation of 638 nm light (0.5 W/cm^2, 1 min), respectively. The images of migration were magnified 100 times. * $p<0.05$, ** $p<0.01$, *** $p<0.001$. (D) Fluorescence image of Huh7 CSCs treated with cPt, light only or MNP with or without irradiation of 638 nm light (0.5 W/cm^2, for 10 s, 60 s and 120 s, respectively). Dead cells and alive cells were stained in red and green, respectively.



Supplementary Figure 5. (A) ROS production caused by MNP in Huh7 CSCs. Fluorescence images (Green: DCF fluorescence; blue: DAPI fluorescence) and (B) quantitative determination via FACS in the Huh7 CSCs received different treatment. The fluorescence images were magnified 200 times. * $p<0.05$, ** $p<0.01$. (C) The survival rate of Huh7 CSCs received different treatment. (D) Detection of mitochondria

injury caused by MNP in Huh7 CSCs. Fluorescence images reflected compact of mitochondria membrane of the Huh7 CSCs received different treatment. The images were magnified 200 times. (E) The images and (F) Fluorescence images of Huh7 CSCs spheroids treated with MNP only, cPt, light only, and MNP with irradiation of 638 nm light (0.5 W/cm^2, 1 min). The white arrows denote the dead cancer cells in tumor sphenoid. The images above and Fluorescence images were magnified 200 times while images below were magnified 400 times.



Supplementary Figure 6. Representative photograph of mice bearing the subcutaneous tumors formed by CCLP-1 CSCs received various therapeutic treatment. Scale bar is 2 cm.