ACS MEASUREMENT SCIENCE Au
AN OPEN ACCESS JOURNAL OF THE AMERICAN CHEMICAL SOCIETY

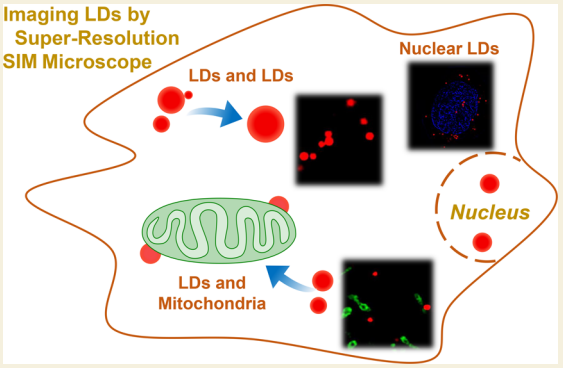
pubs.acs.org/measureau
Article

Super-Resolution Microscopic Imaging of Lipid Droplets in Living Cells via Carbonized Polymer Dot-Based Polarity-Responsive Nanoprobe

Zepeng Huo, Zitong Yu, Weiqing Xu, and Shuping Xu*

Cite This: ACS Meas. Sci. Au 2024, 4, 593−598
Read Online

ABSTRACT: Lipid droplets (LDs) are dynamic subcellular organelles that participate in various physiological processes, and their abnormality can also lead to various diseases. Tracing the dynamics of LDs in living cells will be valuable for understanding cell physiological states. Here, we employed a structured light illumination super-resolution imaging assisted with a carbonized polymer dot (CPD)-based fluorescence nanoprobe to track the travel paths of LDs and other organelles. The CPDs we developed are highly biocompatible with living cells and exhibit a highly sensitive response to solvent polarity, allowing for high specificity in staining LDs in living cells. Aided by these nanoprobes, we successfully observed many real-time LD-involved dynamics in living cells, such as intracellular LD interactions, communications with other organelles, and dynamic behaviors under external stimuli (oxidative stress inducer). These studies deepen our comprehension of the physiological role of LDs and drive the advancement of super-resolution fluorescent probes.



KEYWORDS: *carbon dots, lipid droplets, super-resolution imaging, nanoprobe, organelle interaction*

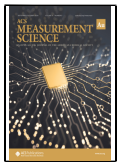
1. INTRODUCTION

Lipid droplets (LDs) are important organelles composed of neutral lipid cores inside and covered by single-layer phospholipids outside, responsible for energy storage and supply in eukaryotic cells, and play an essential role in maintaining cell homeostasis.[1,2] LDs play an important role in regulating cellular homeostasis, including lipid metabolism, protein degradation, and signal transduction, all of which require the participation of LDs.[1] Abnormality of LDs can lead to a variety of diseases including obesity, diabetes, fatty liver, and so forth.[3,4] Therefore, it is important to have a deep understanding of the function of LDs. In recent years, many studies have shown that LDs and other organelle (such as mitochondria and nuclei) regulate intracellular signal transduction and metabolic homeostasis through close interaction.[5−7] Previous evidence suggests the transport of substances such as fatty acids between LDs and mitochondria, indicating close communication and exchange between the two.[8] Therefore, understanding the interaction process between LDs and mitochondria is vital for exploring cellular homeostasis.

Fluorescence imaging is a powerful tool for studying the interaction between subcellular organelles in detail.[9,10] However, due to the diffraction limit of light, the traditional fluorescence microscope cannot obtain detailed structural information on subcellular organelles. Super-resolution imaging technology makes it possible to visualize nanoscale organelle interactions in living cells.[11] Many commercial organic fluorescence dyes are prone to photobleaching or instability in super-resolution imaging mode, which is inapplicable for long-term imaging. Thus, researchers have devoted their efforts to developing new super-resolution fluorescent probes, which will be valuable in exploring the biological process of LDs and visualizing the interaction mode between LDs and other organelle.

Carbonized polymer dots (CPDs), as a new type of carbon dots (CDs), are widely used in the field of biological imaging due to their advantages of good stability and biocompatibility.[12,13] At present, there are also many reports on the CDs that can be applied as fluorescence sensing probes for various imaging techniques, such as multiphoton fluorescence imaging, super-resolution imaging, near-infrared imaging, and photoacoustic imaging.[14−17] Wang et al. developed a carbon dot probe that can be used in the single-molecule localization

Received: July 29, 2024
Revised: August 23, 2024
Accepted: August 30, 2024
Published: September 6, 2024



https://doi.org/10.1021/acsmeasuresciau.4c00049
ACS Meas. Sci. Au 2024, 4, 593−598

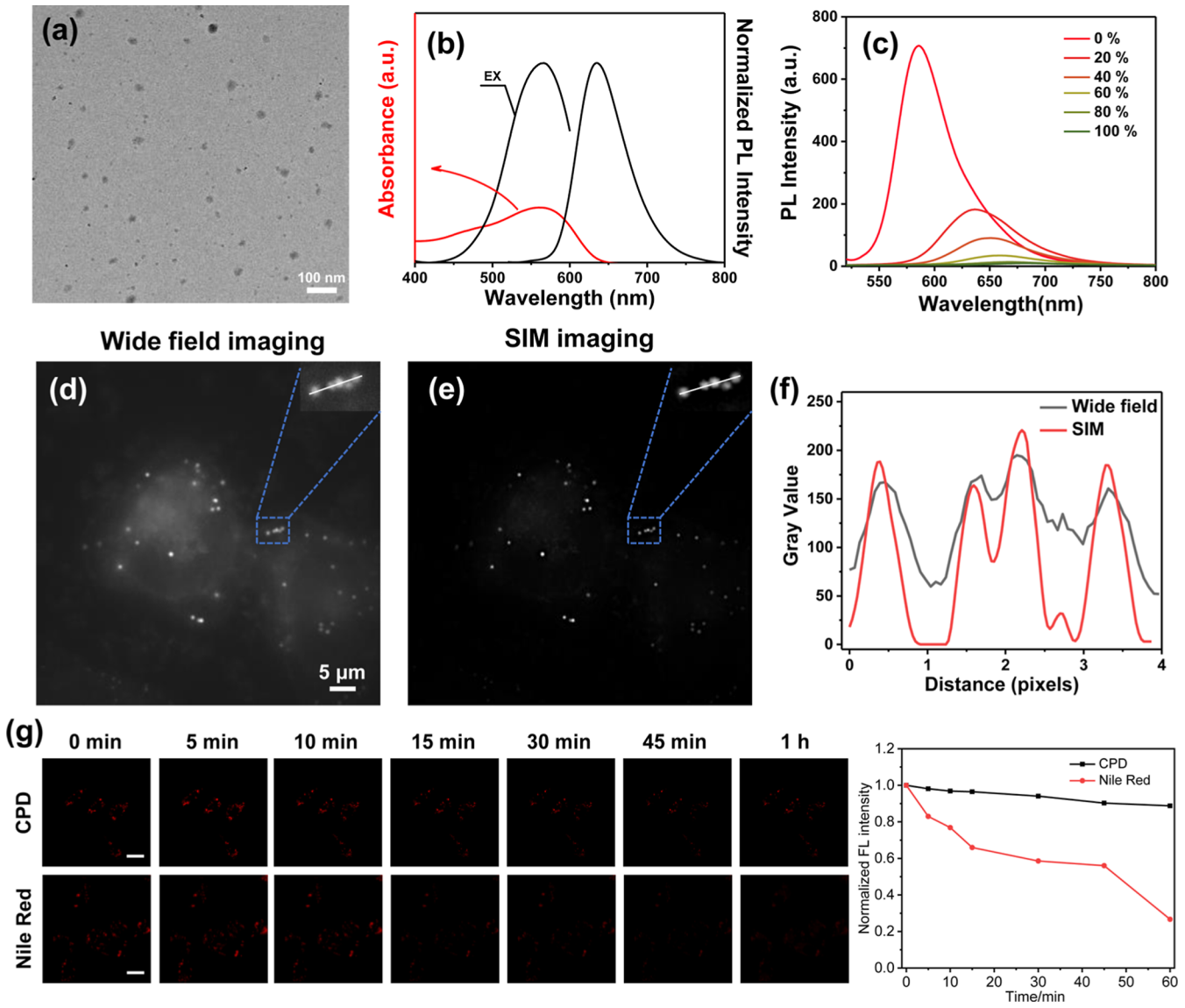


Figure 1. (a) TEM image of the CPDs. (b) UV−vis absorption spectra (red), photoluminescence (PL) excitation spectra (noted as EX), and emission spectra of the CPDs (in ethanol). (c) PL emission spectra of the CPDs in 1,4-dioxane with different water contents. CPD is used for wide-field imaging (d) and super-resolution SIM imaging (e). (f) Plot profile analysis along the white lines in (d) and (e). (g) The photobleaching experiment of CPDs compared to Nile Red for LD staining (excitation wavelength of 561 nm). The scale bars are 10 μm.

microscopy technique and applied it to imaging G protein-coupled receptors on the cell membrane, revealing the distribution pattern of receptors regulated by ligands.[18] Li et al. designed and synthesized a new type of nuclear-targeted CPD. This carbon dot can be used for stimulated emission depletion (STED) microscopic imaging. Their probe has good biocompatibility and stability and shows great potential in the dynamic research of nucleic acid function.[19] The above examples demonstrate a broader application prospect of carbon dots in super-resolution imaging. However, most developed CPD-based imaging probes are water-soluble nanoprobes, which are insensitive to lipids and endoplasmic reticulum. Therefore, it is necessary to develop novel CPD fluorescent probes for targeted imaging of intracellular lipid droplets.

This study developed an LD-targeted CPD-based nanoprobe to track the interaction mode between LDs and LDs with other organelle under SIM imaging. Due to its excellent targeting, good cell permeability, and high signal-to-background ratio, CPD has been successfully used to detect the morphological changes and dynamic processes of LDs in living cells. This reveals the contact sites between LDs and mitochondria. The changes of organelle under oxidative stress conditions in cells were clarified by exploring the dynamic changes of LDs on a nanoscale in living cells. In summary, CPD can be used as a nanoprobe for long-term tracking of LDs' dynamics under SIM, which helps to reveal the cellular biological effects of LDs.

2. EXPERIMENTAL SECTION

2.1. Synthesis of Nanoprobe

CPD was synthesized by ultrasonic treatment of 40 mg *N*,*N*-diethyl-*p*-phenylenediamine, and 10 mL ethanol and then by solvothermal reaction at 200 °C for 12 h in a poly(tetrafluoroethylene) Teflon autoclave. It was purified using dichloromethane and methanol as eluents.

https://doi.org/10.1021/acsmeasuresciau.4c00049
ACS Meas. Sci. Au 2024, 4, 593−598

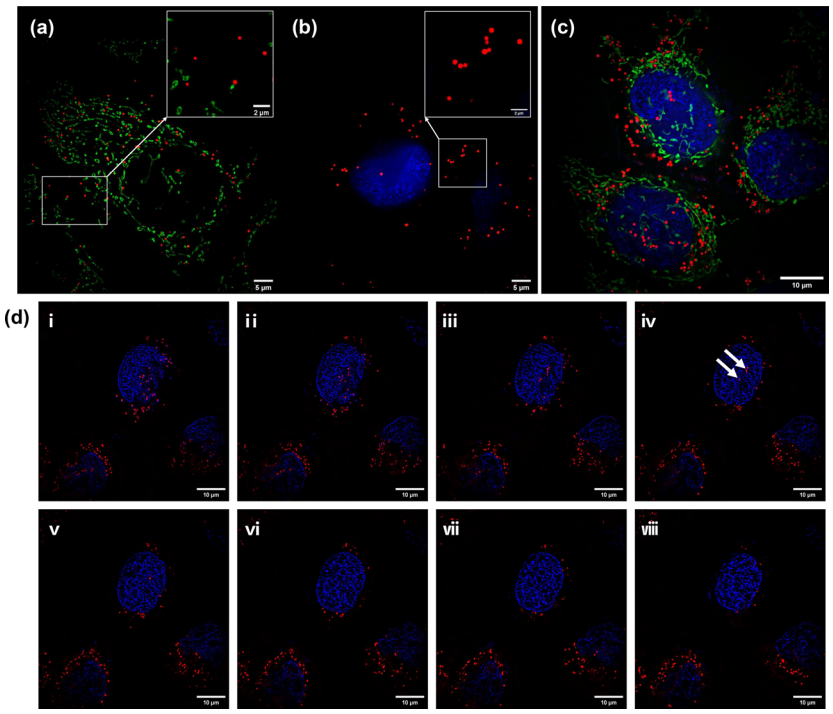


Figure 2. Super-resolution and partially enlarged images of HepG2 cells costained with CPDs and Mito-Tracker Green (a) or Hoechst 33342 (b). (c) SIM images of HepG2 cells costained with Hoechst 33342, Mito-Tracker Green, and CPDs. (d) Depth-dependent images of HepG2 cells stained with CPD (i–viii). The white arrow in iv points toward the nuclear LDs.

2.2. Monitoring the Dynamics of LDs by SIM Imaging

HepG2 cells were inoculated into glass bottom culture dishes and incubated in a humid atmosphere of 37 °C, 5% CO_2, with DMEM and 10% FBS for 24 h. Then, we removed DMEM and used Hoechst 33342 (10 μg/mL, 5 min), Mito-Tracker Green (5 μM. 10 min), and CPD (50 μg/mL, 5 min) to stain the cells in order, followed by washing the cells three times with PBS (pH = 7.4) to remove the remaining dye. Finally, the cells seeded in glass bottom culture dishes and stained will be used for super-resolution SIM imaging.

The super-resolution SIM images were recorded using the HIS-SIM intelligent ultrasensitive super-resolution microscope (Guangzhou Chaoshiji Biotechnology Co., Ltd., China). Among them, the imaging objective uses a 100× oil immersion objective, and the excitation wavelengths of Hoechst 33342, Mito-Tracker Green, and CPD are 405, 488, and 561 nm, respectively.

2.3. Photostability Experiment of CPD in Cells

HeLa cells were seeded in the glass-bottom culture dishes and cultured in DMEM with 10% FBS at 37 °C in a humidified atmosphere of 5% CO_2 for 24 h. Then, we removed DMEM and stained the cells with Nile Red or CPD, respectively. After staining, the cells were washed three times with PBS (pH = 7.4) to remove excess dye. Afterward, the cultured cells were placed on a confocal microscope for imaging (excitation wavelength of 561 nm). Continuous scanning mode is used for imaging, and an image is collected at regular intervals. Then, ImageJ software is used to analyze the fluorescence intensity and obtain numerical values for subsequent analysis.

3. RESULTS AND DISCUSSION

3.1. CPD Design and Characterizations

LD is a complex organelle used to store neutral lipids. Considering the chemical characteristics of LD, appropriate lipophilicity is extremely important for the LD-targeted probe design. Considering the hydrophobic structure provided by the *N*,*N*-diethyl functional groups in the Nile Red structure, we selected *N*,*N*-diethyl-phenylenediamine as the precursor for reaction synthesis through a reverse reaction synthesis strategy. The precursor is constantly dehydrated and carbonized in the reaction process. Solvent participation in the reaction also helps it form a particular D-π-A structure that is environmentally sensitive and can realize polar sensing of microenvironments. The morphology of our prepared CPD was characterized by TEM, and the TEM result indicates that CPD is well dispersed and has a uniform size (Figure 1a).[17] The UV–vis absorption and emission spectra of CPD are shown in Figure 1b. CPD has an absorption peak at 566 nm and a fluorescence peak at 631 nm (in ethanol). Its emission is weak in pure water, but as the polarity of the solvent decreases, its fluorescence gradually increases. As shown in Figure 1c, with the increase of 1,4-dioxane content, the emission intensity of CPD gradually increases. Due to intramolecular charge transfer (ICT), there is a significant blue shift in the maximum emission from 684 to 584 nm.[20] The high lipophilicity of CPD and strong fluorescence emission in low polar solvents demonstrate that it is available for LD imaging.

Before conducting imaging experiments, we tested the cytotoxicity of CPD. The results shown in Figure S1 indicate that when cells were exposed to various concentrations of CPD ranging from 0 to 100 μg/mL, there was no significant effect on cell survival compared to the untreated control group. This suggests good biocompatibility and low cytotoxicity, indicating their suitability for biological systems.

As organelles that maintain lipid and energy homeostasis, many functions of LDs, such as antibacterial, are closely related to the microstructure of LDs and show the dynamic changes of LDs. The life cycle of LDs begins after their formation, including maturation, movement, and turnover, which are also completed through the fusion of LDs and other organelles.[6] To explore spatial resolution, wide-field imaging and SIM imaging

https://doi.org/10.1021/acsmeasuresciau.4c00049
ACS Meas. Sci. Au 2024, 4, 593–598

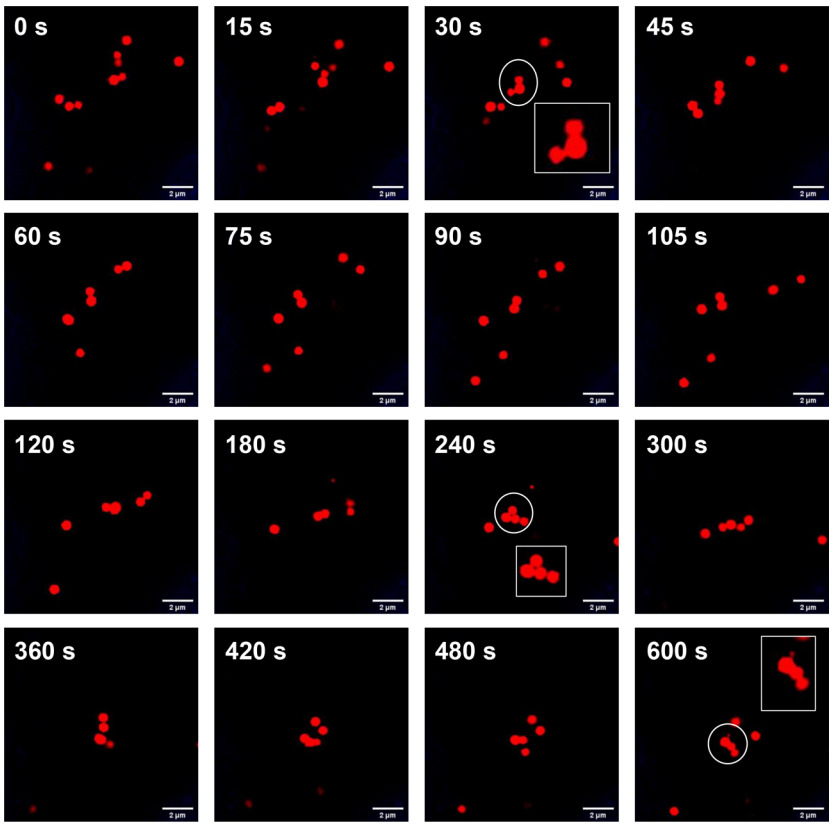


Figure 3. Super-resolution SIM imaging of the LDs contact in the living HepG2 cells. The circles in the figure indicate several typical contact modes of LDs, and the boxes are enlarged images of the circles.

were applied to observe two adjacent CPD-labeled particles. As expected, the SIM imaging results clearly distinguish two tiny droplets positioned close to each other (Figure 1d–f), indicating a higher spatial resolution beyond the wild-field imaging.

The CPDs exhibit higher photostability compared to the commercial dyes. As depicted in Figure 1g, we captured the long-period cell images under laser irradiation for 1 h. Our CPDs showed only 10% signal off in 1 h, whereas the Nile Red exhibited an 80% decrease in fluorescence intensity. Therefore, we can conclude that our developed CPDs demonstrate superior photostability and antiphotobleaching characteristics, making them suitable for imaging under intense laser irradiation and long-term imaging observation.[21]

3.2. LD-Specific Cellular Imaging

To confirm whether CPD targets LDs in living cells, we incubated HepG2 cells with CPDs and their SIM images were taken, as shown in Figure 2a–c, in which the round dotted organelles gave apparent fluorescence. To verify whether the CPD we developed is compatible with commercial nuclear and mitochondrial dyes, we also collected cell imaging results of three dyes costaining. As shown in Figure 2c, three dyes can well stain a variety of organelles in cells without crosstalk, which confirms that CPD can match the common organelle dyes.

The use of 3D-SIM data also shows that these organelles have differentiated distribution with the change of imaging depth and time, which is consistent with the characteristics of LD [Figure 2d(i–viii)]. To evaluate the SIM imaging characteristics of CPD in different cell lines, LDs in HeLa cells before and after oleic acid stimulation were also stained with CPD. It can be observed that there are more LDs in the cells after stimulation with oleic acid (Figure S2). More interestingly, we also found that some LDs in the nucleus were illuminated Figure 2d(iv) (marked by arrows).[22] 3D-SIM data also indicates that CPD can stain nuclear LDs in cells without affecting the normal movement process of LDs. Therefore, we conclude that CPD is a low toxicity and good cell permeability LD-targeting probe.

3.3. Super-Resolution SIM Imaging of LDs

Super-resolution imaging of organelles has become a powerful method for deciphering physiological processes and molecular mechanisms. Considering this, we investigated the interaction between multiple LDs through SIM imaging. We selected a typical region in cell imaging for discussion. From Figure 3, it can be observed that the interaction process of LDs is very fast in a short period of time, indicating the presence of rapid energy exchange processes within cells. During this process, the fusion, division, and other processes between LDs can also be revealed. In addition, we also found that LDs form a filamentous state between LDs before fusion (30, 240 s), or the behavior of large LDs "spitting out" small LDs (600 s), which is similar to previous reports.[23−25] In order to better represent the distribution of intracellular LDs, several sets of photos were taken at different intervals in this experiment and listed in Figure 3. In short, the LDs within cells have different shapes, uneven distribution, and varying trajectories, which are erratic.

3.4. Contact Actions between LDs and Mitochondria

The interaction between organelles is very important for intracellular homeostasis, energy exchange, and material transport.[6] As a dynamic organelle, LDs' interactions with

https://doi.org/10.1021/acsmeasuresciau.4c00049
ACS Meas. Sci. Au 2024, 4, 593−598

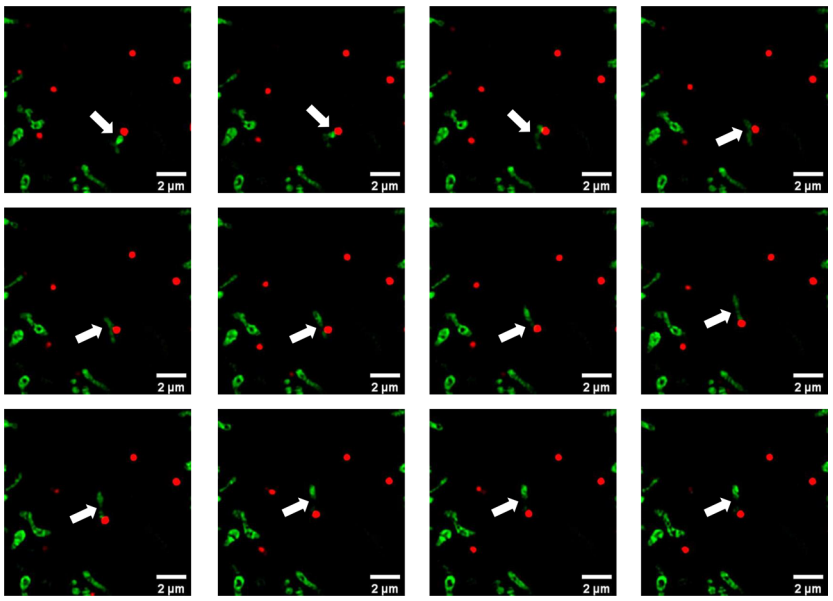


Figure 4. Super-resolution SIM imaging of the LDs and mitochondria in a living HepG2 cell.

other organelles are of great value for the study of intracellular signal transduction.[26] To deeper learn this, we collected the real-time interaction process between LDs and mitochondria, as shown in Figure 4, showing multiple interaction modes between them. It can be observed that mitochondria gradually stretch out, followed by the process of mitochondria dragging LDs "running". Considering the energy transmission function of LDs and the role of mitochondria in intracellular energy supply, we speculate that this phenomenon may be related to intracellular energy transmission. In addition, other regions within the cell also exhibit numerous interaction patterns, such as mitochondrial "encapsulation" of LDs. Reactive oxygen species stimulation can make LD lose vitality (Figure S3). It should be noted that the morphology of mitochondrial crista and the active movement process of mitochondria after CPD staining can be identified (Figure 4), which proves that CPD can realize the staining and tracking of multicellular organelles without affecting the normal physiological activity of cells, indicating the great application potential of CPD in the field of super-resolution SIM imaging.

4. CONCLUSIONS

This study developed a high-performance carbon-based LD-targeted nanoprobe through the design of reactive precursors, which is used to image the cell dynamics of LD and its interaction with mitochondria through SIM imaging. CPD has been successfully used to reveal the interaction mode between LDs and mitochondria at the subcellular level and to reveal the changes in the interaction mode between mitochondria and LDs induced by intracellular reactive oxygen species. Compared to traditional commercial dyes, our probe can also reveal LDs in the nucleus, becoming a powerful tool for exploring the function of nuclear LDs. This research helps to a deeper understanding of the biological functions of LDs and provides ideas for the future development of new super-resolution imaging probes.

The dynamic changes of LDs and their interactions with organelles revealed in this study provide a new perspective for a deeper understanding of the physiological functions of LDs. Future research should further explore the relationship between these findings and the occurrence and development of diseases. Further development based on this probe is expected to provide new diagnostic tools for clinical practice or develop treatment strategies for LD abnormalities.

■ ASSOCIATED CONTENT

Supporting Information

The Supporting Information is available free of charge at https://pubs.acs.org/doi/10.1021/acsmeasuresciau.4c00049.

Materials, instruments, details of cytotoxicity assay, Figures S1−S3 (PDF)

■ AUTHOR INFORMATION

Corresponding Author

Shuping Xu − *State Key Laboratory of Supramolecular Structure and Materials, College of Chemistry, Jilin University, Changchun 130012, P. R. China; Center for Supramolecular Chemical Biology, College of Chemistry and Institute of Theoretical Chemistry, College of Chemistry, Jilin University, Changchun 130012, P. R. China;* orcid.org/0000-0002-6216-6175; Email: xusp@jlu.edu.cn

Authors

Zepeng Huo − *State Key Laboratory of Supramolecular Structure and Materials, College of Chemistry, Jilin University, Changchun 130012, P. R. China*

Zitong Yu − *State Key Laboratory of Supramolecular Structure and Materials, College of Chemistry, Jilin University, Changchun 130012, P. R. China*

Weiqing Xu − *State Key Laboratory of Supramolecular Structure and Materials, College of Chemistry, Jilin University, Changchun 130012, P. R. China; Institute of Theoretical Chemistry, College of Chemistry, Jilin University, Changchun 130012, P. R. China;* orcid.org/0000-0002-1947-317X

Complete contact information is available at: https://pubs.acs.org/10.1021/acsmeasuresciau.4c00049

Notes

The authors declare no competing financial interest.

■ ACKNOWLEDGMENTS

We appreciate an Application Engineer, Zihan Wu from CSR Biotech (Guangzhou) Co., Ltd. China, for her support in imaging data processing. This work was supported by National Natural Science Foundation of China NSFC (grant nos. 21873039, 22173035 and 22373041), the Fundamental Research Funds for the Central Universities, Science and Technology Development Program Projects of Jilin Province (20220101046JC), and Innovative Research Project of State Key Laboratory of Supramolecular Structure and Materials.

■ REFERENCES

(1) Thiam, A. R.; Farese, R. V., Jr.; Walther, T. C. The Biophysics and Cell Biology of Lipid Droplets. *Nat. Rev. Mol. Cell Biol.* **2013**, *14* (12), 775−786.
(2) Walther, T. C.; Farese, R. V., Jr. Lipid Droplets and Cellular Lipid Metabolism. *Annu. Rev. Biochem.* **2012**, *81*, 687−714.
(3) Fan, H.; Tan, Y. Lipid Droplet-Mitochondria Contacts in Health and Disease. *Int. J. Mol. Sci.* **2024**, *25* (13), 6878.
(4) Zadoorian, A.; Du, X.; Yang, H. Lipid Droplet Biogenesis and Functions in Health and Disease. *Nat. Rev. Endocrinol.* **2023**, *19* (8), 443−459.
(5) Cohen, S.; Rambold, A. S.; Lippincott-Schwartz, J. Mitochondrial and Lipid Droplet Dynamics Regulate Intra- and Intercellular Fatty Acid Trafficking. *Mol. Cell. Oncol.* **2018**, *5* (5), No. e1043038.
(6) Dejgaard, S. Y.; Presley, J. F. Interactions of Lipid Droplets with the Intracellular Transport Machinery. *Int. J. Mol. Sci.* **2021**, *22* (5), 2776.
(7) Henne, M. And Three'sa Party: Lysosomes, Lipid Droplets, and the ER in Lipid Trafficking and Cell Homeostasis. *Curr. Opin. Cell Biol.* **2019**, *59*, 40−49.
(8) Enkler, L.; Spang, A. Functional Interplay of Lipid Droplets and Mitochondria. *FEBS Lett.* **2024**, *598* (10), 1235−1251.
(9) Fam, T. K.; Klymchenko, A. S.; Collot, M. Recent Advances in Fluorescent Probes for Lipid Droplets. *Mater.* **2018**, *11* (9), 1768.
(10) Yang, Z.; Xu, T.; Li, H.; She, M.; Chen, J.; Wang, Z.; Zhang, S.; Li, J. Zero-Dimensional Carbon Nanomaterials for Fluorescent Sensing and Imaging. *Chem. Rev.* **2023**, *123* (18), 11047−11136.
(11) Yang, Z.; Sharma, A.; Qi, J.; Peng, X.; Lee, D. Y.; Hu, R.; Lin, D.; Qu, J.; Kim, J. S. Super-Resolution Fluorescent Materials: An Insight into Design and Bioimaging Applications. *Chem. Soc. Rev.* **2016**, *45* (17), 4651−4667.
(12) Xia, C.; Zhu, S.; Feng, T.; Yang, M.; Yang, B. Evolution and Synthesis of Carbon Dots: From Carbon Dots to Carbonized Polymer Dots. *Adv. Sci.* **2019**, *6* (23), 1901316.
(13) Meng, W.; Wang, B.; Ai, L.; Song, H.; Lu, S. Engineering White Light-Emitting Diodes with High Color Rendering Index from Biomass Carbonized Polymer Dots. *J. Colloid Interface Sci.* **2021**, *598*, 274−282.
(14) Zhao, W. B.; Chen, D. D.; Liu, K. K.; Wang, Y.; Zhou, R.; Song, S. Y.; Li, F. K.; Sui, L. Z.; Lou, Q.; Hou, L.; Shan, C. X. Near-Infrared I/II Emission and Absorption Carbon Dots *via* Constructing Localized Excited/Charge Transfer State for Multiphoton Imaging and Photothermal Therapy. *Chem. Eng. J.* **2023**, *452*, 139231.
(15) Zhou, R.; Wang, C. G.; Lu, G. Y. Advances in Organic Fluorescent Probes for Super-Resolution Imaging of Cellular Lipid Droplets. *Chin. Opt.* **2022**, *15* (6), 1228−1242.
(16) Li, W.; Kaminski Schierle, G. S.; Lei, B.; Liu, Y.; Kaminski, C. F. Fluorescent Nanoparticles for Super-Resolution Imaging. *Chem. Rev.* **2022**, *122* (15), 12495−12543.
(17) Li, H.; Xu, T.; Zhang, Z.; Chen, J.; She, M.; Ji, Y.; Zheng, B.; Yang, Z.; Zhang, S.; Li, J. Photostable and Printable Fluorescence Carbon Quantum Dots for Advanced Message Encryption and Specific Reversible Multiple Sensing of $Cu^{2+}$and Cysteine. *Chem. Eng. J.* **2023**, *453*, 139722.
(18) Wang, Q.; Zhang, Q.; He, H.; Feng, Z.; Mao, J.; Hu, X.; Wei, X.; Bi, S.; Qin, G.; Wang, X.; Ge, B.; Yu, D.; Ren, H.; Huang, F. Carbon Dot Blinking Fingerprint Uncovers Native Membrane Receptor Organizations *via* Deep Learning. *Anal. Chem.* **2022**, *94* (9), 3914−3921.
(19) Li, H.; Ye, S.; Guo, J.; Wang, H.; Yan, W.; Song, J.; Qu, J. Biocompatible Carbon Dots with Low Saturation Intensity and High Photobleaching Resistance for STED Nanoscopy Imaging of the Nucleolus and Tunneling Nanotubes in Living Cells. *Nano Res.* **2019**, *12* (12), 3075−3084.
(20) Huo, Z.; Cao, X.; Sun, D.; Xu, W.; Yang, B.; Xu, S. Carbonized Polymer Dot Probe for Two-Photon Fluorescence Imaging of Lipid Droplets in Living Cells and Tissues. *ACS Sens.* **2023**, *8* (5), 1939−1949.
(21) Jing, Y.; Liu, G.; Zhang, C.; Yu, B.; Sun, J.; Lin, D.; Qu, J. Lipophilic Red-Emitting Carbon Dots for Detecting and Tracking Lipid Droplets in Live Cells. *ACS Appl. Bio Mater.* **2022**, *5* (3), 1187−1193.
(22) Barbosa, A. D.; Siniossoglou, S. New Kid on the Block: Lipid Droplets in the Nucleus. *FEBS J.* **2020**, *287* (22), 4838−4843.
(23) Liu, T.; Chen, J.; Dai, Y.; Wang, Z.; Chen, P.; Zhou, Y.; Wang, H.; Li, Y.; Mo, Z.; Yang, C.; Zhang, X.; Sun, B.; Yin, J.; Li, L.; Li, G.; Ma, J. Harnessing Polarity-Dependent Fluorescent Probe for Lipid Droplet Metabolism Dissection and in Vivo Nonalcoholic Fatty Liver Disease Diagnosis. *Sens. Actuators, B* **2024**, *399*, 134825.
(24) Hu, L.; Pan, J.; Zhang, C.; Yu, K.; Shen, S.; Wang, Y.; Shen, X.; Gu, X.; Han, J.; Wang, H. Polarity-Sensitive and Lipid Droplet-Specific Red Emission Fluorophore for Identifying Fatty Liver of Living Mice through in Vivo Imaging. *Biosens. Bioelectron.* **2022**, *216*, 114618.
(25) Liu, X.; Lu, X.; Zhu, T.; Du, W.; Yang, Z.; Cao, H.; Wang, S.; Tian, Y.; Zhang, Z.; Zhang, R.; De Souza, S. C.; Tian, X. Revealing Lipid Droplets Evolution at Nanoscale under Proteohormone Stimulation by a BODIPY-Hexylcarbazole Derivative. *Biosens. Bioelectron.* **2021**, *175*, 112871.
(26) Benador, I. Y.; Veliova, M.; Liesa, M.; Shirihai, O. S. Mitochondria Bound to Lipid Droplets: Where Mitochondrial Dynamics Regulate Lipid Storage and Utilization. *Cell Metab.* **2019**, *29* (4), 827−835.

https://doi.org/10.1021/acsmeasuresciau.4c00049
ACS Meas. Sci. Au 2024, 4, 593−598