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Introduction to special issue on single cell analysis

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Increasing evidence has shown that cell populations are not homogeneous, but rather heterogeneous, even within very small cell populations. Bulk measurements based on the homogenized cell population do not account for the critical changes occurring in individual cells and are sometimes misleading. Cellular heterogeneity characteristics may be the key to address previously unsolved questions in disease development and progression. Therefore, it is crucial to develop novel single cell analysis techniques, including spectroscopy, imaging, sensing, manipulating, and sorting. This special

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issue, including three review articles and five original research articles, highlights recent progress in the development of single-cell optical analysis techniques and their applications in biological discovery, disease diagnosis, and treatment.

A number of advanced optical imaging methods have been recently developed for single cell analysis. Stimulated Raman scattering (SRS) microscopy, an attractive imaging technique with advantages of high chemical selectivity, high spatial resolution, and high imaging speed, has become a powerful tool for biology and medicine. Jing Huang and Minbiao Ji reviewed the principles of SRS, discussed the technical developments and implementations of SRS microscopy, then highlighted and summarized its applications on biological cellular machinery, and finally shared visions of potential breakthroughs in the future.[1] Light field microscopy (LFM) is an elegant non-scanning imaging tool to achieve real-time three-dimensional (3D) volumetric imaging of single cells with a single shot. Beibei Gao *et al.* reviewed the principle and development of LFM, discussed the improved approaches based on hardware designs and 3D reconstruction algorithms, and presented the applications in single-cell imaging.[2] As an emerging single-cell imaging modality, microwave-induced thermoacoustic imaging (MTAI), with the assistance of functional nanoparticles, has shown broad prospects in biomedical and clinical applications. Xiaoyu Tang *et al.* reviewed the recent progress, challenges, and future directions of MTAI integrated with functional nanoparticles.[3] Besides imaging instrumentation, processing algorithm is also very important for single-cell analysis. Shutong Liu *et al.* proposed a new feature extraction method in fluorescence imaging of nucleus to achieve automatic and accurate identification of apoptosis with low cost in terms of time,[4] which may facilitate large-scale single-cell analysis.

The advanced single-cell optical methods have been increasingly applied for biological discovery and disease diagnosis. Exosome has been considered as a promising biomarker for diagnosis of early-stage cancer. Xiao Ma *et al.* achieved label-free breast cancer detection and classification by convolutional neural network-based exosomes surface-enhanced Raman scattering.[5] Actin cytoskeleton plays crucial roles in various cellular functions. Fulin Xing *et al.* reported the regulation of actin cytoskeleton via photolithographic micropatterning, which provides new insights into how geometry of extracellular matrix (ECM) regulates the organization of actin cytoskeleton.[6] Siyi Qiu *et al.* revealed microbial spore responses to microwave radiation by single-cell analysis, providing a new perspective on the responses of living single cells to microwave radiation.[7] Fanyi Kong *et al.* showed continuous infrared light suppressed transmembrane Na currents irrelevant to K channel on the freshly isolated hippocampal neuron cell, which could be explained by a membrane capacitance mediation process.[8]

Finally, we would like to thank all the contributing authors for making this issue possible. More articles focused on Single Cell Analysis will be published in the next special issue.

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