ISSN: 0256-307X

中国物理快报
Chinese Physics Letters

Volume 25 Number 11 November 2008

A Series Journal of the Chinese Physical Society
Distributed by IOP Publishing

Online: http://www.iop.org/journals/cpl
http://cpl.iphy.ac.cn

CHINESE PHYSICAL SOCIETY

CHIN.PHYS.LETT. Vol. 25, No. 11 (2008) 4013

Distinguishing Structure Change of Cells Based on Analysis of Light Scattering Patterns *

JIN Yong-Long(金永龙)**, YANG Fang(杨芳), WANG Meng(王孟), ZHANG Yu(张宇), GU Ning(顾宁)

Jiangsu Key Laboratory for Biomaterials and Devices, State Key Laboratory of BioElectronics, Southeast University, Nanjing 210096

(Received 25 April 2008)

We develop a new method to distinguish structural change of cells based on light scattering and Fourier spectra analysis. The light scattering detection system is composed of a laser source, an optical microscope, a CCD with high resolution and low distortion. After the scattering patterns of cells are recorded by the CCD, the Fourier spectra are obtained by the intensity distribution of scattered light. In the experiment, the change of cell structure is designed by sonication treatment. It is found that different typical peaks can be shown in the Fourier spectra of MCF7 cells with and without sonication treatment, which indicates that this method can be used to distinguish the structural change of cells.

PACS: 42.68.Mj, 87.18.Hf

The detection of light scattering is a relatively low cost, high speed and real-time method without any other accessory material. Therefore, it has been extensively applied to many detection technological fields.[1−6] Biological cells contain a number of different types of organelles in the cytoplasm, such as nucleus, mitochondria and lysosomes etc.. These organelles can act as light scattering centres because they have higher refractive index than the cytoplasm. When the cells are illuminated by a laser beam, the light will be scattered around the cells. Different structures of cells will lead to different scattered light patterns. Thus, the microstructure information of cells may be acquired by the different specific scattering light pattern features. This method may have great potential to distinguish the cancerous cells and normal cells in the biomedical applications.[7−9] Generally speaking, the structures of cancer cells, including nuclear, membrane, and cytoskeleton, etc, are different from the normal cells.[9] The identification of structure change of cancer cells is very helpful for further understanding their functional characteristics. In this study, we design a light scattering detection system to capture the structural change of cells by analysing the light scattering signal. A 650 nm fibre coupled laser is used to excite single cells, and the CCD images of light scattering patterns of cells are recorded. After all the images are collected, the Fourier spectra transformation are performed according to the light scattering patterns to obtain the information about the structural change of cells. It is demonstrated that different structures of MCF7 cells with and without sonication treatment could be distinguished by comparison of the Fourier spectra, which may provide an important and accurate method for the detection of cancer cells with identifying their structure changes.

The human breast cancer cell lines MCF7 were cultured in RPMI 1640 media containing 10% fetal bovine serum (FBS), 100 U/ml penicillin and 100 μg/ml streptomycin at 37°C in a humidified 5% CO_2 atmosphere. Cells were harvested from cultures and were resuspended in the fresh media. Microspheres were purchased from BD Biosciences Company, USA.

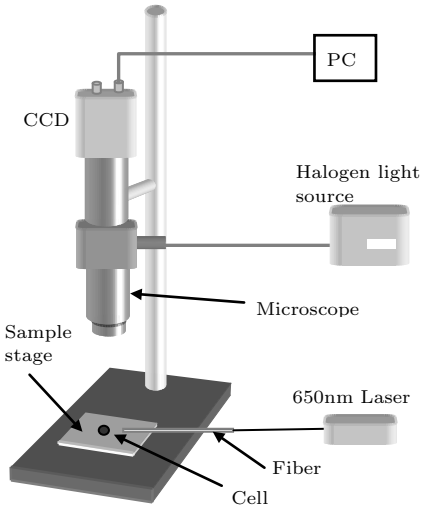

Fig. 1. Schematic diagram of the detection system.

MCF7 cell samples were divided into two groups. One is control group without ultrasound exposure. The other is treated group with ultrasound exposure. An arbitrary waveform generator (Agilent 33250A, USA) is used to produce a sinusoidal signal. The central frequency of the transducer is 1MHz and the focal

*Supported by the National Natural Science Foundation of China under Grant No 90406023.

**Email: hiroi@163.com

distance is 8 cm. The transducer and test-tube filled with cell suspension were mounted in a tank with degassed water. Output of the function generator was adjusted to 1000 mV under the 1MHz transducer frequency and the sample tube was exposed to the sound field produced by the transducer for 40 s. The acoustic pressure was measured to be 0.48 MPa by using the calibrated hydrophone.

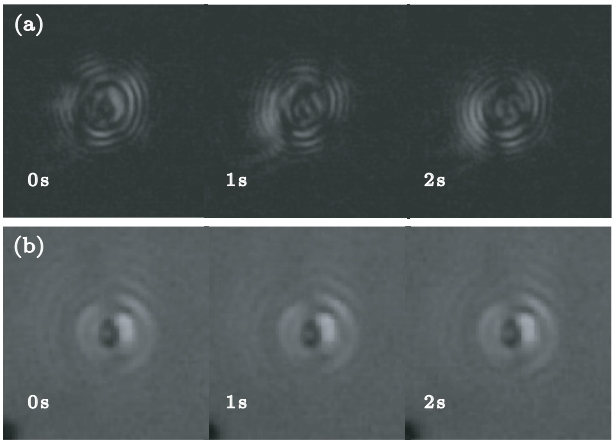

Fig. 2. Light scattering patterns of different samples (interval is 1 s). (a) MCF7 cell without ultrasound exposure, (b) standard microspheres.

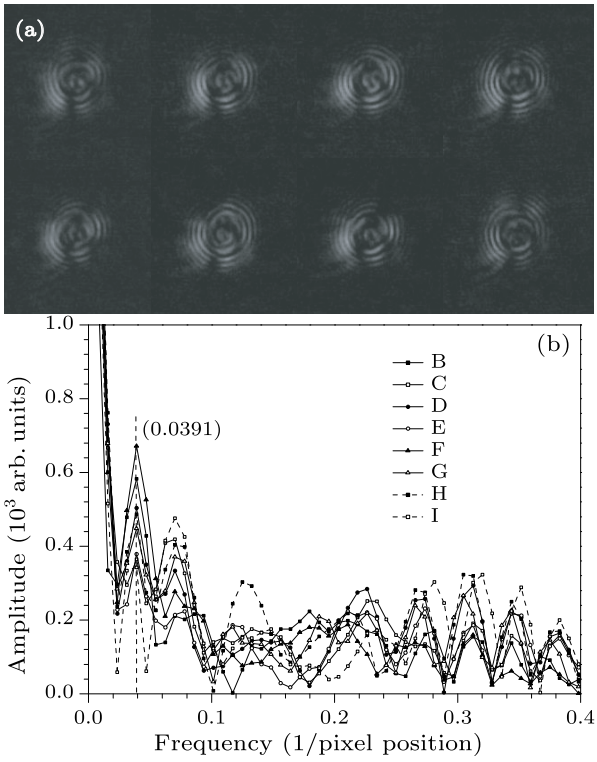

Fig. 3. Some scattering patterns (a) and their Fourier spectra (b) of MCF7 cell without ultrasound exposure. These Fourier spectra have the same typical peaks at 0.0391 (1/pixel position).

A Navitar Zoom6000 system (USA) and a SENTech STC-N63 CCD camera were used in the optical system. The laser source is a 650-nm fibre coupled laser. The transmission media is a single mode fibre and the front end diameter of fibre is 20 μm. The schematic diagram of the detection system is shown in Fig. 1. The detection process is as follows: positioning cells to sample stage, adjusting microscope to focus on a cell for observation, aligning the fibre front end to the cell and adjusting the distance within a few micrometres to guarantee the incident light to be parallel light. On opening the laser power, the laser beam illuminated the cells and light scattering patterns were formed. After the light scattering patterns were recorded by the CCD camera, the scanning of the scattered light intensity distribution was performed on the scattering pattern. Last, Fourier transform on the scanned intensity distribution was carried out.

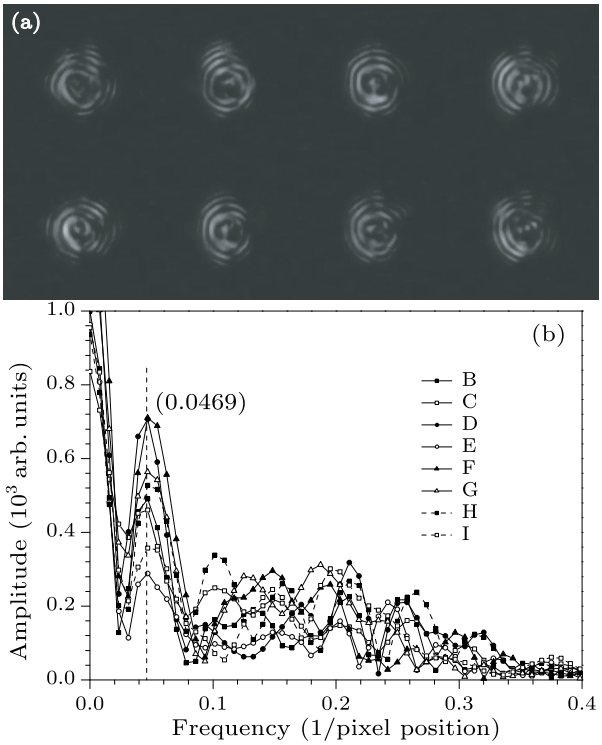

Fig. 4. Some scattering patterns (a) and their Fourier spectra (b) of MCF7 cell treated by ultrasound exposure. These Fourier spectra have the same typical peaks at 0.0469 (1/pixel position).

Upon the laser beam illumination, the light scattering patterns were formed and captured by the CCD camera. Figure 2 shows the light scattering patterns of MCF7 cells without ultrasound treatment and microspheres. It is found that the cells light scattering patterns are changed with time. However, the light scattering patterns of microspheres remain unchanged. The reason may be that for living cells, the organelles and cell membrane are not static and the overall structure of cell is dynamically changed, resulting in a random change of light scattering pat-

terns. The structure and morphology of microspheres prepared by polymer are constant. Thus no change is observed in the light scattering patterns. Because of the dynamic fluctuation of MCF7 cell scattering patterns, we could not obtain the cell's feature from one picture of light scattering pattern. It has been reported that the size variations or/and the refractive index variations will change the typical frequency peaks of Fourier spectra obtained by intensity distribution of the scattered light of cells.[10] Therefore, in order to extract the structural information of cells, Fourier transform of scattering patterns has been performed. Some scattering patterns of MCF7 cells without ultrasound exposure and their Fourier spectra are shown in Fig. 3. It can be observed that the patterns of scattered light are different from each other, but all the Fourier spectra have a typical peak at 0.0391 (1/pixel position). Could this typical peak at 0.0391 serve as a feature of MCF7 cells? In order to understand the typical peak, a comparative experiment was explored. The MCF7 cells, changed on the structure by sonication treatment, were also performed by the same imaging and Fourier transform. The scattering patterns and Fourier spectra of MCF7 cells treated by ultrasound exposure are shown in Fig. 4. Because the light scattered patterns of MCF7 cells treated by ultrasonic exposure are also different from each other, it is difficult to distinguish the difference between MCF7 cells with and without ultrasonic exposure only according to the light scattered patterns. However, their Fourier spectra have a typical peak at 0.0469 (1/pixel position), which is different from those of the MCF7 cells without ultrasound exposure. The typical peak of MCF7 cells was changed from 0.0391 to 0.0469 after the ultrasound treatment. The reason may be that under ultrasound exposure, the damage or perforation of the cell membrane occurs.[11,12] The structural change of MCF7 cells may result from the diffusion of contents in cytoplasm outside of the cells and the increase of cell membrane permeability after ultrasound exposure.[13] From the result, we presume that the typical peak of Fourier spectra can serve as a specific spectral feature which reveals the structural change of cells. Although this result is limited to qualitative analysis currently, by optimizing the detection system and enhancing the detection precision, this detection technique can be used as a quantitative analysis tool of cell structure.

In summary, we have presented a light scattering pattern analysis technique for distinguishing different structures of cells, based on the relationship between light scattering pattern and structures of cells. The light scattering patterns are recorded by a CCD camera and transformed into Fourier spectra to obtain the specific typical peak, which indicates the structure change of cells. The result shows that this technique may distinguish the structural difference of MCF7 cells before and after ultrasound exposure. It is expected that this technique can be further developed as an analysis tool for distinguishing cells with different structures such as cancer cells and normal cells.

References

[1] Ghosh N, Buddhiwant P, Uppal A et al 2006 *Appl. Phys. Lett.* **88** 084101
[2] LI X, Allen T and Vadim B 2005 *IEEE J. Sel. Top. Quant. Electron.* **11** 759
[3] Shu X M, Fang J, Shao Qet al. 2006 *Chin. Phys. Lett.* **23** 385
[4] Konstantin S, Rebekah D, Kirk G et al 1999 *Opt. Express* **5** 302
[5] Mourant J R, Canpolat M, Brocker C et al 2000 *J. Biomed. Opt.* **5** 131
[6] Singh K, Liu C, Capjack C et al 2004 *IEE Proc. Nanobiotechnol.* **151** 10
[7] Perelman L T, Backman V, Wallace M et al 1998 *Phys. Rev. Lett.* **80** 627
[8] Kim Young L, Liu Y, Wali Ramesh Ket al 2003 *IEEE J. Sel. Top. Quant. Electron.* **9** 243
[9] Backman V, Wallace M B, Perelman L T et al 2000 *Nature* **406** 35
[10] Su X T, Wojciech R, Clarence C et al 2007 *Proc. SPIE* **6446** 64460W
[11] Liu Q H, Wang X B, Wang P et al 2006 *Ultrasonics* **45** 56
[12] Koichi O, Katsuro T, Toshiki U et al 2001 *Med. Electron Microsc.* **34** 249
[13] Lu Q and Qiu T Q 2006 *J. South Chin. Univ. Technol. (Natural Science Edition)* **34** 51 (in Chinese)