北京航空航天大學
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Long working distance DHM for space application

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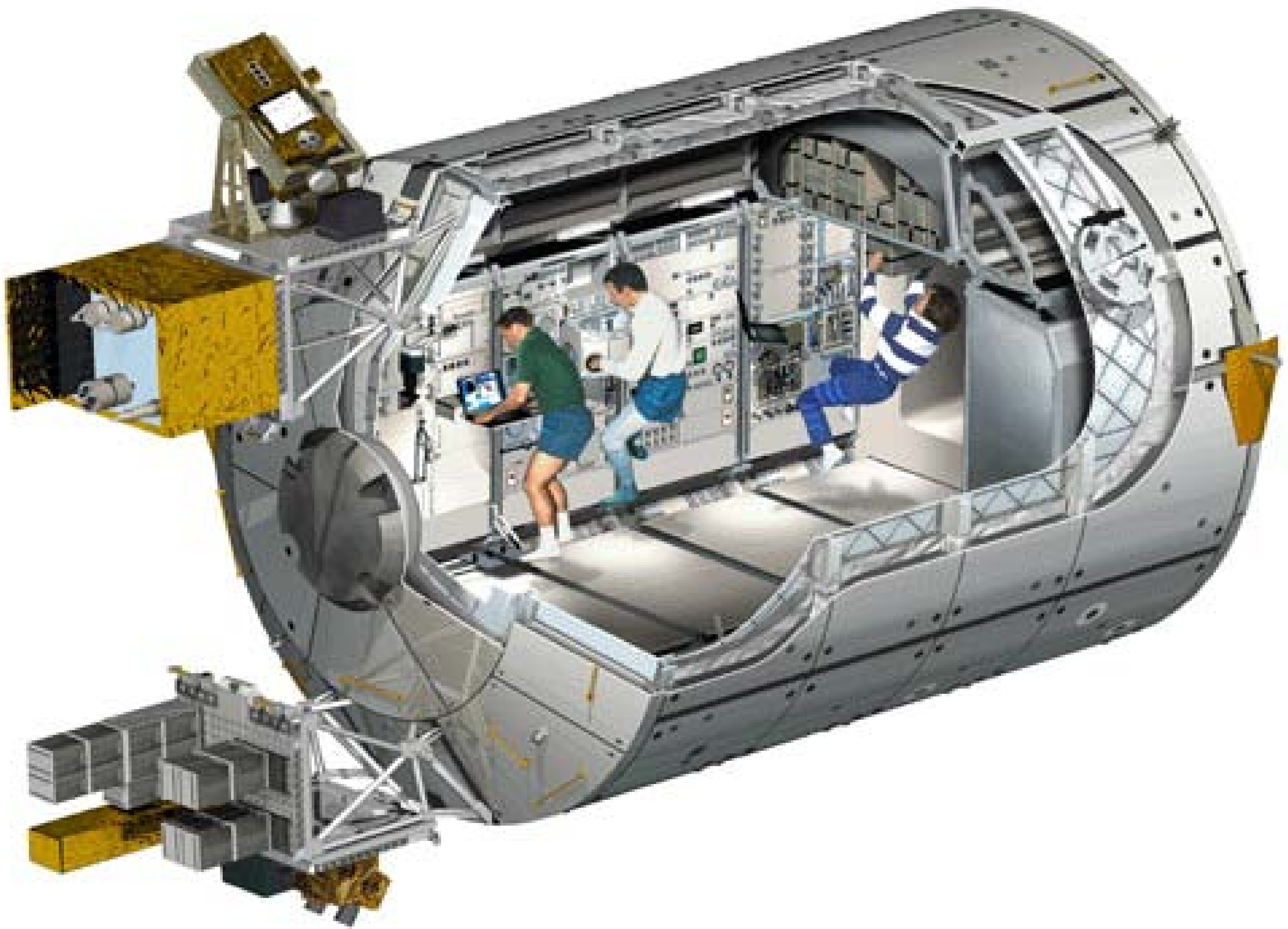
Content

- Why do we make this research
- What is mean for long working distance
- How make it possible
- Works on that

Micro-observation in space plateform

- The payload for micro-observation
  - Cells
  - Materials change
  - Optical lens' damage
- The equipments for monitor the structures of the plateform
  - Outside damage
  - Payload outside

Space biological experiment



Micro-observation in space plateform

- The payload for micro-observation
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  - Optical lens' damage
- The equipments for monitor the structures of the plateform
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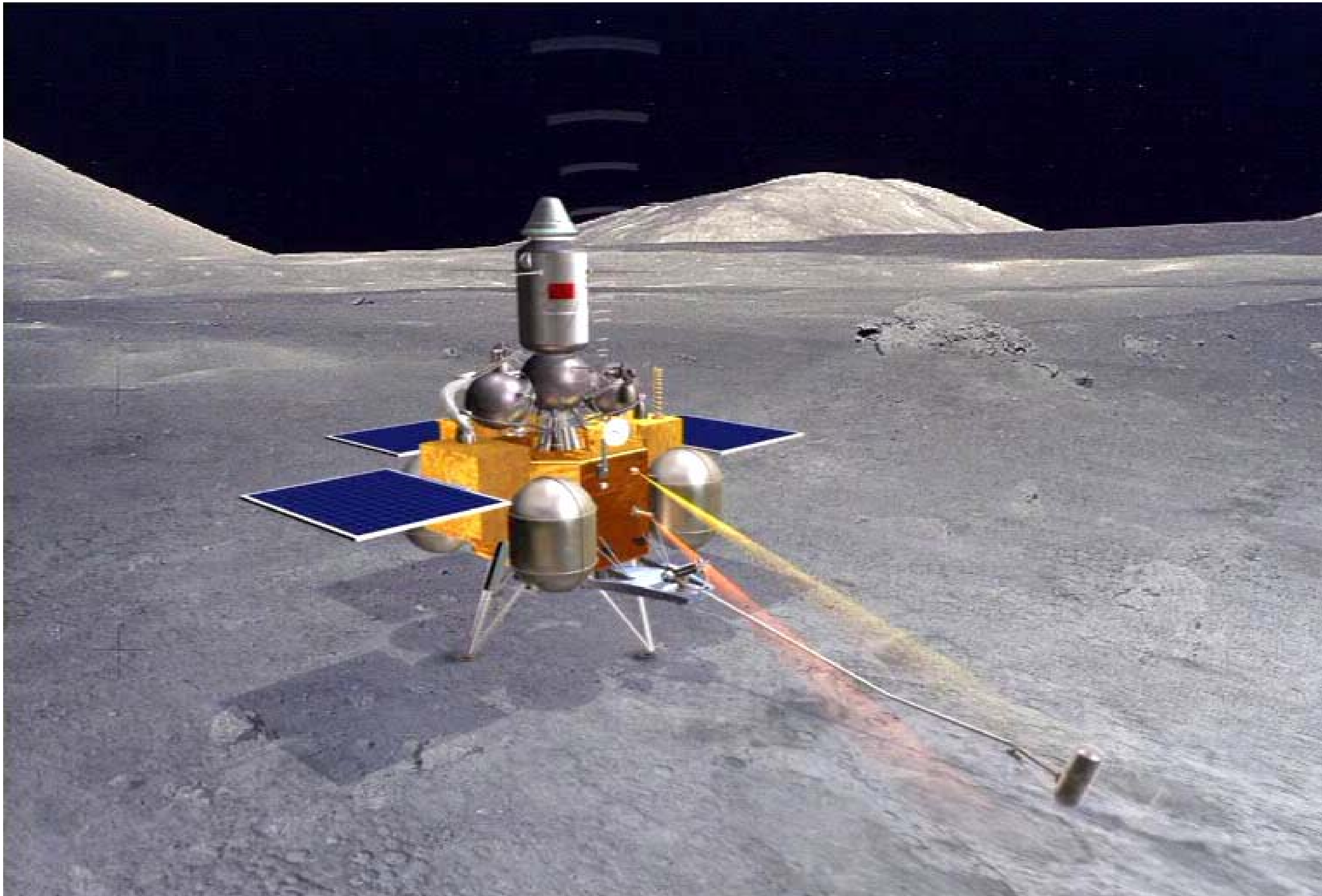
Surface cracks of spacecraft

Surface explorations



Micro-observation in space platform

- The payload for micro-observation
  - Cells
  - Materials change
  - Optical lens' damage
- The equipments for monitor the structures of the platform
  - Outside damage
  - Payload outside

Microscopy is a powerful tool for Space S & T.

Microscopy usually refers to:

✈Optical microscopy

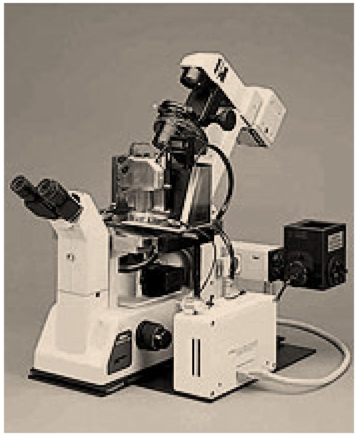
Only obtain 2D information of the object

Resolution and image quality are restricted by object lens

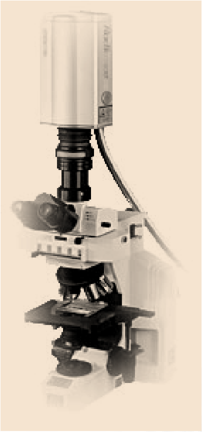
✈Scanning microscopic techniques

Obtain 3D information of the object

Complex structure, trivial procedures and rigid working environment



AFM



LSCM

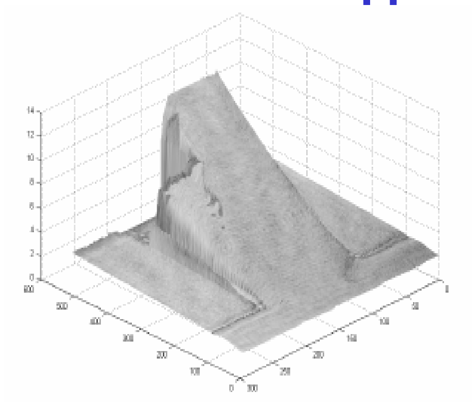
STM

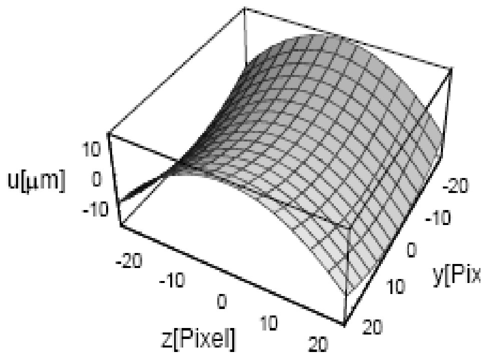
Digital holographic microscopy (DHM), which is based on digital holography, aims at investigating small samples, such as life cells, MEMS, micro-optics, etc.

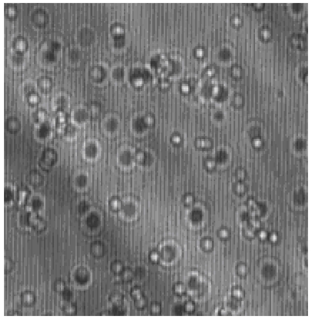
DHM has numerous advantages:

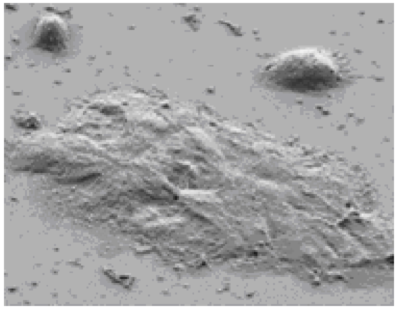
- **It is a noninvasive, noncontact and in-situ measurement technique.**
- **It allows for 3D quantitative analysis.**
- **Since no scanning is involved, it may operate in real time.**
- **It is marker-free and no pretreatment is required.**

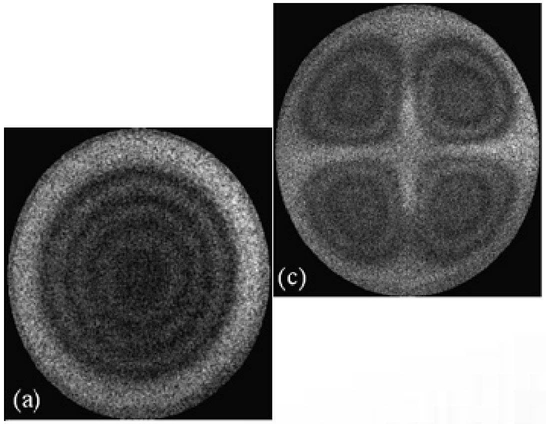
DHM could be applied to:

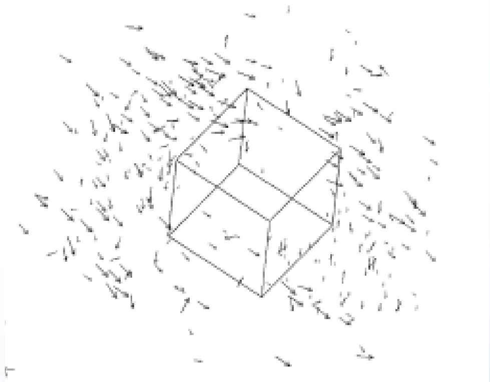












3D imaging

& profile analysis

Deformation & vibration

measurement

Particle field

observation

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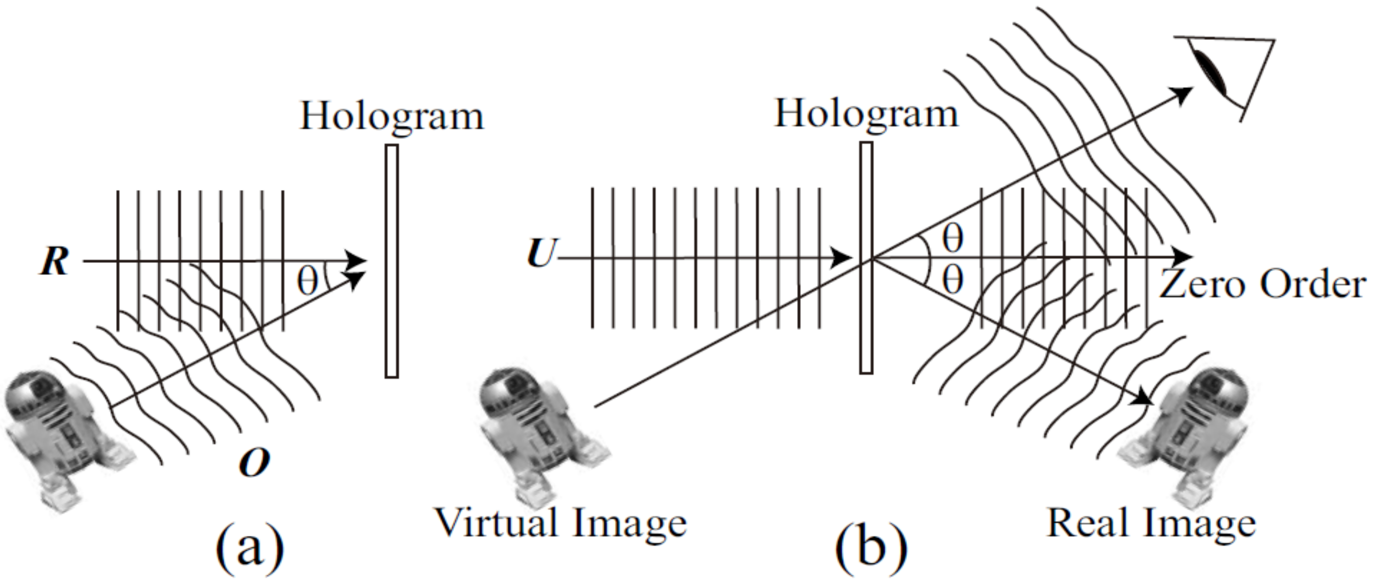
Introduction to Digital Holography

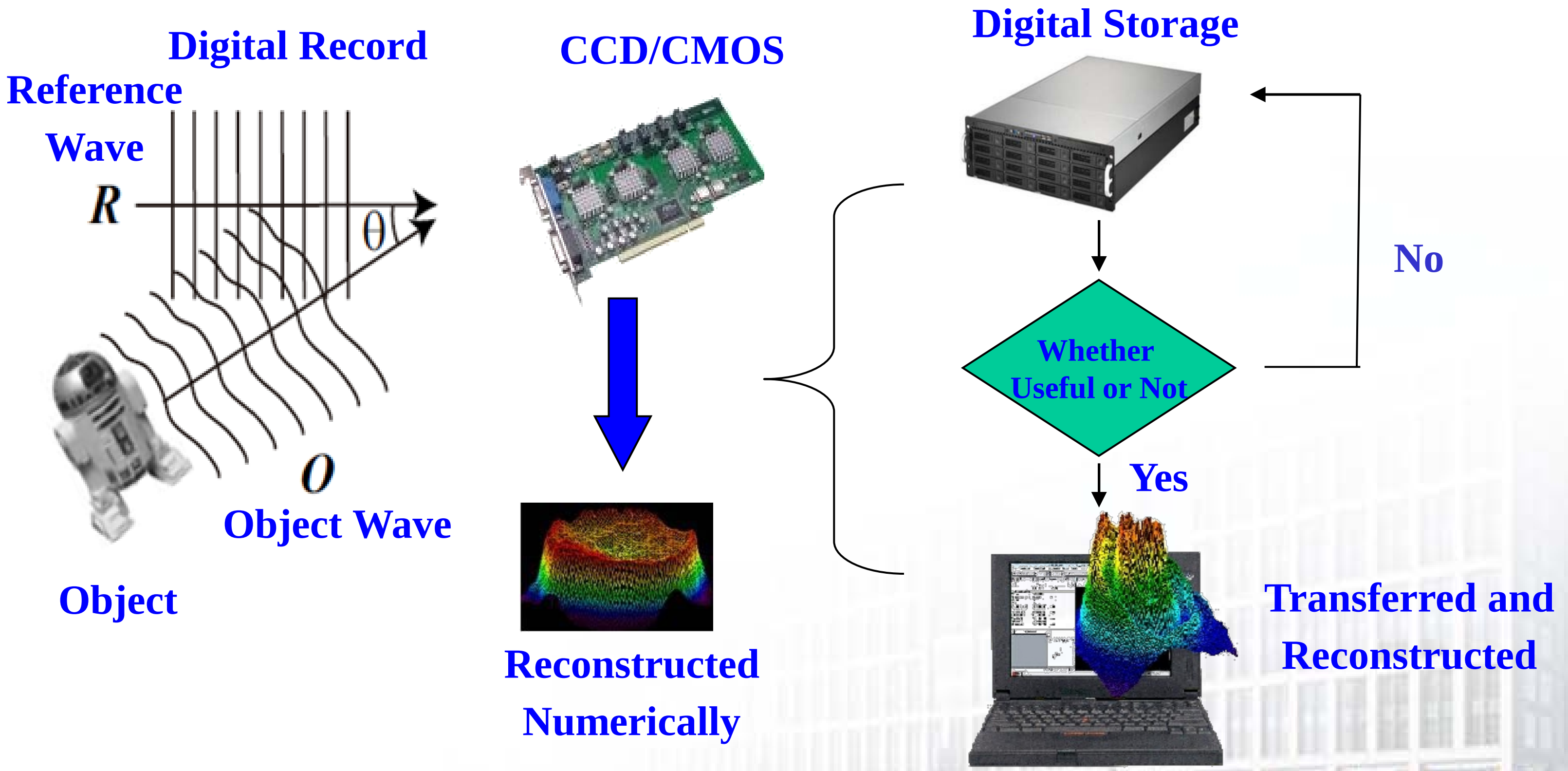
- Holography is a imaging technique that can recover the whole information of the object under inspection from the holographic interferogram.



Recording **Reconstruction**

Introduction to Digital Holography

- **Digital holography (DH) is based on the optical holography, which is a novel imaging technique that can quantitatively recover both the intensity and the phase information of the object under inspection with the simulated reference wave from the single the hologram recorded digitally by CCD and transferred to a computer.**



Introduction to Digital Holography

Mathematical explanation of holography:

Recording:

$$H = |R+O|^2 = (R+O)(R^* + O^*)$$

$$= R^2 + O^2 + 2|RO|\cos(\Delta\varphi)$$

$$= \underbrace{R^2 + O^2}_{\text{zero-order term}} + \underbrace{OR^* + RO^*}_{\text{conjugate terms}}$$

H **denotes the intensity of hologram.**

R **and** ***O*** **denote the complex amplitude of reference and object wave, respectively.**

Reconstruction:

$$RH = R(R^2 + O^2) + \underline{O|R|^2} + R^2O^*$$

If $|R|^2 = 1$ → O

Once O in hologram plane is obtained, it could be propagated to image plane by simulating a diffraction procedure:

- Fresnel algorithm: 2D FFT

$$b'(n\Delta x', m\Delta y') = e^{i\pi d\lambda\left(\frac{n^2}{N^2\Delta\xi^2}+\frac{m^2}{M^2\Delta\eta^2}\right)} FFT\left[\mathrm{h}(k\Delta\xi, l\Delta\eta)\mathrm{r}^*(k\Delta\xi, l\Delta\eta)e^{\frac{i\pi}{d\lambda}\left(k^2\Delta\xi^2+l^2\Delta\eta^2\right)}\right]$$

- Convolution algorithm: 2D FFT & IFFT

$$b'(n\Delta x', m\Delta y') = FFT^{-1}\left\{FFT\left\{h(k\Delta\xi, l\Delta\eta)\cdot r^*(k\Delta\xi, l\Delta\eta)\right\}\cdot FFT\left\{g(k\Delta\xi, l\Delta\eta)\right\}\right\}$$

- Angular spectrum algorithm: 2D FFT & IFFT

$$b'(n\Delta x', m\Delta y') = FFT^{-1}\left\{FFT\left\{h(k\Delta\xi, l\Delta\eta)\cdot r^*(k\Delta\xi, l\Delta\eta)\right\}\cdot e^{\frac{i2\pi d}{\lambda}\sqrt{1-\left(\frac{k\Delta\xi}{d}\right)^2-\left(\frac{l\Delta\eta}{d}\right)^2}}\right\}$$

Then the amplitude and phase image could be calculated.

Amplitude: $I(n\Delta x', m\Delta y') = \left|b'(n\Delta x', m\Delta y')\right|^2$ **Phase:** $\phi(n\Delta x', m\Delta y') = \arctan\frac{\mathrm{Im}\left\{b'(n\Delta x', m\Delta y')\right\}}{\mathrm{Re}\left\{b'(n\Delta x', m\Delta y')\right\}}$

Digital Holographic Microscopy

Nowadays, there are two methods to increase the lateral resolution in DHM:

◆**Pre-magnification approach:**

A Microscopic Objective (MO) is used to obtain a magnified image of the sample. The resolution depends on the numerical aperture (NA) of MO.

Only one hologram is needed to retrieve both amplitude and phase image.

The aberrations induced by MO must be compensated.

◆**Synthetic aperture approach:**

A series of holograms are captured in different recording conditions. Then these holograms are synthesized to obtain resolution enhanced image.

Since no lenses are involved , the working distance of the system isn't constrained to can be much longer, and the reconstructed images doesn't subject to lens aberration.

The record and synthesis of several holograms are time consuming.

Long-working-distance Synthetic Aperture DHM

Research Purpose

- An three-dimensional in-situ microscopy imaging with high resolution based on the digital holography at long working distance for the space in-suit microscopy measurement is proposed. Its superiorities are:

(1)It is a noncontact, in-situ measurement technique, which can obtain the 3D quantitative information of the object under inspection;

(2)It is provided of the flexible working distance and high-resolution imaging;

(3)It is in accordance with the basic design requirements for the miniaturization and the integration of the Load space science exploration.

Technical Specifications

- Capabilities of three-dimensional in-suit microscopy imaging at long work distance, which is varied from the 10*cm* to the 80*cm*;
- Lateral resolution could achieve to few tens of micrometers, and axial resolution could be increased to more than a dozen micrometers at the longest work distance.

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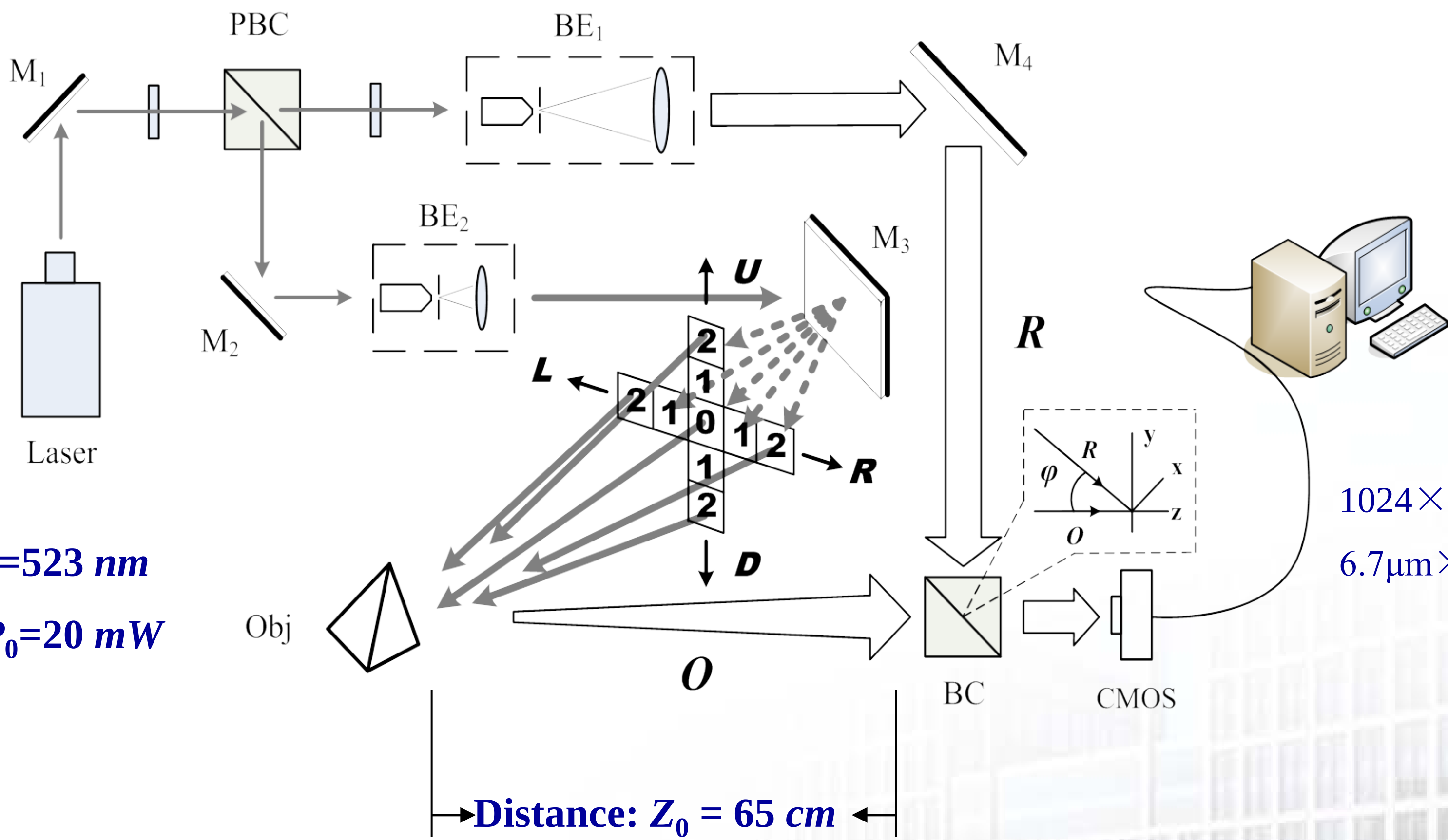
Long-working-distance Synthetic Aperture DHM

We developed a synthetic aperture method for DHM in our previous work. Different tilted plane wave illuminations were utilized in this approach.



λ*=523 *nm

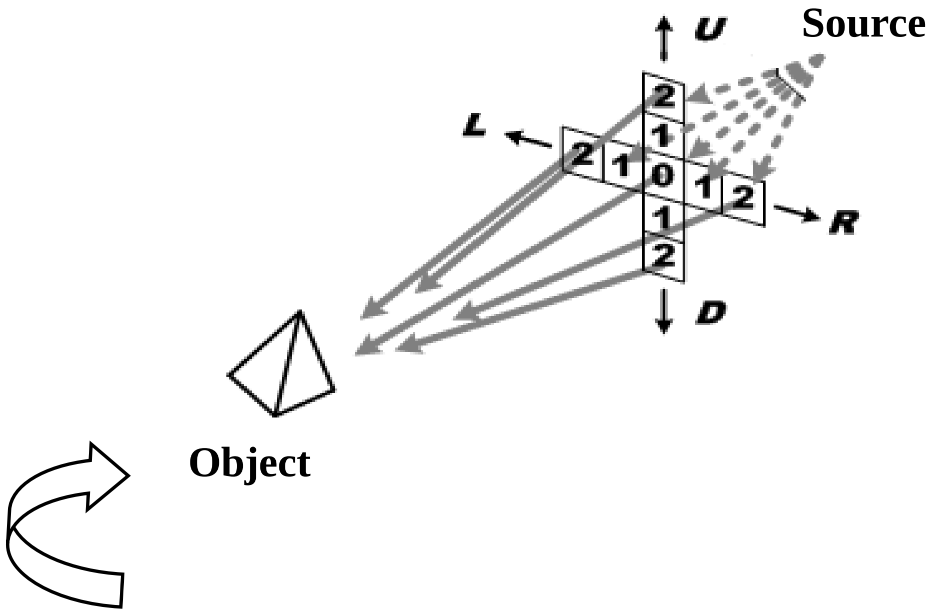
P_0*=20 *mW

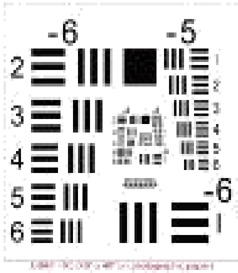
1024×1024 pixels

6.7μm×6.7μm

Schematic diagram of our method

Long-working-distance Synthetic Aperture DHM



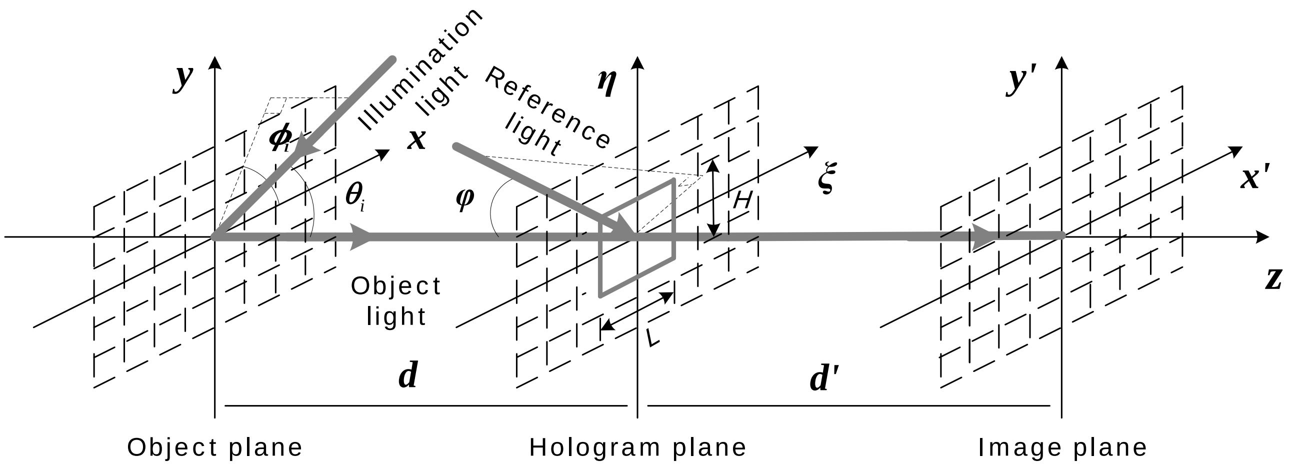


USAF-1951
Test Target

&

10mm ×10mm ×10mm

Long-working-distance Synthetic Aperture DHM



Reflective complex field in the object plane:

$$U_i(x,y)=b(x,y)A_i(\gamma_i,\zeta_i)$$

Spatial frequency spectrum of the reconstructed image in the image plane:

$$\tilde{U}_o(f_x,f_y)=A_0C\tilde{b}(f_x+\gamma_i,f_y+\zeta_i)\mathrm{rect}\left(\frac{\lambda df_x}{L}\right)\mathrm{rect}\left(\frac{\lambda df_y}{H}\right)$$

Spatial frequency spectrum of the synthesized image:

$$\tilde{U}_o^{sum}(f_x,f_y)=\sum_i A_0C'\tilde{b}(f_x+\gamma_i,f_y+\zeta_i)\mathrm{rect}\left(\frac{\lambda df_x}{L}\right)\mathrm{rect}\left(\frac{\lambda df_y}{H}\right)$$

$$=A_0C'\tilde{b}(f_x,f_y)\mathbf{SA}(f_x,f_y)$$

Synthetic aperture function:

$$\mathbf{SA}(f_x,f_y)=\sum_i \mathrm{rect}\left[\frac{\lambda d(f_x+\gamma_i)}{L}\right]\mathrm{rect}\left[\frac{\lambda d(f_y+\zeta_i)}{H}\right]$$

Long-working-distance Synthetic Aperture DHM

Multiple holograms are recorded with different titled illuminations

The complex amplitudes are reconstructed and magnified from these holograms by using digital arithmetic.

The synthesized image is obtained by incoherent superposition

The resolution is enhanced

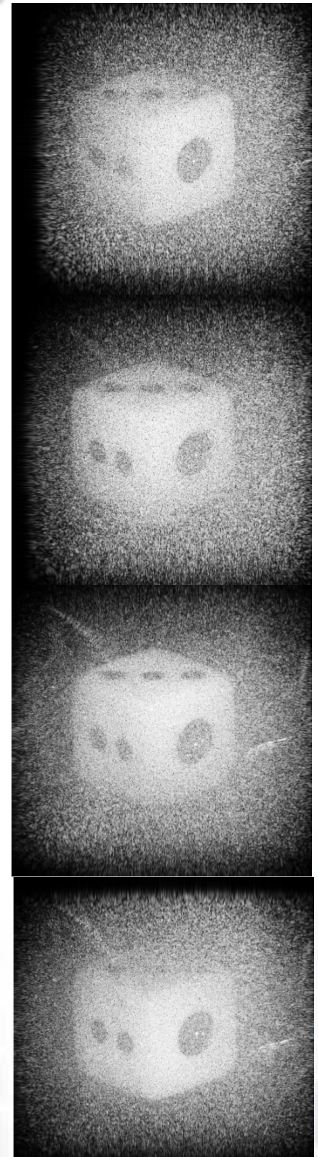
The speckle noise is suppressed

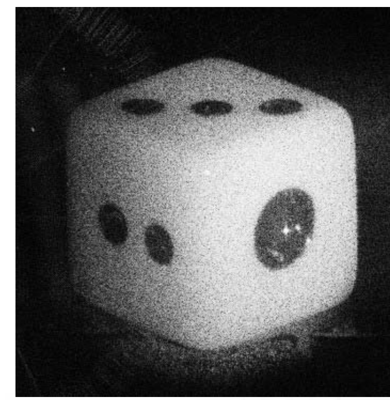
R2

L2

U2

D2





365 pixels×365 pixels

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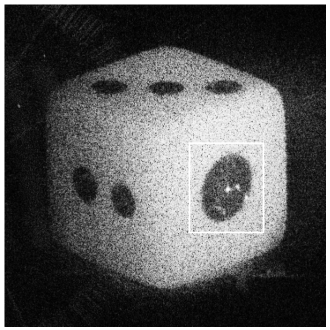
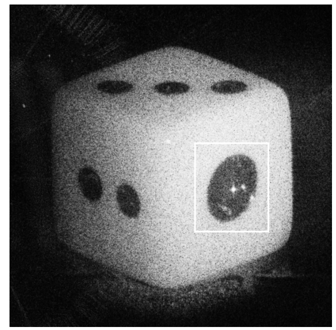
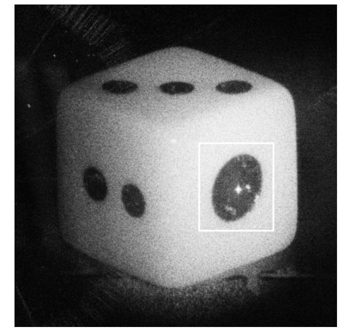
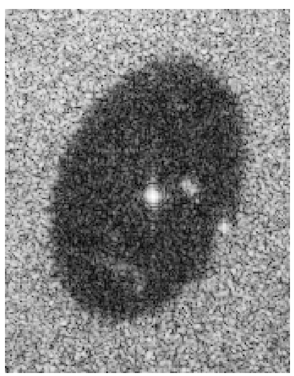
Long-working-distance Synthetic Aperture DHM

$\sum^{1} = \mathbf{0}$ | $\sum^{5} = \mathbf{R1 + L1 + U1 + D1 + 0}$ | $\sum^{9} = \mathbf{R1 + R2 + L1 + L2 + U1 + U2 + D1 + D2 + 0}$

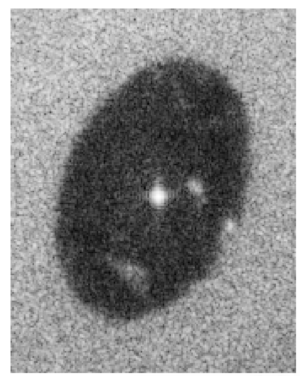
795pixels ×795pixels

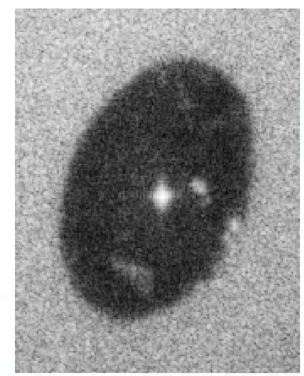
169pixels ×216pixels



Reconstructed with 1 hologram



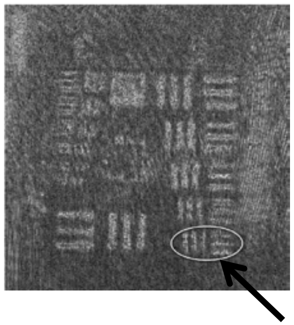
Reconstructed with 5 holograms



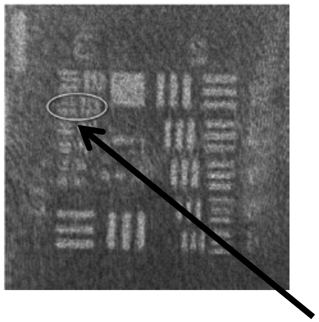
Reconstructed with 9 holograms

These results show that the image quality is effectively improved and the speckle noise is well suppressed with proposed method.

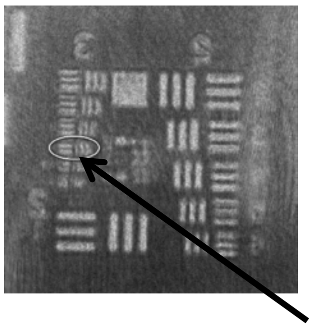
Long-working-distance Synthetic Aperture DHM



7.13 line-pairs/mm

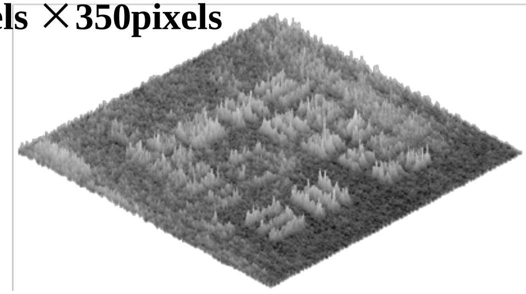


10.1 line-pairs/mm

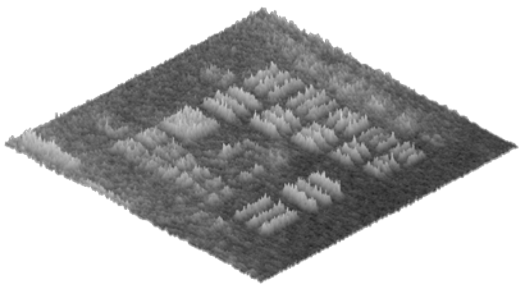


12.7 line-pairs/mm

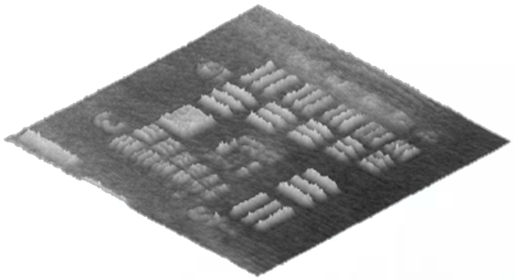
350pixels ×350pixels



Reconstructed with
1 hologram



Reconstructed with
5 holograms



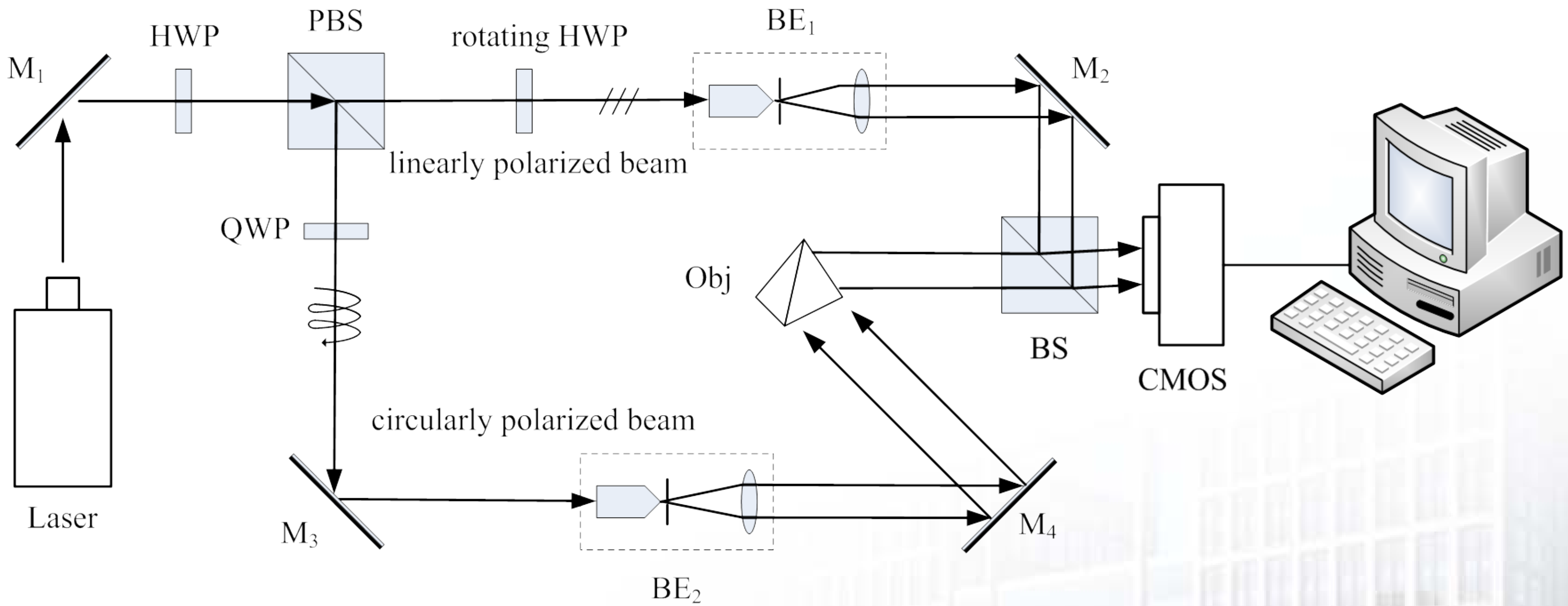
Reconstructed with
9 holograms

These results were obtained with resolution test target USAF-1951. The spatial resolution is effectively enhanced.

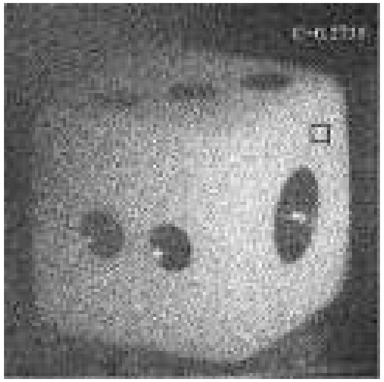
Pan Feng, Xiao Wen, Rong Lu. "Long-working-distance synthetic aperture Fresnel off-axis digital holography", Optics Express. 2009, 17(7):5473-5480， Virtual Journal of Biomedical Optics. 2009, 5(1).

Speckle noise reduction in digital holography by use of multiple polarization holograms

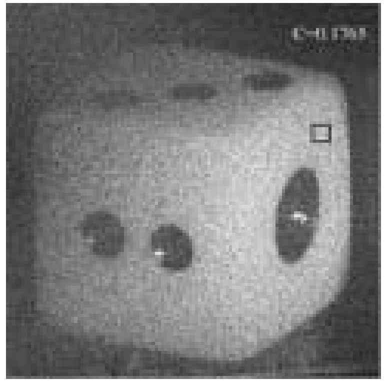
We propose a novel speckle reduction technique for digital holography by recording multiple polarization off-axis holograms. The speckle noise is suppressed by averaging the reconstructed intensity fields, and the resolution is improved.



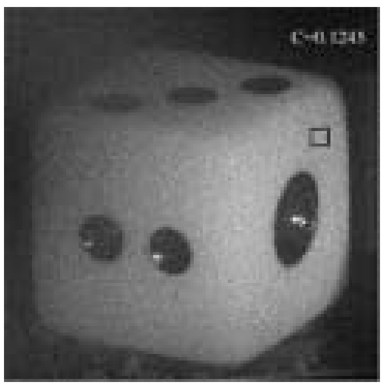
Schematic diagram of experimental setup



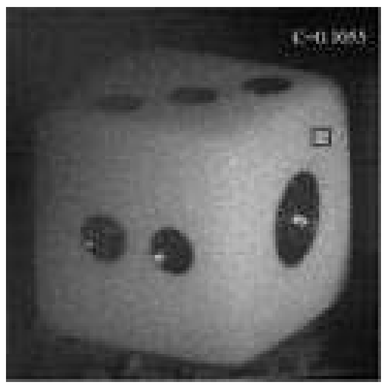
Reconstructed with 1 hologram



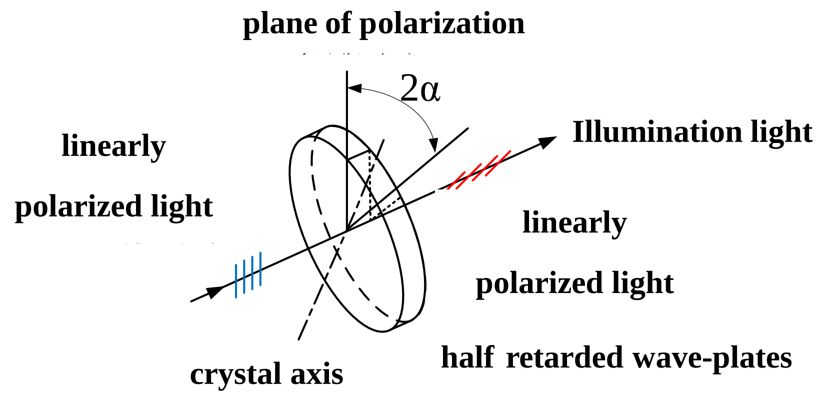
Reconstructed with 3 hologram

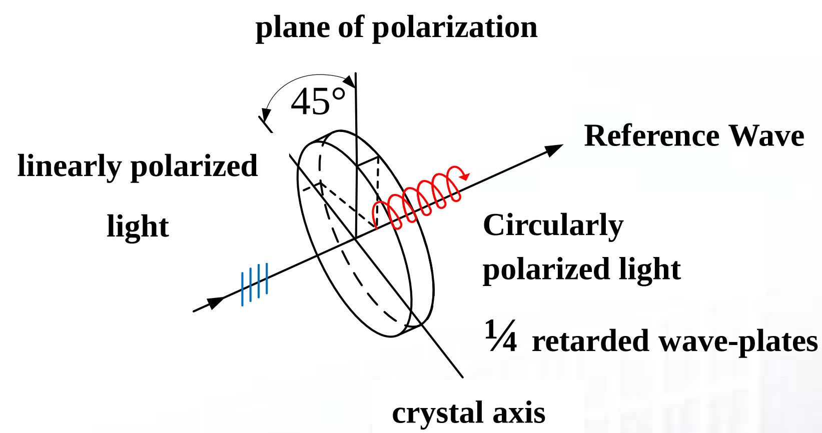


Reconstructed with 9 hologram



Reconstructed with 18 hologram

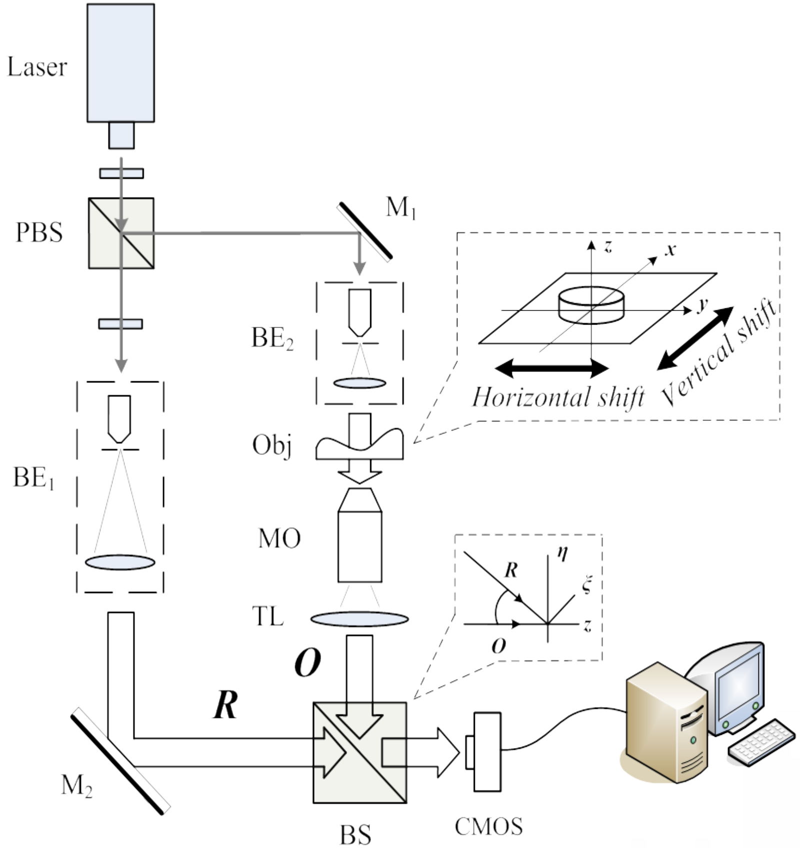




- ✓ *Lu Rong, Wen Xiao, Feng Pan, et al , "Speckle noise reduction in digital holography by use of multiple polarization holograms". Chinese Optics Letters. 2010, 8(7):653-655.*
- ✓ *Wen Xiao, Jing Zhang, et al, "Improvement of speckle noise suppression in digital holography by rotating linear polarization state", (accepted by Chinese Optics Letters).*

Coherent noise reduction in digital holographic phase contrast microscopy by slightly shifting object

By slightly shifting the specimen, a series of digital holograms with different coherent noise patterns is recorded. Through averaging these images, the coherent noise is reduced and the quality of phase contrast image is improved.



Schematic diagram of our method

Complex amplitude of specimen in object plane:

$$U_b(x,y)=b(x,y)\cdot s(x,y)$$

Complex amplitude of object wave in the recording plane:

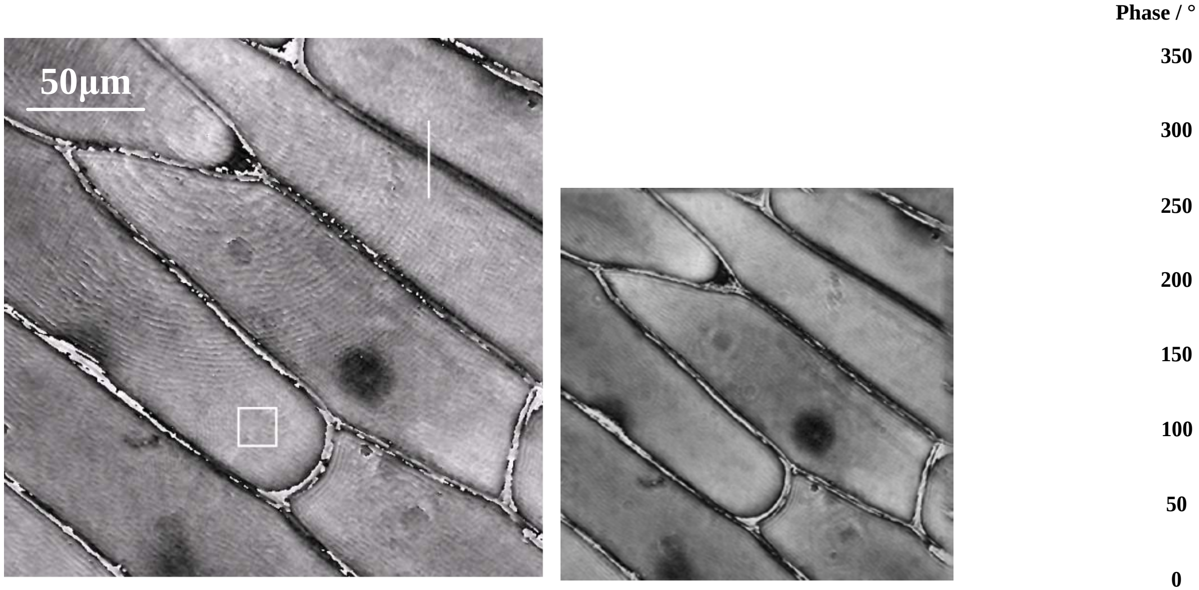
$$U_h(\xi,\eta)=\exp\left(i\frac{2\pi}{\lambda}\cdot\frac{\xi^2+\eta^2}{2d}\right)\cdot\mathbf{F}\left[U_b(x+\Delta x,y+\Delta y)\exp\left(i\frac{2\pi}{\lambda}\cdot\frac{x^2+y^2}{2d}\right)\right]$$

Complex amplitude of object wave in the imaging plane:

$$U_i(x',y')=C\cdot\mathbf{F}\left\{\left[U_h(\xi,\eta)\mathbf{rect}\left(\frac{\xi}{a},\frac{\eta}{b}\right)\right]\cdot\exp\left(-i\frac{2\pi}{\lambda}\cdot\frac{\xi^2+\eta^2}{2d}\right)\right\}$$

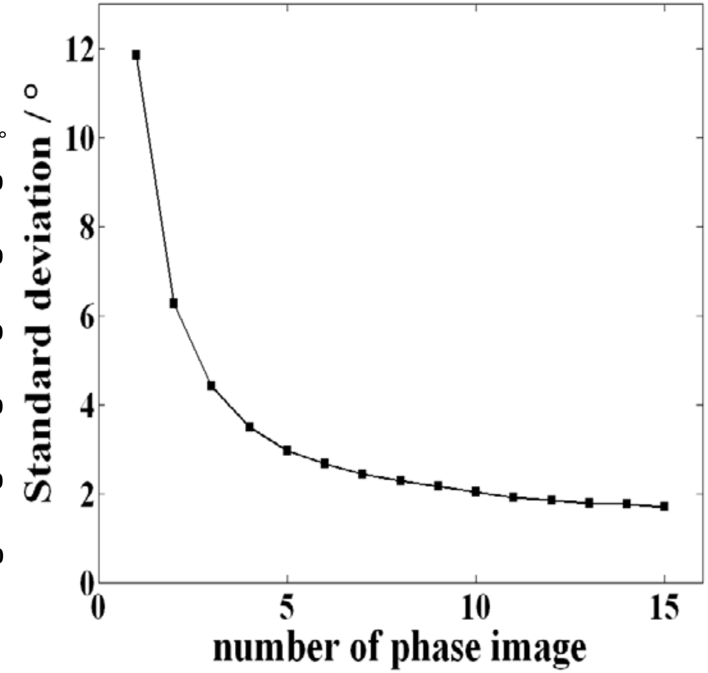
$$=C\cdot\left\{b(x+\Delta x,y+\Delta y)s(x+\Delta x,y+\Delta y)\exp\left(i\frac{2\pi}{\lambda}\cdot\frac{x^2+y^2}{2d}\right)\right\}$$

$$*\mathbf{sinc}\left(\frac{ax}{\lambda d},\frac{by}{\lambda d}\right)$$



(a) (b)

Reconstructed phase image by the proposed method, (a): result of a single hologram, (b): result after averaging phase images.



Cross correlation between the phase distributions of a uniform region indicated by white rectangle in the left figure.

Feng Pan, Wen Xiao, et al, "Coherent noise reduction in digital holographic phase contrast microscopy by slightly shifting object", (accepted by Optics Express)

Monitoring of Living Cells Growth under Microgravity

Gravity strongly influences the growth of living cells.

Microgravity environment was generated by a superconducting magnet system.

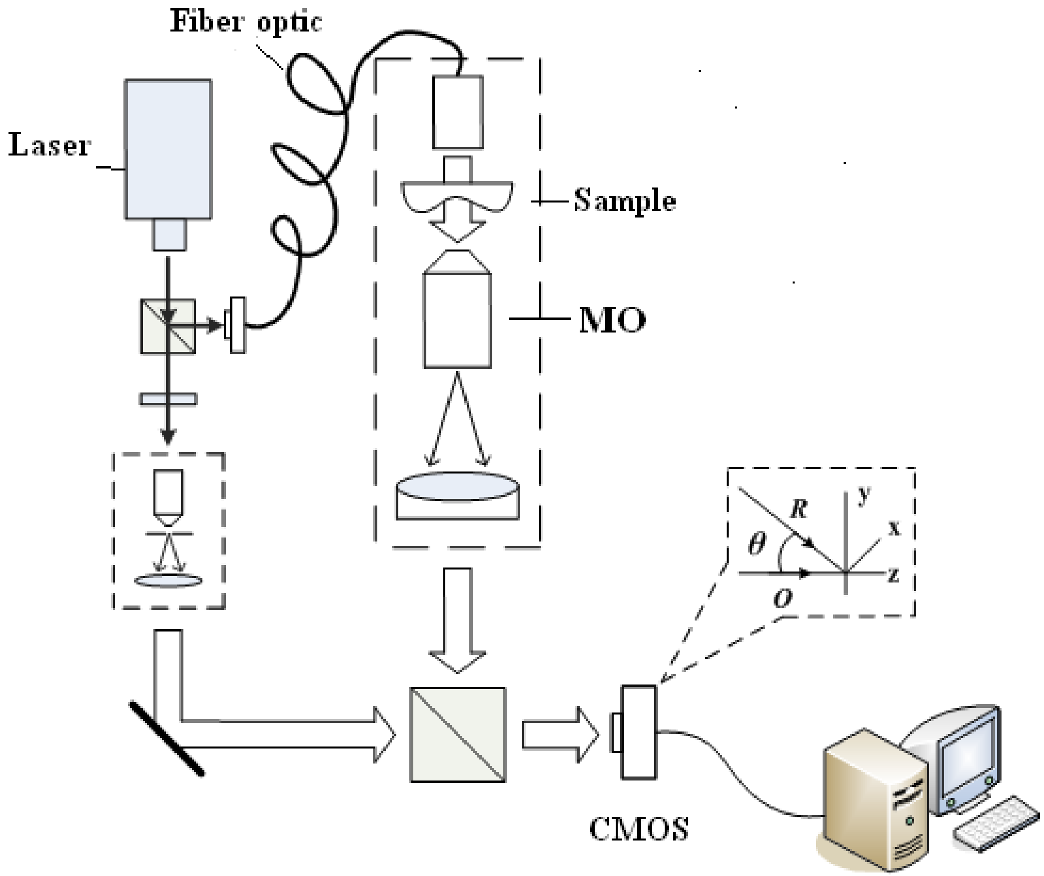
Mouse bone cells were cultivated at the 0g point, for 6 or 12 hours. Then we could monitor the growing procedure of bone cells after microgravity stimulation by DHM system.

The benefits of DHM system are as follows:

- **It is noninvasive and noncontact.**
- **It works in real time.**
- **It permits 3D quantitative analysis.**

Superconducting magnet system

DHM living cell observation system:



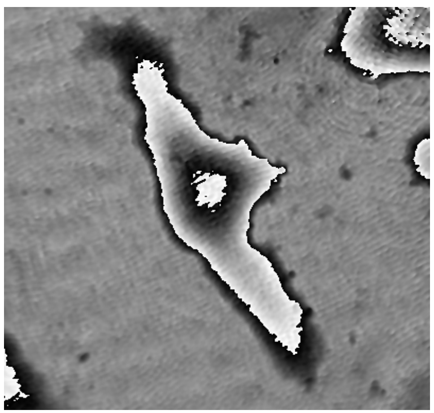
Schematic diagram

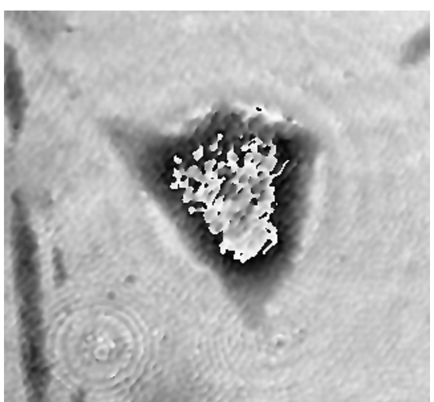
Photograph of optical setup

Monitoring of Living Cells Growth under Microgravity

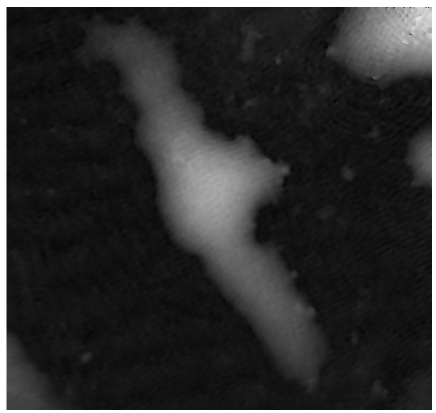
Living bone cell observation results (20x MO):

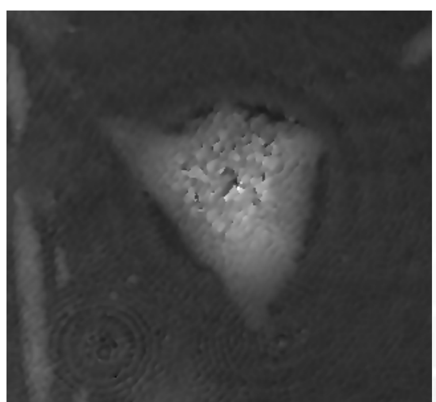
Phase image



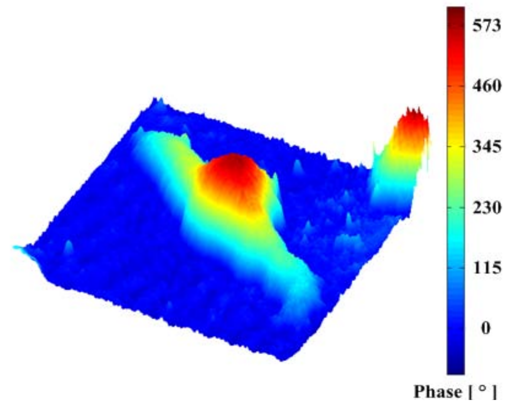


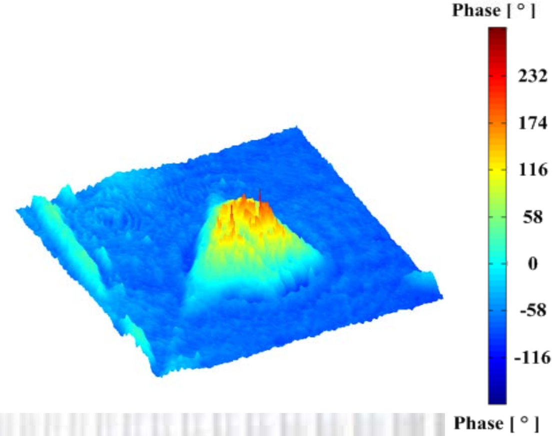
Unwrapped phase image





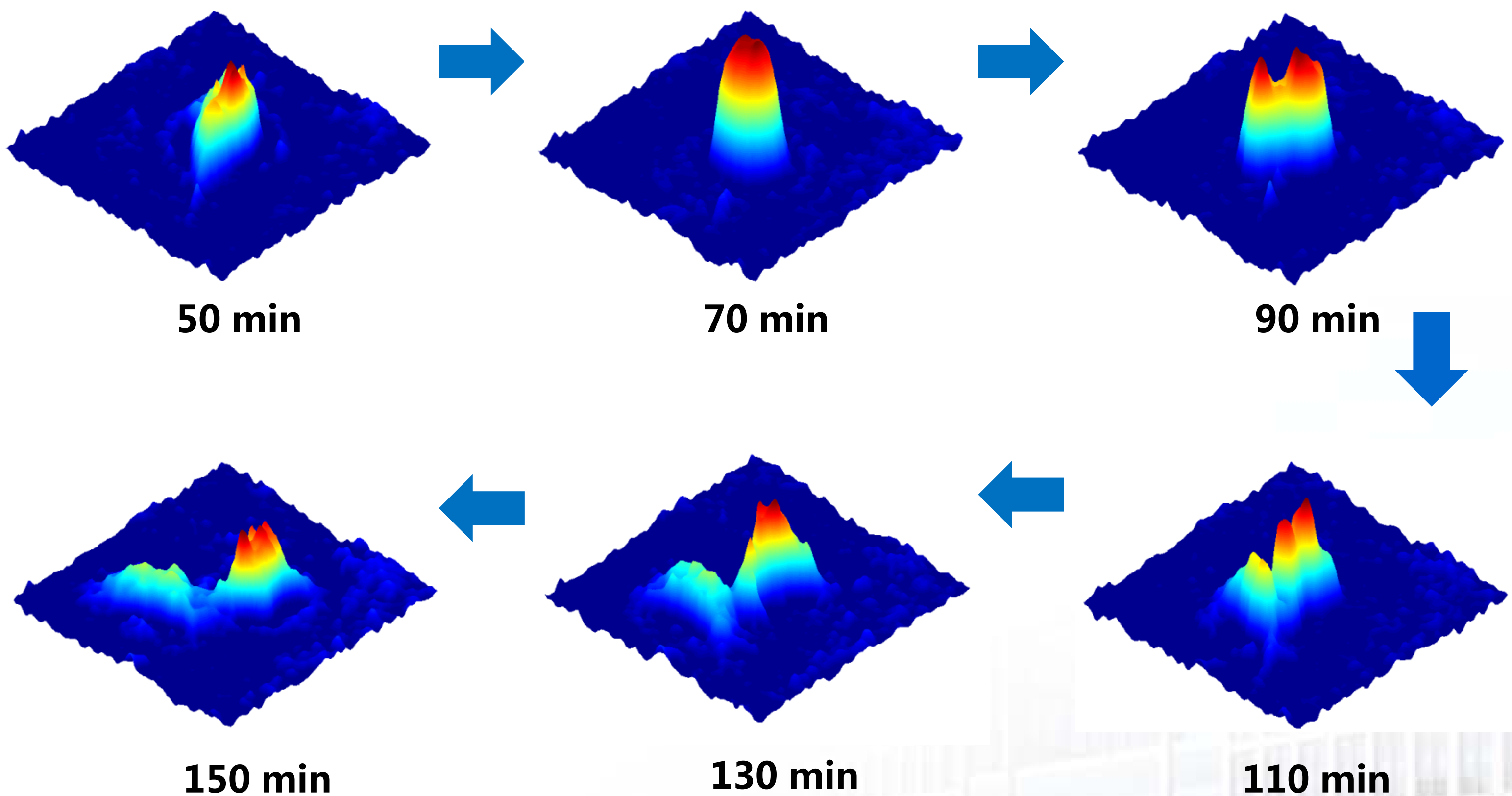
3D rendering of phase image





Monitoring of Living Cells Growth under Microgravity

Dynamic analysis of the division procedure of mouse bone cell, after 0g, 12 hours cultivation :

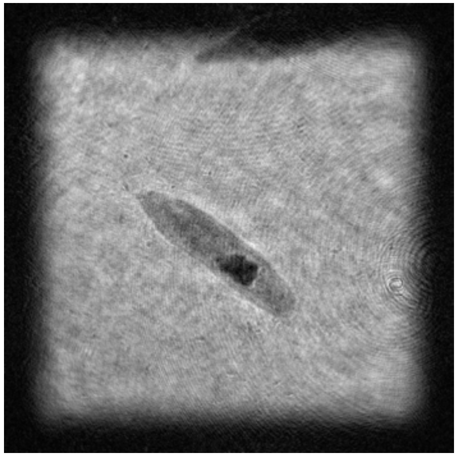


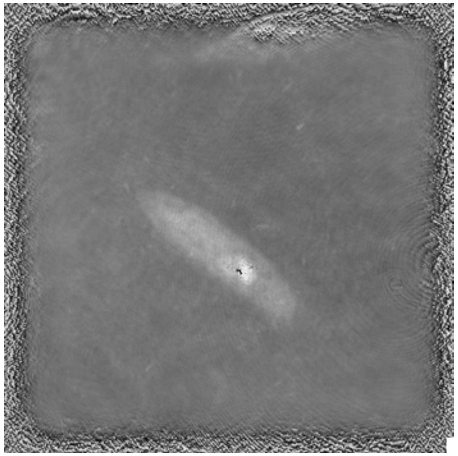
S.Liu, F.Pan, Z.Wang, F.J.Wang, L.Rong, P.Shang, W.Xiao. "Long-term quantitative phase-contrast imaging of living cells by digital holographic microscopy". Laser Physics, 2011, 21(4).

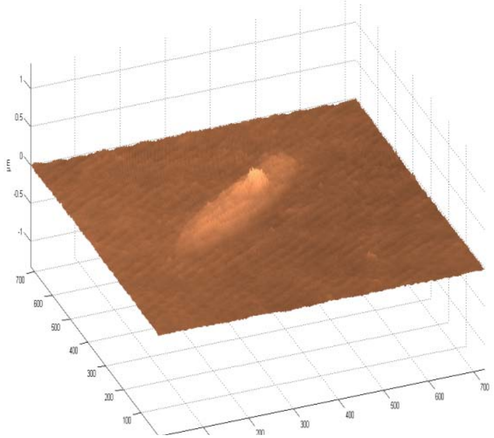
Biological Slices Observation Results

Amplitude image | **Phase image** | **3D rendering of phase image**

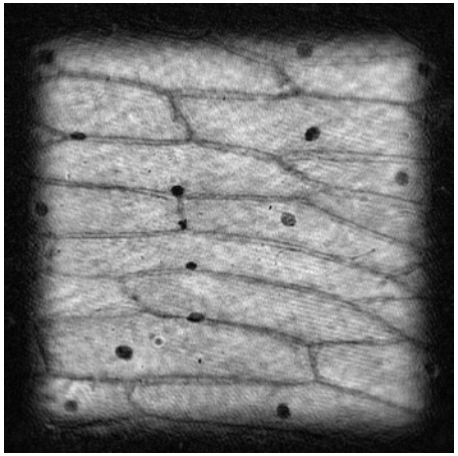
Paramecium

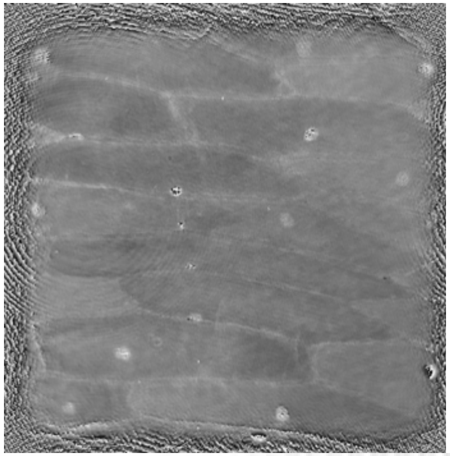


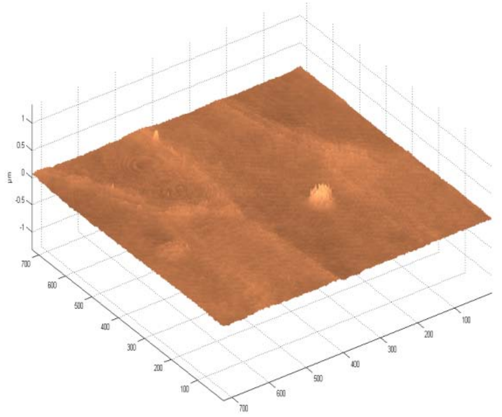




Onion





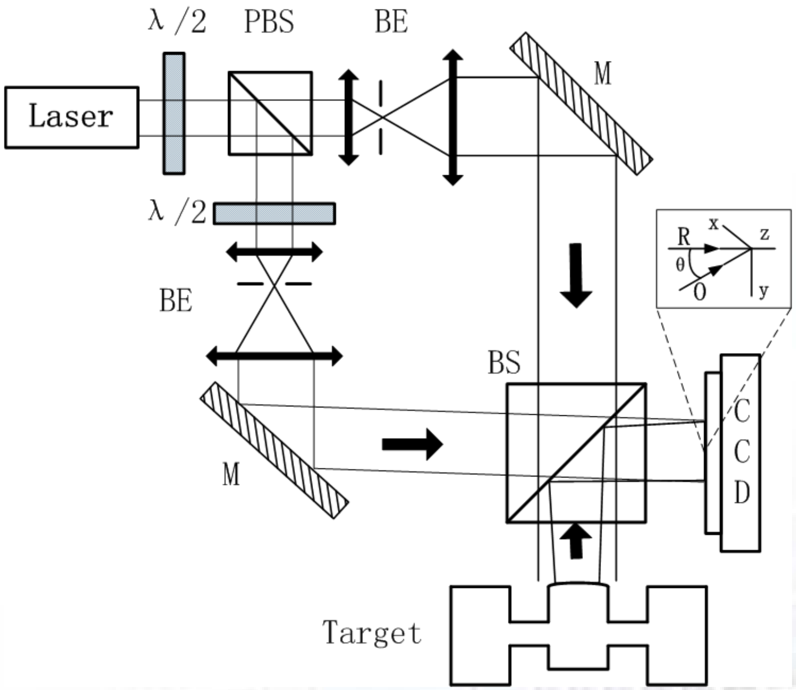


Profile and Defect Examination of Lens Surface

We have applied DHM to examine profile and defect of lens surface.

The in-situ measurement of lens surface could be realized with DHM.

Object under testing



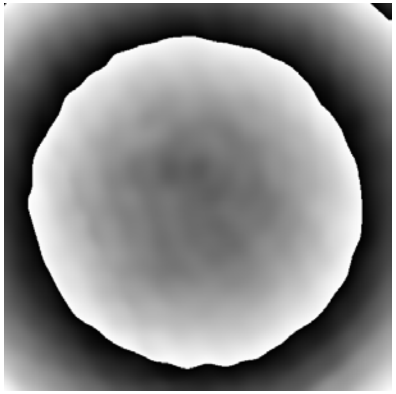
Schematic Diagram

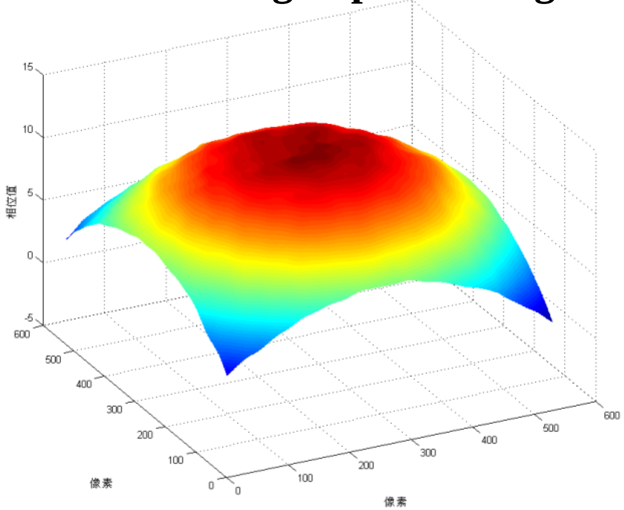
Profile and Defect Examination of Lens Surface

Testing results:

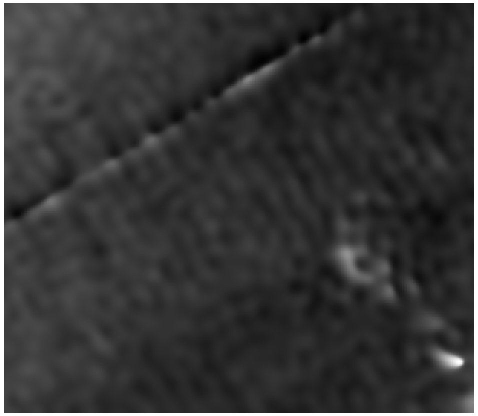
Phase image | **3D rendering of phase image**

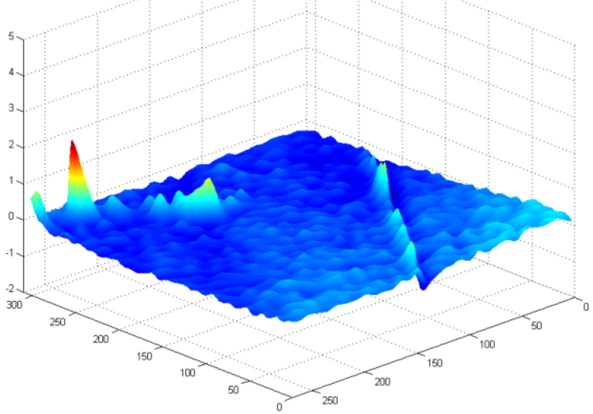
Profile:





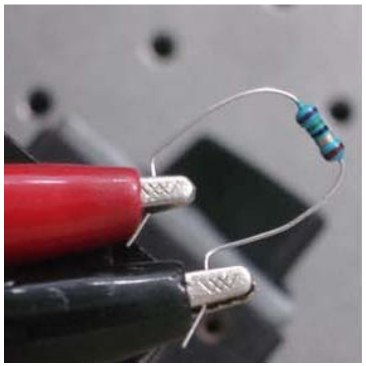
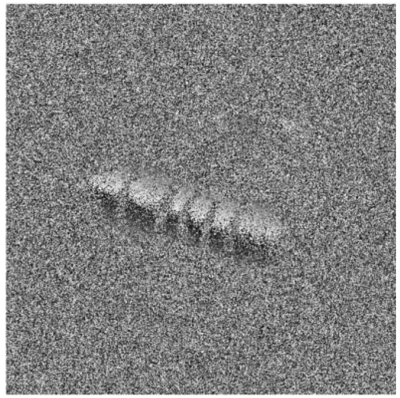
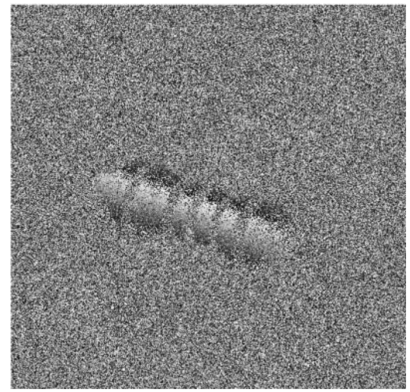
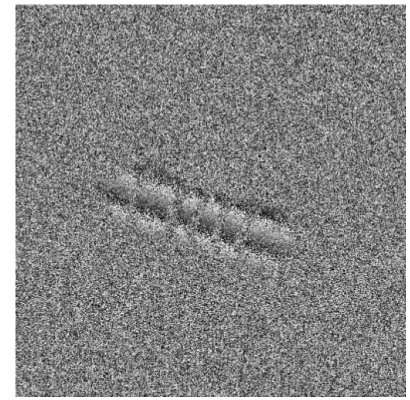
Defect:



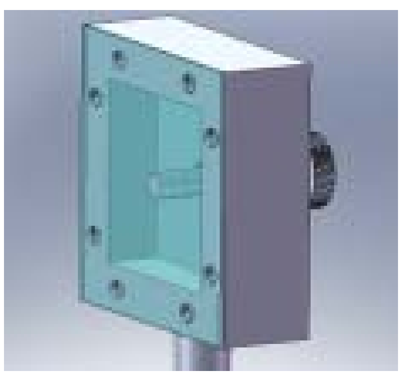
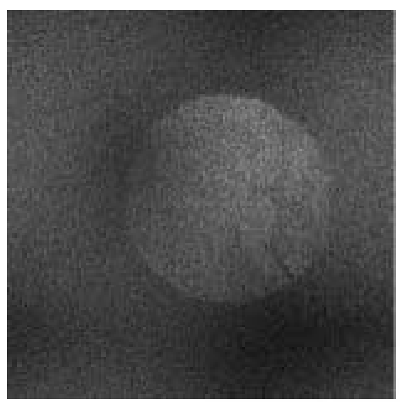
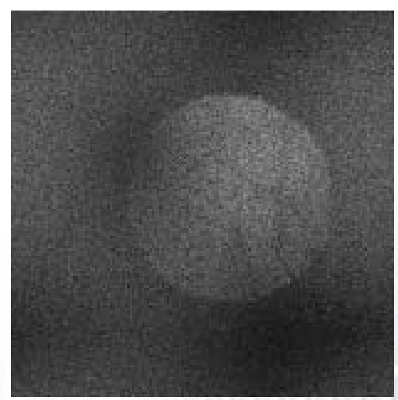
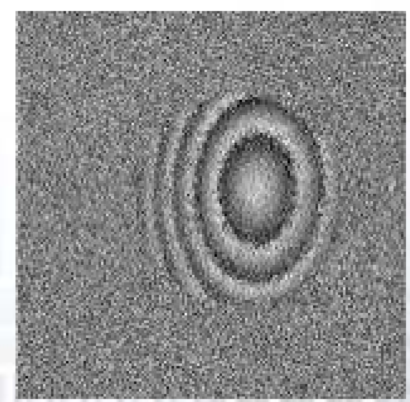


Fanjing Wang, Wen Xiao, Feng Pan, Shuo Liu, Lin Cong, Rui Li, Lu Rong. "Measurement of optic component by digital holography with least square method". (accepted by Optic laser Engineering)

Micro-deformation Measurement of the Resistance

Micro-deformation Measurement of the Aluminum

Conclusions

- Digital Holographic Microscopy (DHM) is a useful technique allowing for 3D in-situ measurement. It could be adjusted and applied in space.

- With many advantages, DHM is highly suitable for dynamic observation of biological procedures. It is competitive in the morphological analysis of cells and tissues.

- Although the resolution and image quality are restricted by CCD or CMOS, it would not be such a great problem because the performance of electronic devices will be remarkably improved in the future.

谢谢