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研究種目：基盤研究(A) (一般)

研究期間：2016～2018

課題番号：16H02218

研究課題名(和文) コヒーレントX線回折による酵母核内の核酸分布イメージング

研究課題名(英文) Visualization on the distribution of nucleic acids in yeast nucleus by using coherent X-ray diffraction

研究代表者

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交付決定額(研究期間全体)：(直接経費) 35,300,000円

研究成果の概要(和文)：シンクロトロン放射光を用いた低温X線回折イメージング・トモグラフィーと、X線自由電子レーザーを用いた低温X線回折イメージングにおける試料作製および実験装置の高度化をおこなうとともに、新たな構造解析法を考案した。それらをもとに、バクテリア細胞や酵母細胞核の普遍的な内部構造などを可視化するとともに、バクテリア細胞や酵母細胞の三次元構造解析を行い、生体非結晶粒子の非侵襲イメージングを確立した。

研究成果の学術的意義や社会的意義

低温X線回折イメージング・トモグラフィー実験では、細胞個々の特性や細胞周期に応じた物質分布の変化を、一方、XFELを用いるX線回折イメージング実験では、大量の細胞試料の投影電子密度を得ることが可能となった。本課題の研究成果は、シンクロトロン放射光とX線自由電子レーザーを相補的利用によって、細胞の個性と多様性を見ることにつながり、他のイメージング手法では成しえない非侵襲的イメージングの大きな進展につながることを期待できる。今後、本手法が細胞イメージングの重要な柱の一つとなるだけでなく、非結晶金属ナノ材料試料の構造研究にも裾野を広げていくと期待できる。

研究成果の概要(英文)：In this project, we improved fundamental techniques including automatic control procedures of diffraction apparatuses, preparation of frozen-hydrated biological specimens, and measurement of spatial coherence of X-ray beam, in cryogenic X-ray diffraction imaging tomography using synchrotron X-rays and cryogenic X-ray diffraction using X-ray free electron laser pulses. In addition, we developed methods to obtain reliable electron density maps in phase retrieval calculation in structure analyses including the utilization of manifold learning. Based on these developments, we successfully established X-ray diffraction imaging technique for non-crystalline biological specimens, such as cells and organelles, through the visualization of common structures in yeast nucleus and cyanobacterium cell as well as the three-dimensional structures of yeast and bacteria cells without sectioning and chemical labeling necessary in other imaging techniques.

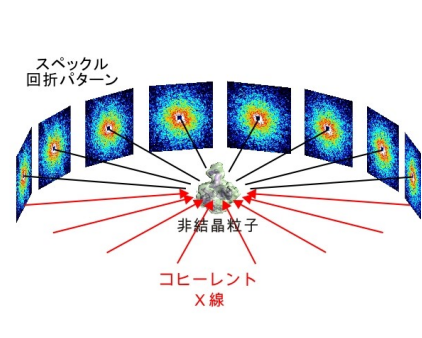
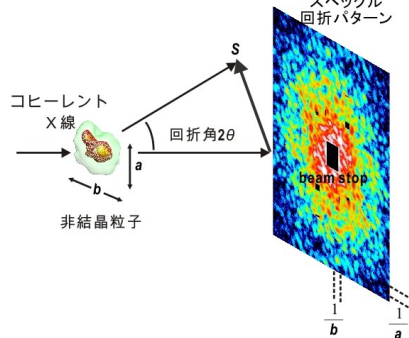
研究分野：生物物理学

キーワード：X線回折イメージング 放射光 X線自由電子レーザー 低温X線回折実験 位相回復 三次元再構成  
細胞核 細胞非侵襲イメージング

(EMXb18B0(b)g/S4kRb11  
 1vSMb0iMG6 □.2A □ ((b)g/S4N □ □ 6brrb  
 09MGSKK8)6(x □ □ 6brrb  
 □ 09KSCV2A68IMB7ELsb60q09A N.□  
 8 □ 6E812AITS/(b)K0b  
 □ 1 μm 100 nm Mb00b3M26d68 □  
 677K(S4E) □ 6 □ □

0j2b18  
 5006#H150K8M786k86 □  
 Kv □ (□ □ 54b) (xgE(188 □ □ 2 LAPM  
 M)zbSB46S60iKDs67b'g/S40 □ □  
 6G□(KM)z□ □ X-ray free electron laser: XFEL □ 98d(11)z  
 8 □ □ (M)zG □ □ X-ray diffraction imaging: XDI □ □ □  
 (b7u b □ 4 □ (x □ 100 nm 2z(00iMG □ □ KS □

1j2b2  
 XDI ε8S □ '56b98 (V)z □ ) □ 0q(UK  
 G#(36ISb) □ G24:K □  
 UV)zεKS0q7GM □ □ 1 e □ 9EM)z □  
 1M08bE67Gε0/6106  
 KK0qxH1b11MGOS406 □ rS0 □  
 UV)zPKG3IOG#p7K □ backprojection 2  
 0q7(xbUSB6 □ □ 1 □ □



□ 1 XDI 9b  
 0E □  
 b □  
 6 □  
 6 □ XFEL-XDI  
 εXb( □  
 X8 □  
 9E □  
 M(G3K □  
 G#E □  
 7M □

2j2B □ □ 1 □  
 2j □ XDI 9 □ 9 □ b □  
 G# □ 8) (bS48u000iM □  
 /(6K □ □ 12E □ C9# □  
 'g K1vK87 □ □ KS □

XDI 2#MSub9 □  
 p7M □ Su □ UXB/ □

2j □ □ □ 0q 80b9 □ □ □ 6, 10 □  
 0qεD □ 5 □ □ 10Mq(11MSuε(4h □ poly-lysine  
 (PLL) K1SE(4wV0q □ □ □ 5k )/13

D4)5 □ □ K# □  
 3)M □ □ 2 □  
 XFEL-XDI □ □  
 M b Su □ 7 (□ /10×10 μm<sup>2</sup>  
 b98kK □ S □ □  
 εM)z2M(M □  
 □ focused ion beam 88  
 0KSp0q(Cm8p'gK  
 S □ □ 12E □ 6827 □  
 426 K □ □ )b0q# □  
 0i □ KSG □ □ 10q  
 bDE □ AMIOG □  
 BKS □ □

for SR XDI tomo

for XFEL-XDI

□ 2 10q8Bm8pgK1S6×  
 82742610q09p □

XFEL-XDI 900 30 Hz j) 50 μm/33 ms

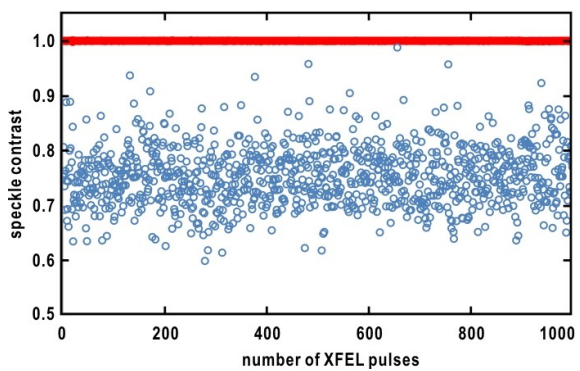
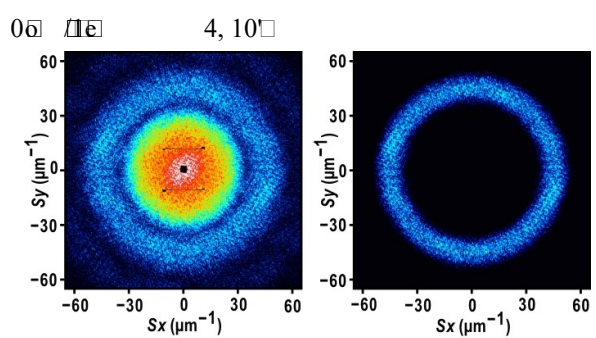
1, 6, 11 XFEL CMS

0q

8: G@DMS SACLA b3 Program Logic Controller XDI 900 CMS

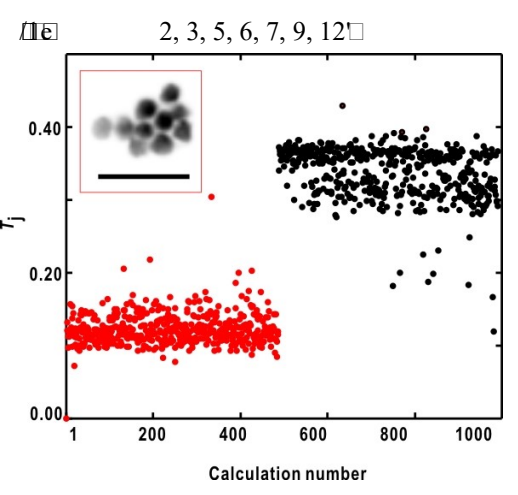
12 b0q SACLA b3 Program Logic Controller

XFEL 0 SACLA 30Hz j) HGK C06M 2014 SACLA b2 XFEL b56 0.7 K87KS 1.0 M SACLA 70i5 G2 K 7KS5 7K 7KS56 XFEL 8G1K S XFEL H0qbG K: xM Ib :U)z (V Ob'v S



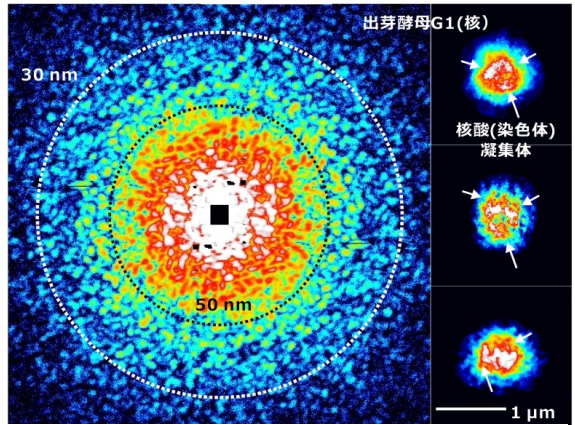
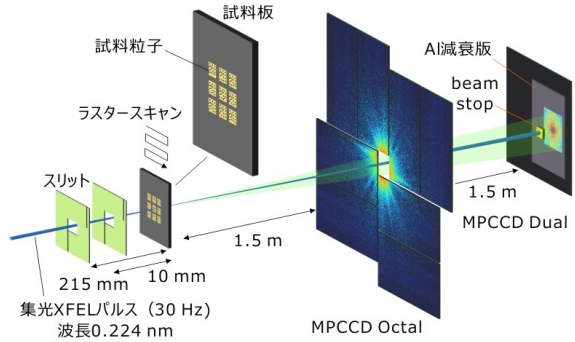
3 5 Sk0qbG Mel05)G bSu5M 2e27 Ob'vS56

2) 2)ivK87b4E G@b)G01@ SS8b K vGK87 0e8 1000 Gb)G01qv K87 4EM SuG 2k'566K b(x60 018M2CM 87 b6M X IC 40b K G 888 4 2IDG85V)z OS 4E bV IOS fG7b856 V) M2DK XFEL-XDI 900 6KSxb45(b574 B6x 4S45K GbbH)9N/AS 802SBH S



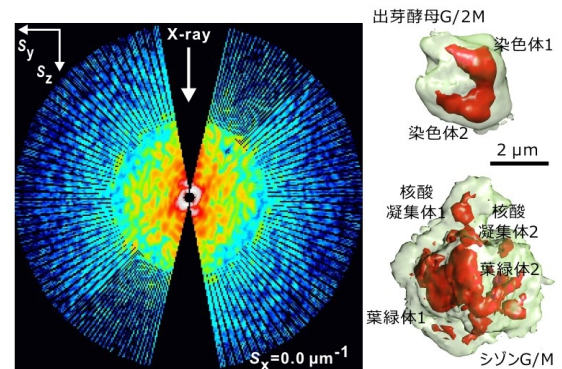
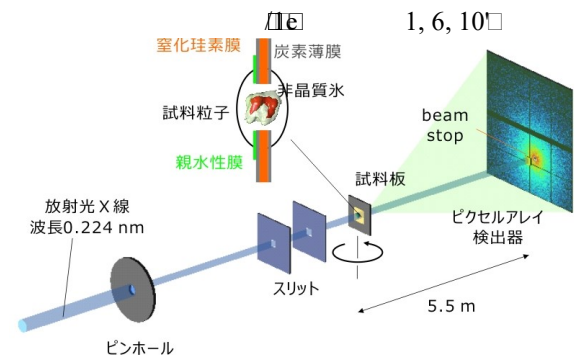
2) 0 XFEL-XDI 6484 b 0i 1e 6, 8, 10, 11, 2

multi-port CCD 8  
 500 nm 4)BKS  
 S0  
 T'6  
 6 46M  
 M1797 bG  
 MG  
 8  
 7  
 850 nm  
 8M  
 7  
 250 nm  
 7  
 250 nm  
 46  
 S  
 8  
 8  
 SIB28  
 MG  
 500 nm



5 XFEL-XDI 950M4  
 6M  
 )G

CXDI 9  
 SPring-8 BL29XU  
 4  
 U/  
 0  
 70 K  
 9  
 5 μm  
 6 μm  
 1.5  
 7  
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 PM  
 100 nm  
 U  
 100 nm  
 20 nm  
 x016G  
 S  
 50 nm



6 XDI  
 VG(M  
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○ <http://www.phys.keio.ac.jp/guidance/labs/nakasako/nakasako-lab.html>

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