

Effect of deuterium oxide on X-ray diffraction pattern in glycerinated skeletal muscle fibers by a CCD X-ray detector

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Introduction

Muscle contraction results from cyclic formation and breaking of cross bridges between myosin head and actin binding site. Energy for contraction is supplied by ATP hydrolysis. Since molecular weight of deuterium oxide D₂O is larger than that of H₂O, binding force and characteristics of reaction to other chemicals are different. D₂O increases force and stiffness by about 20% with slow rising phase, and decreases ATPase activity by 40-50% [1][2]. To obtain information about structure change, the effect of substituting D₂O for H₂O studied by X-ray diffraction.

Materials and Methods

The rabbit glycerinated muscle fibers were mounted isometrically into the experimental chamber at 2.4 μm of sarcomere length, and set to monochromatized X-ray beam path of wavelength 0.155nm from beam line 15A of synchrotron radiation. The two dimensional diffraction patterns from relaxed, rigor and Ca²⁺ activated fibers in D₂O were recorded by the CCD X-ray detector with force developments, and compared to those in H₂O. All experiments were made at room temperature.

Results

Figure 1. shows the diffraction patterns from relaxed muscle fibers in H₂O and D₂O. In relaxed muscle fibers, diffraction pattern in D₂O is much clearer than that in H₂O. Especially myosin layer lines appeared remarkably. These phenomenon occurred when sequence of H₂O and D₂O is changed. Figure 2. shows intensity profiles of 1,0, 1,1 equatorial reflection in (A) and 143, 215 meridional refraction in B from relaxed in H₂O, D₂O and contracting muscle fibers in D₂O. 1,0 intensity change in D₂O increased, but 1,1 changed little. Both 143 and 215 intensity changes in D₂O increased more than 2 times. Remarkable differences are not seen between rigor or contracting fibers in H₂O and D₂O. These results suggest that helical regularity of myosin head of relaxed muscle fibers in D₂O increases more

than that in H₂O. It might be related that in D₂O fraction of myosin head of M.ADP.Pi state increase.

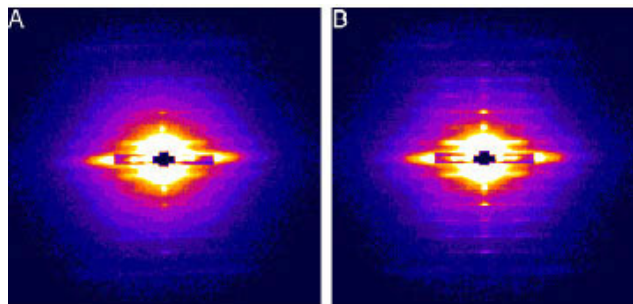


Figure 1. Diffraction patterns of H₂O (A) and D₂O (B) from relaxed muscle fibers.

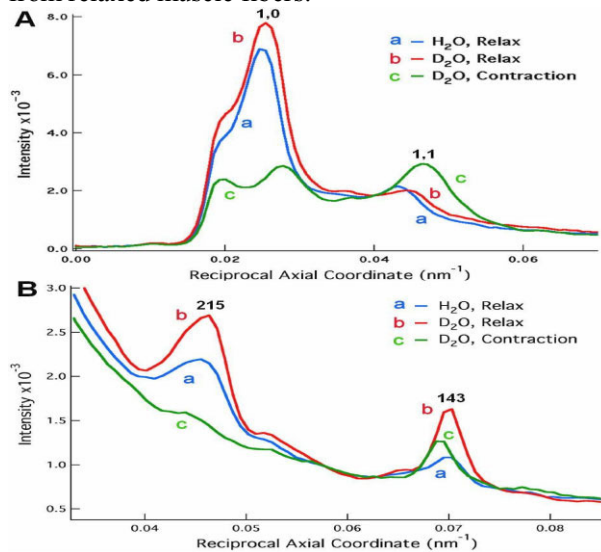


Figure 2. Intensity profiles of 1,0, 1,1 equatorial (A) and 143, 215 meridional refraction (B)

References

- [1]S.Chaen et al.,Biochim. Biophys.Acta 1506 (2001)
- [2]T.Kobayashi et al., Biochim. Biophys. Acta 1659 (2004)

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