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Alteration of the helical twist associated with the shortening of the thin actin filaments upon activation of skeletal muscles

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Introduction

Twisting changes of the actin helical structure associated with its filament extensibility upon activation of frog skeletal muscles stretched to non-overlap length of thin and thick filaments were investigated by measuring the axial spacing of the actin-based first layer-line component in the X-ray diffraction pattern. In the first layer-line complex, the thin and thick filament-based components could be resolved in the radial region close to the meridian, by a Gaussian deconvolution method [1]. We inspected the axial spacing change of the first actin layer line together with those of the actin 5.9-nm and 5.1-nm layer lines upon activation.

Experimantal

Living semitendinosus muscles of the bullfrog were used for this study. We measured 2D X-ray diffraction patterns from muscles stretched to non-overlap length of thin and thick filaments in resting and activated states. X-ray experiments were performed at BL15A. The 2D diffraction patterns were recorded on image plates. The specimen-to-detector distance was ~2m.

Results and Discussion

There appeared two layer-line components in the very inner radial region (0.020 R 0.039 nm^{-1}); one component located at ~1/38.0 nm^{-1}, almost the same axial level as the first troponin-related meridional reflection and the other sit at ~1/42.9 nm^{-1} which was the myosin-based first layer line. The axial spacing of the 38.0-nm component decreased by ~0.10% upon activation of an overstretched muscle, similar to the average spacing change of the troponin-related meridional reflections. Thus, this component originates from the troponin molecules, associated with the first troponin-related meridional reflection. In the next inner region (0.039 R 0.063 nm^{-1}), there existed a diffuse and weak component centered at ~1/36.6 nm^{-1}, which sit at the axial level higher than at 1/38.0 nm^{-1} and seemed to be the actin-based component. Upon activation it decreased in spacing by ~1.38% below its resting value. The 5.9-nm and 5.1-nm layer lines, corresponding to the pitches of the left- and right-handed genetic helices of the monomer in the actin filament, decreased respectively in the axial spacing by ~0.01% and ~0.15% below their resting values upon activation. The observation that the spacing change of the 5.1-nm reflection was larger than that of the 5.9-nm reflection was consistent with our previous reports [1][2]. Figure 1 summarizes the spacing changes of the inner and next inner regions of the first actin layer line together with those of the 5.9-nm and 5.1-nm actin layer lines. The spacing decrease of the first actin layer line estimated from those of the 5.9-nm and 5.1-nm layer lines was ~1.18% (a white bar graph), close to the observed spacing change of the 36.6-nm component, as above. The spacing decrease of the actin-based 2.7-nm meridional reflection estimated from those of the 5.9-nm and 5.1-nm reflections was ~0.09%. This was also very close to the observed change in our previous experiments where the overstretched muscles were activated [2][3]. These results confirmed that the shortening of the thin actin filament caused by the activation was closely associated with a twisting change of the right-handed helices of the actin filament.

References

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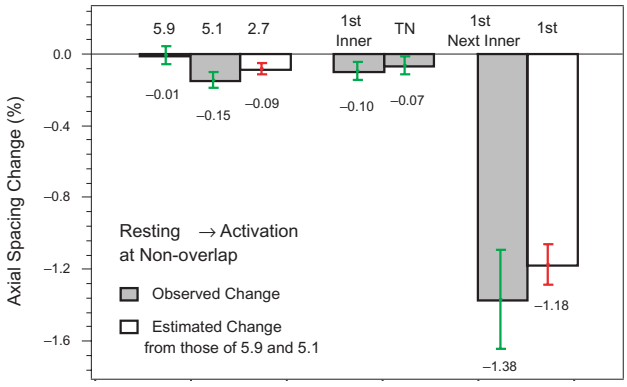

Fig.1 Spacing changes of the first actin layer-line components and the troponin meridional reflections (TN), together with those of the 5.9-nm and 5.1-nm layer lines.

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