FINAL TETANUS FORCE OF FROG SEMITENDINOSUS MUSCLE AFTER RAMP SHORTENING AT VARIOUS LENGTHS

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Abstract: Clarifying the dynamic properties of a muscle is indispensable to construct the model of the muscle. However, when the muscle was shortened during an isometric contraction on a descending limb where the muscle length was longer than the optimal length (*L***0), its properties were not fully clear. The purpose of this study is to investigate the** relationship between the final force (P_e) after muscle **shortening and shortening length on the descending limb A frog semitendinosus muscle was shortened from an initial muscle length (120% of** L_0 **) to four final muscle length (115, 110, 105 and 100% of** L_0 **).** *P***e was greater than the isometric force at the initial** length. P_e increased with an increase of the amount of shortening length up to 10% of L_0 . The peak of P_e **was obtained when the amount of shortening length was in 10% to 15% of** L_0 **.**

Introduction

It is necessary to figure out the dynamic properties of a muscle in order to understand a movement control of human's hand and foot correctly. Gordon et al. [1] investigated the relationship between a muscle length and an isometric force. As a result, the existence of a descending limb was showed. The descending limb is the region where the isometric force decreases with the increase of the muscle length. On the descending limb, it was known that a final force after a muscle stretching under a tetanical stimulation was greater than the isometric force at the final muscle length and increased with an increase of a stretching length [2 - 5]. On the other hand, Maréchal and Plaghki [3] showed that the final force after a muscle shortening under the tetanical stimulation was lower than the isometric force at the final muscle length after the muscle shortening. Schachar [6] pointed out that the final force increased with the increase of the shortening length as compared with the isometric force at the final muscle length after the muscle shortening.

We have focused on the final force, and studied on a comparison between the final force after the muscle shortening and the isometric force at the initial muscle length before the muscle shortening [7]. The relationship between the final forces after the muscle shortening and the shortening length was examined. From the pervious study, it was clarified that the final force depended on the shortening length. A change of the properties by a history dependant should be

considered in the case where the muscle length had a larger change. However, the unnormalized value was used as the shortening length in the study. For a quantitative comparison of different samples, the shortening lengths need to be normalized by the optimal length L_0 .

The purpose of this study is to investigate the relationship between the final force after the muscle shortening and the shortening length on the descending limb.

The force response was measured while a muscle ramp shortening under an isometric contraction at the various shortening lengths which were normalized by *L*0. 11 samples of the semitendinosus muscles of frog, *Rana catesbeiana* were used in an experiment.

Materials and Methods

Fig. 1 shows an experimental setup. Ringer's solution (normal saline solution of frogs, ingredients: NaCl 115.5 mM, KCl 2.0 mM, CaCl $_2$ 1.8 mM and NaHCO₃ 2.0 mM) was filled to the glass chamber (10 x) 50 x 10 mm). It was circulated using the roller pump (FURUE SCIENCE RP-NE2) and always maintained at the fresh state. The solution's temperature was kept at 3.8 (± 0.7) °C using the cooler (BIOCRAFT MODELC-1).

The muscle was fixed using the actuator and the manipulator of the mechanical stimulator (DIA MEDICAL DPS-270). A pair of electrodes (0.5 x 50 x 10 mm) made of platinum was arranged on the sides in the chamber. The electric stimulation (amplitude: 300 mA, pulse width: 0.4 ms, interval of pulses: 70 ms) was given to the muscle using the electric stimulator (DIA MEDICAL DPS-1100D). The force and the muscle length were measured using the tensiometer (resolution: 2.0 mN) attached in the actuator, and the linear displacement gage (resolution: 0.05 mm). They were recorded to the computer (NEC PC9821FA, MS-DOS).

In this experiment, the force was normalized by the maximum isometric force P_0 , and the muscle length was normalized by the optimal length L_0 . The descending limb was determined the region where the muscle length was longer than L_0 (> 100% of L_0).

Figure 1: Experimental setup

After 2.3 seconds from the beginning of the electric stimulation, the muscle under the isometric contraction was shortened at a constant velocity (20% of L_0 /s) from the initial muscle length L_i (120% of L_0) to the final muscle length L_f on the descending limb. The muscle shortening had four shortening lengths (5% of L_0 , 10% of *L*0, 15% of *L*0, and 20% of *L*0).

The treatment of animals used for this experiment was followed 'Guiding principles for the care and use of animals in the field of physiological sciences (Physiological Society of Japan, 2003)'.

Results

Fig. 2 shows a typical of the experimental results. In Fig. 2, the axis of ordinate represents the force, the muscle length, and the electric stimulation in order, and the axis of abscissa represents time. The curve (a) denotes changes of the force and the muscle length with time when the amount of the muscle shortening is 5% of *L*0, (b) 10% of *L*0, (c) 15% of *L*0, and (d) 20% of *L*0. The final force (P_e) was measured at (a)', (b)', (c)' and (d)' with each length.

 P_e at (a)' was greater than the isometric force at L_i (P_{isom, L_i}) . Also in (b)', (c)', and (d), the same result was obtained as (a)'.

Fig. 3 shows the relationship between the force and the muscle length in Fig. 2. In Fig. 3, the axis of ordinate represents the force, and the axis of abscissa represents the muscle length. A green line indicates the isometric force - the muscle length relationship, and a red line indicates the relationship between P_e and the shortening length.

A force response in the case of (c) was the following progress; (I) the muscle was at a resting state, and (II) the force reached a steady state at 120% of L_0 . Then, after the start of the ramp shortening, the force decreased as indicated by the arrow. (III) The force increased again when the muscle length amounted to 105% of L_0 . Finally, (IV) the force reached the steady state. P_e increased in order of (a), (b), and (c). The peak of P_e was obtained at (c).

 P_0 of this muscle was 272.1 mN, and L_0 of it was 19.8 mm. The nearly same results were obtained from the other muscles.

Figure 2: Changes of force and muscle length with time

Figure 3: Relationship between force and muscle length during muscle shortening at various lengths

Discussion

In Fig. 3, it was shown that the muscle length at which the peak of P_e was obtained was longer than L_0 . We carried out also the experiment where the initial muscle length was 130% of L_0 . The same results were obtained when the initial muscle length was 130% of *L*0. This showed that the muscle length at which the peak of *P*e was obtained didn't depend on the initial muscle length before the muscle shortening.

There were few reports concerning to the muscle shortening experiments. In this study, the final force was measured when the muscle was shortened under the contraction. It was shown that the relationship between the final force after the muscle shortening and the isometric force at the initial muscle length before the muscle shortening on the descending limb. This relationship has not reported previously.

In Fig. 3, the final force actually increased as shown (a), (b) and (c). This result was indicated that the active muscle had the properties of a negative stiffness. In previous studies, some researchers reported that the final force after the muscle stretching under the contraction was greater than the isometric force measured at the initial muscle length [2, 3, 4, 5]. These properties were a positive stiffness like a elasticity. The results from the previous and this study suggested the active muscle had the properties of both the positive and negative stiffness.

A joint angle of human's the upper and lower limbs is controlled stably by the flexor-extensor muscle pair *in vivo*. Therefore, it is indispensable to consider the dynamics of the flexor-extensor muscle pair in order to understand the movement control of human. On the descending limb, there were three hypothesises as a mechanism which indicated the instability; the sarcomere length nonuniformity [8], an accumulation of fatigue [9] and an engagement of cross bridge [3]. However, none has been proved yet. The dynamics of

the flexor-extensor muscle pair may concern this phenomenon.

Fig. 4 (a) shows the flexor-extensor muscle pair, and (b) shows the structure of the structure of the mechanical model. The extensor muscle has the stiffness K_e , and the flexor muscle has the the stiffness *K*f.

Figure 4: (a) Flexor-extensor muscle pair; (b) Structure of mechanical model

The dynamic equation of this model is given by eq. (1).

$$
F = (K_e + K_f)X + (D_e + D_f)\dot{X} + M\ddot{X}
$$
 (1)

where *X* denotes a displacement, *F* denotes an external force, *K* denotes the stiffness, *D* denotes a viscosity, and *M* denotes a mass.

Note that the muscle has the positive stiffness the muscle length which is longer than the initial muscle length [2 - 5]. On the other hand, it has the negative stiffness at the muscle length which is shorter than the initial muscle length as showed in Fig. 3. The stability depends on the total stiffness $(K_e + K_f)$. In the case that the total stiffness is the positive value, *X* is stable. However, *X* is unstable when the total stiffness is the negative value. Thus it is necessary to concern the result obtained in this study in order to construct the model of the flexor-extensor muscle pair.

From the above discussion, the simulation experiments in the flexor-extensor muscle pair will be the important future works.

Conclusions

At the region where the muscle length was longer than the optimal length (L_0) , the muscle under the tetanic contraction was shortened. 11 semitendinosus muscles of frog were used in this experiment. The initial muscle length L_i was 120% of L_0 , and the shortening speed was 20% of L_0 /s. The shortening lengths were changed with 5% of *L*0, 10% of *L*0, 15% of *L*0, and 20% of L_0 . The final force (P_e) in each shortening lengths

was measured. The following results were obtained from this experiment.

- P_e was greater than the isometric force at the initial muscle length before a muscle shortening in the case of 5% of L_0 , 10% of L_0 shortenings.
- P_e increased with the increase of the shortening length up to 10% of L_0 .
- The peak of P_e obtained at the shortening length, in 10% of L_0 to 15% of L_0 .

It was suggested that the relationship between the final force after the muscle shortening and the shortening length was an important point to clarify the dynamics of the flexor-extensor muscle pair.

The future works are to perform the simulation experiment using the model of the muscle considering the flexor-extensor muscle pair, and to make the contraction dynamics of the muscle clear.

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