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Hypothesis

The role of the skeletal muscle myosin light chains N-terminal fragments

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Abstract The myosin regulatory and essential light chains in skeletal muscle do not play a role as significant as in scallop or smooth muscle, however, there are some data suggesting that the skeletal myosin light chains and their N-terminal parts may have a modulatory function in the interaction of actin with myosin heads. In this paper four conformational states of the myosin head with respect to the regulatory light chain bound cation (magnesium or calcium) and phosphorylation are proposed. Communication between regulatory and essential light chains and putative binding of the N-terminus of A1 essential light chain to actin is discussed.

Key words: Myosin light chain; Actin-myosin interaction

1. Introduction

Since publication of the work of Rayment et al. [1] describing the crystallographic model of myosin head, the question of the localization of myosin light chains within the structure of the head seems to have been answered in an exact way. The light chains are enveloping the long α-helix of the 20 kDa domain. The essential light chain (ELC) is in contact with the parts of the molecule constituting the active site region, and the regulatory light chain (RLC) is bound to the C-terminal part of the heavy chain of subfragment-1. The fragment of the skeletal muscle myosin head which contains the light chains can be called, by analogy with that of scallop myosin, the 'regulatory domain', in contrast to the rest of the head, called the 'motor domain', containing the active site and actin binding surface. The regulatory domain of scallop myosin has been crystallized and its three-dimensional structure has been proposed [2]; it appears to resemble roughly that of the 'regulatory domain' of chicken skeletal muscle. All three components of the myosin head, the heavy chain and both regulatory and essential light chains are necessary to achieve regulation of the scallop myosin activity [3]. In scallop myosin the specific (triggering) calcium binding site is located in the essential light chain. The calcium binding site on ELC is stabilized by the interaction with heavy chain and RLC. This site is absent in vertebrate striated muscle ELCs. In the mechanism of force generation by a crossbridge proposed by Rayment et al. [4] the information about ATP hydrolysis is transmitted to the regulatory domain and the relative movement of this domain (bending of the head, lever arm mechanism) leads to the generation of movement. The reverse process does also occur: the information about the calcium binding to the ELC in scallop myosin and phosphorylation of the smooth muscle RLC is transmitted to the motor domain of myosin and regulates its activity. In the skeletal muscle myosin the regulatory domain has a distinctly less important role than in the scallop or smooth muscle myosin, however, the phosphorylation of RLC and the cation binding to RLC and the isoform of ELC present in the heads have modulating effects on the mechanical and biochemical properties of the myosin-actin interaction and force generation. The regulatory domain influences the function of the motor domain also in vertebrate striated muscle.

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2. Physiological role of phosphorylation of the vertebrate striated muscle myosin light chains and cation replacement in RLC

The phosphorylation of RLC in vertebrate striated muscle does not act as a trigger of contraction, as it does in smooth muscle, but rather has a modulatory function. This subject has been reviewed thoroughly by Sweeney et al. [5]. During contraction the myosin light chains phosphorylation level depends on the activities of two enzymes: myosin light chain kinase and phosphatase. The activity of kinase supersedes the activity of phosphatase; therefore prolonged stimulation of muscle or low frequency stimulation leads to an increase of the phosphorylation level which decreases relatively late after the stimulation has ceased. The phosphorylation has certain modulatory effects on the contracting muscle. It correlates with potentiation of the maximal extent of the isometric twitch tension [6]. It also increases the isometric tension produced in skinned fibers at submaximal Ca^{2+} concentrations [7] and increases the rate of force development [8]. Sweeney et al. [5] have postulated that RLC phosphorylation acts in vertebrate striated muscle as a factor enhancing the performance of muscle, a kind of 'memory' that the muscle has recently been active. The RLC phosphorylation enhances the force if the muscle was previously activated and allows the force to be maintained at a certain level even if, during prolonged contractions, the Ca^{2+} concentration becomes decreased.

In addition to phosphorylatable serine RLCs have another site which can be recognized as a potential regulatory site. This is the calcium binding EF-hand domain I which binds the divalent cations Mg^{2+} and Ca^{2+}. During activation of muscle these sites can be, at least partially, saturated by Ca^{2+} despite high physiological levels of Mg^{2+} [9–11]. Although the process of exchanging of the RLC bound Mg^{2+} (relaxation) for Ca^{2+} (activation) is too slow to account for quick development of force [12] during activation of contraction some binding of Ca^{2+} may occur under conditions of prolonged activation. It might

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be possible that, due to intermolecular cooperativity within the thick filament [13,14], even incomplete saturation by calcium might be sufficient for changing the properties of a majority of myosin molecules. Since the binding of Mg^{2+} and Ca^{2+} to isolated RLC causes different conformational changes [15] one could expect that the differences in conformation of the myosin head complex would lead to a change in contractile properties dependent on the cation bound to RLC. Metzger and Moss [11] investigated the basis of Ca^{2+} sensitivity of the weak to strong crossbridge transitions in mammalian skeletal muscle. They found that the rate of force development increased with Ca^{2+} concentration in the physiological range. This effect was related to the RLC. These findings confirm the role of RLC as a part of a second system of Ca^{2+}-dependent regulation which does influence the parameters of contraction.

3. The presence of isoforms of essential light chains and their physiological significance

In the vertebrate skeletal fast muscle two isoforms of essential light chains, A1 and A2 are present. Both of them are products of alternative splicing on a single light chain gene [16]. A1 is longer than A2. In the rabbit fast muscle A1 has the same amino acid sequence as A2 over 141 C-terminal residues (Fig. 1a). An additional fragment of 41 amino acid residues is present at the N-terminus. Between the sequence common to both isoforms and the N-terminal fragment there is an eight residues long homologous sequence with five amino acid residues substitutions. The biochemical differences between the myosin isoenzymes have been thoroughly studied both with whole myosin and its soluble fragments S1 containing only the myosin head. Pope et al. [17] and Pastra-Landis et al. [18] found that the presence of a particular essential light chain did not affect the steady state rate of ATPase activity of myosin in the presence of actin under conditions approximating physiological ones. In the case of S1 it was found that the affinity of S1(A1) for actin was significantly higher than that of S1(A2) whereas the V_{max} for S1(A1) was two-fold lower [19–21]. However, this difference disappeared with an increase of ionic strength toward more physiological values [22]. It has been also observed that S1(A1) polymerizes G-actin more efficiently than does S1(A2) [23]. In rigor conditions A1 both in S1 and HMM can be crosslinked to actin by an zero length crosslinking agent [24,25]. Additionally, it was shown by NMR studies that the N-terminal of A1, which is mobile in solution became immobilized in the presence of actin [26]. Further NMR studies showed that the N-terminal

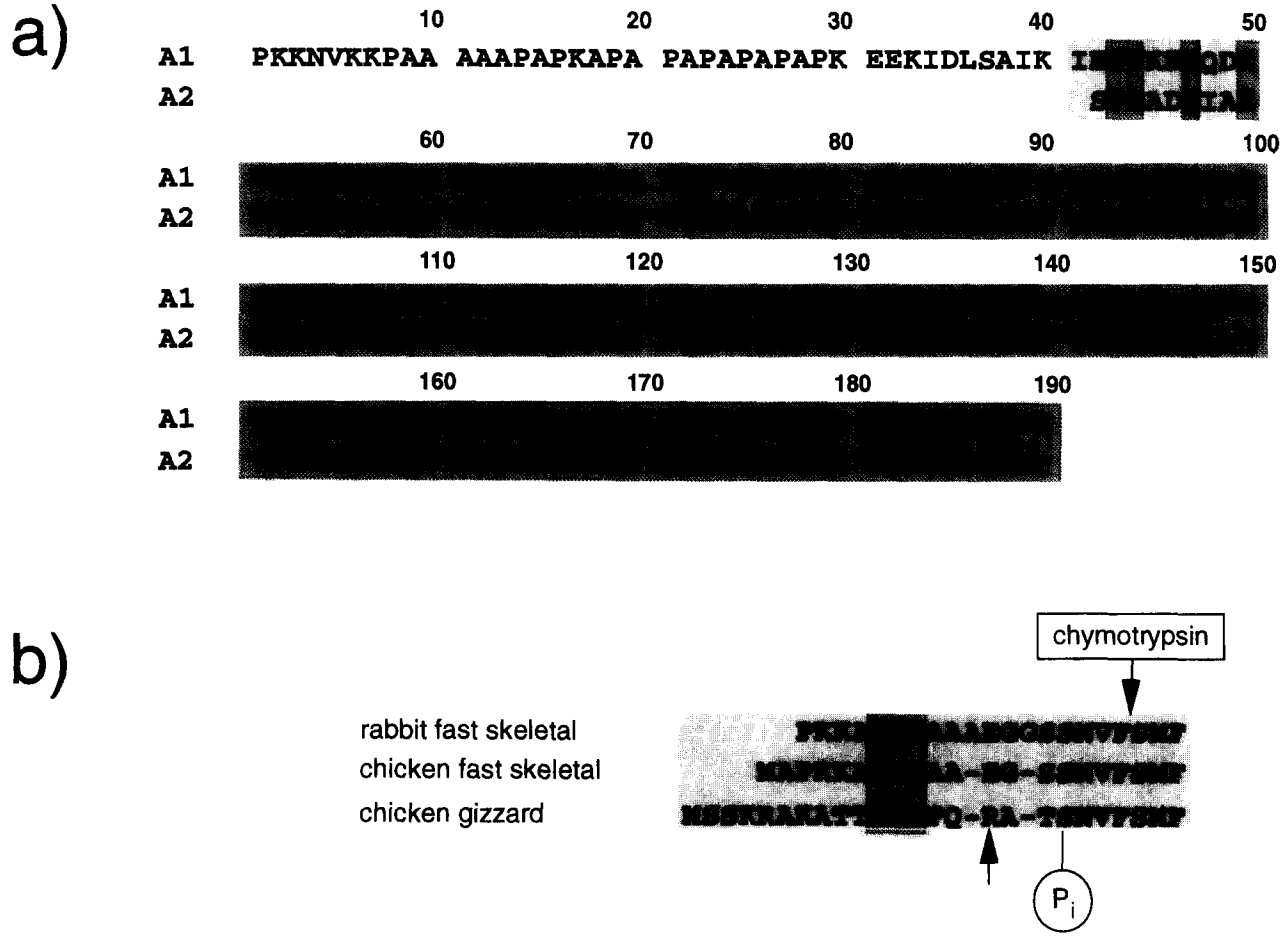

Fig. 1. Sequence alignment of the rabbit fast skeletal muscle essential myosin light chains isoforms (a) and of the N-terminal parts of regulatory light chains (b). Sequences of the essential light chain isoforms, A1 and A2 are aligned and the homologous fragments are placed in boxes. N-Terminal parts of RLCs from rabbit fast skeletal, chicken fast skeletal and chicken gizzard muscles are shown in b). The cluster of basic residues is indicated by the box. Arginine-16 in gizzard RLC, important for the myosin light chain kinase activity, and chymotryptic cleavage site in rabbit RLC are indicated by arrows. The position of phoshorylatable serine is also marked.

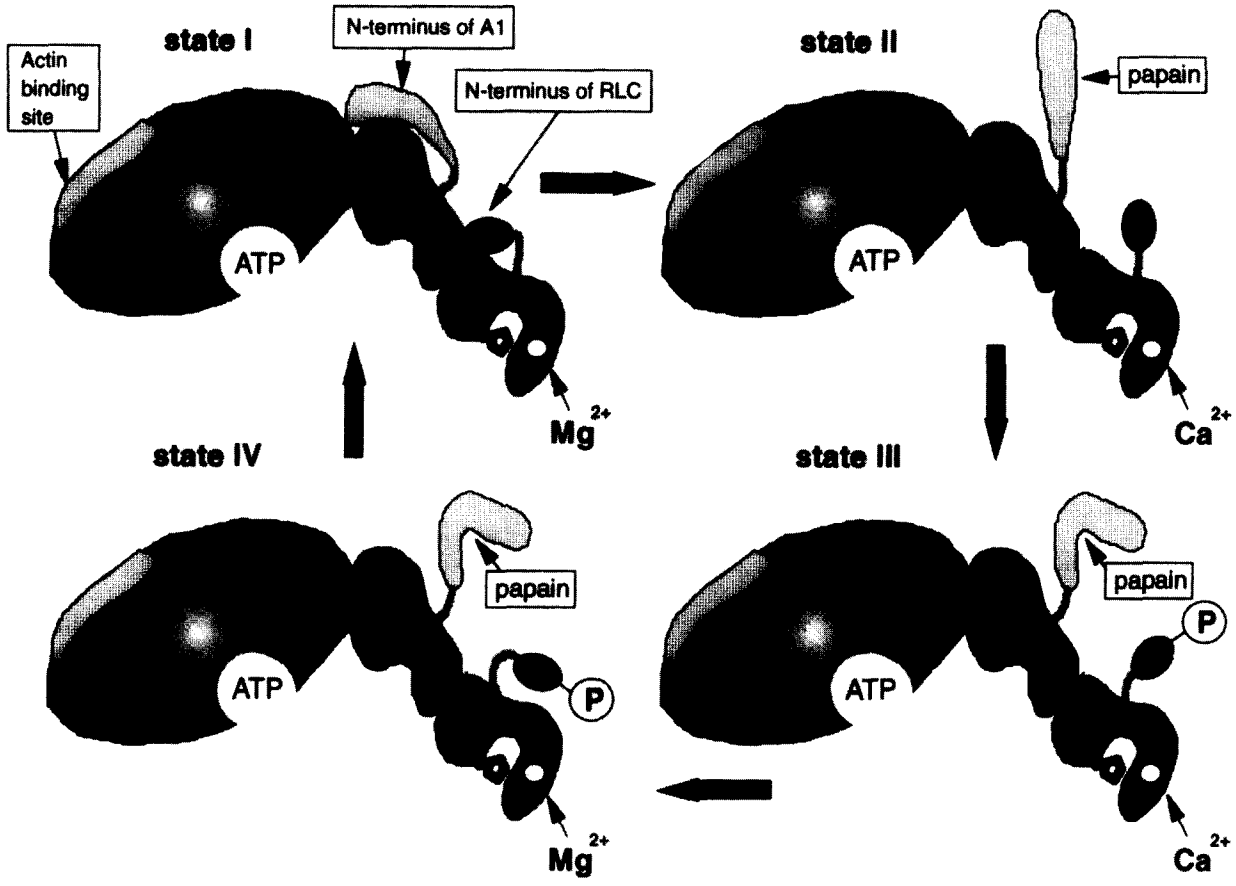

Fig. 2. The scheme of four conformational states of myosin head in respect to RLC phosphorylation and RLC bound cation. The atomic coordinates of α-carbons from chicken skeletal myosin subfragment-1 [1] (obtained from the Protein Data Bank – Brookhaven National Laboratory) were used to build the backbone model. The model was then rotated to obtain a view with exposed N-terminal fragments of light chains. The contours of the motor and regulatory domains denote the shape of the molecule. Long heavy chain C-terminal α-helix is shown to illustrate the connection between motor and regulatory domains. Since, in the crystallographic structure, information about location of the N-terminal fragments of A1 and RLC is absent, a graphic representation of the lacking parts, corresponding roughly to their size, is proposed. The scheme describes the conformational changes of only the N-terminal parts of light chains conveying the conception of the structural changes rather than actual bending or straightening of the termini. To make the presentation more clear neither the conformational changes of whole RLC, A1 or the motor domain occurring due to RLC phosphorylation and replacement of the RLC bound cation, nor the conformational changes occurring during ATP hydrolysis by myosin head, are shown. State I – occurs in the relaxed muscle and in the very first moments after activation. The N-terminus of A1 is not exposed to papain cleavage. State II – occurs in the first period of activation. The N-terminus of A1 is strongly exposed to papain cleavage. State III – occurs during prolonged activation (level of phosphorylation reaches its highest values). The N-terminus of A1 is exposed to papain cleavage. State IV – occurs in the muscle cell with decreased Ca^{2+} concentration after further prolonged stimulation. The N-terminus of A1 is exposed to papain cleavage. The extent of exposition of the N-terminus of A1 for papain cleavage in the presence of actin and ATP was determined from the data described by Stępkowski et al. [38].

region of A1 interacts directly with the C-terminal region of actin [27]. However, most of these data differentiating S1(A1) from S1(A2) were obtained at very low ionic strength. Both the crosslinking and interaction of A1 with actin seen in the NMR experiments were diminished with the increase of ionic strength [27,28]. More convincing evidence on the differences between A1 and A2 isoforms came from velocity measurements. Lowey et al. [29] tested, in the motility assay, myosin which contained either A1 or A2 isoforms of ELCs and found that the A2 light chain containing myosin translocated actin filaments with a velocity almost twice as high as the A1 containing myosin. These in vitro results correspond to the physiological studies where a correlation was found between the presence of A1 or A2 isoforms and the shortening velocity of isolated skeletal muscle fibers [30,31]. The fibers with a relatively higher A2 content shortened faster than those containing more A1. Chronic low frequency stimulation of fast rabbit skeletal muscle led to an increase in the A1 light chain content concomitant with a decrease in A2 which suggested adaptation to slower working conditions [32].

The major difference found between the properties of the A1 and A2 containing isoforms of myosin is the difference in the velocities of contraction rather than differences found in the properties of myosin in solution. It is clear that the N-terminal fragment of A1 can be responsible for the observed differences possibly due to the interaction of A1 N-terminus with actin. Such an interaction has been postulated in three dimensional reconstruction of the actin filaments decorated with S1(A1) [33]; however, in the actin filament–myosin head models based on crystallographic data [4,34] such an interaction seems to

be impossible. This discrepancy needs further elucidation. Andreev [35] observed crosslinking of the A1 N-terminus to actin at high actin to S1 ratio but not at full decoration. Since, in living muscle, actin is not fully saturated by myosin heads it might have the freedom to interact with the N-terminus of A1.

4. Significance of the N-terminal region of A1 in the actin–myosin interaction

Only one, the longer isoform of the essential light chain is present in cardiac myosin. Margossian et al. [36] found that, in S1 preparations obtained by papain digestion of cardiac myosin (with the N-terminus of A1 cleaved off, referred to as A1′), reassociation of intact A1 with the A1 deficient S1 caused increased affinity and increased V_{max} in comparison with preparations of S1A1′ (with shorter A1). However, in the experiments with the rabbit cardiac myosin which had been treated with papain to obtain preparations with the N-terminus of A1 digested off (but with the preserved intact heavy chain) Moczarska and Kąkol [37] observed an increase by one order of magnitude of the myosin affinity to actin. Similar results were obtained by Stępkowski et al. [38] for the rabbit fast skeletal muscle myosin, indicating that removal of the N-terminal residues of A1 leads to a strikingly increased affinity of myosin filaments towards actin. Such a behaviour was not observed with the myosin enriched in the shorter isoform of the essential light chain, A2 [20], suggesting that, the A1′-containing preparation is different from the A2 containing preparation. The removal of the N-terminus of A1 in myosin leads to destabilization of the head with time as observed by reduced K^+ ATPase activity [38]. The destabilization could result from dimerization of heads observed by Margossian et al. [36] for the cardiac papain S1 and by us for the rabbit cardiac and skeletal myosin (unpublished observations).

The function and role of the N-terminal part of A1 was also studied in several laboratories with the use of antibodies. An antibody directed against the N-terminal part of A1 caused a four-fold decrease of the cardiac myofibrillar ATPase activity [36]. Boey et al. [39] using an antibody against the N-terminus of A1 in skeletal myosin achieved a complete inhibition of the S1(A1) actin-activated ATPase whilst the activity of S1(A2) was not affected; however, the antibody bound also to A2. The ATPase activity of myosin and the rate of superprecipitation were also inhibited; surprisingly, the affinity of S1(A1) to actin, when studied at very low ionic strength, was not affected by antibody. Morano et al. [40] studied the influence of a monoclonal antibody against the N-terminus of ventricular human A1 on the isometric force of contraction of chemically skinned left ventricular fibers. An about 30% increase of the isometric force was observed at high calcium concentrations. Another approach used by Morano et al. [40] to study the role of the N-terminus of LC1 was the addition of a synthetic peptide corresponding to the N-terminal sequence of human cardiac A1 (amino acid residues 5–14) to the skinned ventricular fibers, which resulted in a 50% increase in the isometric force at submaximal calcium concentrations (pCa 5.5).

The N-terminal region of A1 seems to play an important role in the actin–myosin interaction in vitro; however, its influence observed in the experiments with the use of S1 was different than in those using myosin. It is not clear at present what could be the exact role of the A1 N-terminus binding to actin, if it does occur in living muscle. One possibility is a modification of kinetic parameters of the ATP hydrolysis cycle leading to slowing down of the velocity of shortening or velocity of actin filaments in the motility assay.

5. The role of light chains and their N-terminal parts in the mechanism of energy conversion by the skeletal muscle myosin head

The three-dimensional structure of myosin head implies that light chains, both essential and regulatory, may play a role in the overall mechanism of conformational changes of the myosin head leading to the generation of force. This issue as concerns skeletal, smooth, scallop muscle and non-muscle myosins has been reviewed by Trybus [42].

Recently, VanBuren et al. [43] reported that, in the single actin filament force measurements, skeletal muscle myosin with removed essential light chains produced half of the isometric force of control myosin. The force production ability of myosin with removed RLC was almost unchanged. However, as reported previously, the removal of either RLC or ELC or both had a significant slowing down effect on the velocity of actin filaments in the motility assay (unloaded conditions) [44], suggesting that both classes of light chains contribute to the conformational changes of the skeletal muscle myosin head involved in generation of movement or force. There are two lines of evidence which suggest direct communication between RLC and A1 light chains. The monoclonal antibody binding in a Ca^{2+}-dependent manner to the ELC, demonstrated by Shimizu et al. [45] may suggest that light chains undergo conformational changes when one of them (presumably RLC) binds Ca^{2+} or Mg^{2+}. Stępkowski et al. [38] has found, for the rabbit skeletal muscle heavy meromyosin and myosin, that limited proteolysis of A1 by papain is dependent both on the cation bound to RLC and the phosphorylation state of RLC. In both cases, those of binding of antibody to A1 or limited proteolysis of the N-terminus of A1, a close direct conformational communication occurs between the two light chains A1 and RLC, involving probably also the heavy chain.

Since such a communication does occur, it is reasonable to assume that, in the working muscle, the state of phosphorylation of RLC and its saturation either with Ca^{2+} or Mg^{2+} may affect the possible interaction of the A1 N-terminus with actin. This may be one of the ways of modulating the actin–myosin interaction dependent on the level of free Ca^{2+} ions in the sarcoplasm. This mechanism may apply as well to the cardiac and slow skeletal muscle where only the longer isoform of the essential light chains is present.

The changes of RLC state (cation bound and phosphorylation) influence also the motor domain of myosin head and its interaction with actin. The following effects were observed.

(a) The rate of tryptic digestion of the myosin heavy chain in the 20/50 kDa junction in the presence of actin was dependent both on phosphorylation and the kind of cation saturating the regulatory light chain (Mg^{2+} or Ca^{2+}) [46].

(b) The conformational state of the actin filament in rigor complex with heavy meromyosin was found to depend on the RLC saturating cation and phosphorylation. The existence of two states of rigor complexes of F-actin and heavy meromyosin, flexible and rigid, has been postulated [47].

The N-terminal part of RLC is involved in the transmission of conformational changes of RLC to the A1 or actin binding site since its removal leads to desensitization of the above described phenomena (a and b) to cation exchange [48]. Wagner [49] reported that the N-terminal part of RLC and hinge region are necessary for the calcium-dependent heavy meromyosin binding to regulated actin. Thus the N-terminus of RLC in the fast skeletal muscle myosin is involved in communication between the regulatory domain and the motor domain. Similarly, the N-terminal part of smooth muscle RLC is important for several properties of smooth muscle myosin. Extensive research allowed to link specific sites in the aminoacid sequence to 10S-to-6S conformational transition (ability to form filaments) and the stimulation of ATPase activity [50] and to myosin light chain kinase requirements for substrate recognition [51,52]. Similarly like in the case of skeletal muscle RLC, removal of N-terminal sequence of smooth muscle RLC destroyed certain functions of the smooth muscle myosin [50,51]. Despite some differences in sequence between the skeletal and smooth muscle RLC (especially Glu^{11} in position of Arg^{16} – important for kinase substrate recognition in smooth muscle RLC [51,52]; Fig. 1b) the N-terminus of RLC has a significant role in determining the properties in both types of myosin.

It can be postulated that both the N-terminal region of A1 and N-terminal region of RLC play an important role in the modulation of the skeletal muscle actin-myosin interaction by responding to the phosphorylation of RLC and magnesium-for-calcium exchange in the RLC cation binding site located in the EF-hand domain I, and that these effects require the hinge region of the myosin molecule. Taking into consideration the facts described in this paper, the following scheme of the influence of light chains on the interaction of myosin with actin in working muscle is proposed: The postulated four conformational states of the myosin head with respect to phosphorylation of RLC and the RLC bound cation are summarized in Fig 2. Transition from state I to II would influence the parameters of contraction (rate of force development) [11]. Transition from state III to IV (both states phosphorylated and with exposed N-terminus of A1) caused by a decrease of the free calcium concentration and replacement of Ca^{2+} by Mg^{2+} in the RLC's EF-hand domain would lead to maintenance of high isometric tension [7]. The elevated phosphorylation level would also cause a post tetanic potentiation [6] as compared to state I where the N-terminus of A1 is not exposed by actin. According to this model the phosphorylation causes desensitization of conformational changes of the myosin head with respect to the exchange of the RLC bound cation. The key process in the molecular mechanism by which light chains influence the interaction of myosin head with actin is the transmission of information to the essential light chain and the motor domain about the phosphorylation state of RLC and the saturating cation. The longer isoform of the essential light chains present in the fast skeletal muscle in parallel with the shorter isoform, and present as the single form in the slow skeletal and cardiac muscle, may take part in the interaction of the myosin head with actin through the A1 N-terminal part. This process seems to be sensitive to calcium concentration and the level of RLC phosphorylation. Such a mechanism may act as a fine modulator of the muscle performance adapting it to changing functional demands. The presence of the light chains isoforms in different types of muscles with different requirements for work output suggests that the existence of such a mechanism is very probable.

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References

[1] Rayment, I., Rypniewski, W.R., Schmidt-Base, K., Smith, R., Tomchick, R. D. Benning, M.M. Winkelmann, D.A., Wesenberg, G. and Holden, H. (1993) Science, 261, 50–58.
[2] Xie, X., Harrison, D.H., Schlichting, I., Sweet, R.M., Kalabokis, V.N., Szent-Gyorgyi, A.G. and Cohen, C. (1994) Nature 368, 306–312.
[3] Kwon, H., Goodwin, E.B., Nyitray, L., Berliner, E., O'Neall-Hennessey, E., Melandri, F.D. and Szent-Gyorgyi, A..G. (1990) Proc. Natl. Acad. Sci. USA 87, 4771–4775.
[4] Rayment, I., Holden, H., Whittaker, M., Yohn, C.B.,Lorenz, M., Holmes, K.C. and Milligan, R.A. (1993) Science 261, 58–65.
[5] Sweeney, H.L., Bowman, B.F. and Stull, J.T. (1993) Am. J. Physiol. 264, C1085–C1095.
[6] Houston, M.E., Green, H.J. and Stull, J.T. (1985) Pfluegers Arch. 403, 348–352.
[7] Persechini, A., Stull, J.T. and Cooke, R. (1985) J. Biol. Chem. 260, 7951–7954.
[8] Metzger, J.M., Greaser, M.L. and Moss, R.L. (1989) J. Gen. Physiol. 93, 855–883.
[9] Watterson, J.G., Kohler, L. and Schaub, M. (1979) J. Biol. Chem. 254, 6470–6477.
[10] Holroyde, M.J., Potter, J.D. and Solaro, R.J. (1979) J. Biol. Chem. 254, 6478–6482.
[11] Metzger, J.M. and Moss, R.L. (1992) Biophys. J. 63, 460–468.
[12] Bagshaw, C.R. and Reed, G.H. (1977) FEBS Lett. 81, 386–390.
[13] Chantler, P.D., Sellers, J.R. and Szent-Gyorgyi, A.G. (1981) Biochemistry 20, 210–216.
[14] Stępkowski, D., Orlova, A.A. and Moos, C. (1994) Biochem. J. 300, 153–158.
[15] Alexis, M.N. and Gratzer, W.B. (1978) Biochemistry 17, 2319–2325.
[16] Matsuda, G., Maita, T. and Umegane, T. (1981) FEBS Lett. 126,111–113.
[17] Pope, B., Wagner, P.D. and Weeds, A.G. (1981) Eur. J. Biochem. 117, 201–206.
[18] Pastra-Landis, S.C., Huiatt, T. and Lowey, S. (1983) J. Mol. Biol. 170, 403–422.
[19] Weeds A.G. and Taylor, R.S. (1975) Nature 257, 54–56.
[20] Wagner, P.D., Slater, C.S., Pope, B. and Weeds, A.G. (1979) Eur. J. Biochem. 99, 385–394.
[21] Hayashibara, T. and Miyanishi, T. (1994) Biochemistry 33, 12821–12827.
[22] Chalovich, J.M., Stein, L.A., Green, L.E. and Eisenberg, E. (1984) Biochemistry 23, 4885–4889.
[23] Chaussepied, P. and Kasprzak, A.A. (1989) Nature 342, 950–953.
[24] Sutoh, K. (1982) Biochemistry 21, 3654–3661.
[25] Onishi, H. and Fujiwara, K. (1990) Biochemistry 29, 3013–3023.
[26] Prince, H.P., Trayer, H.R., Henry, G.D., Trayer, I.P., Dalgarno, D.C., Levine, B.A., Cary, P.D. and Turner, C. (1981) Eur. J. Biochem. 121, 213–219.
[27] Trayer, I.P., Trayer, H.R. and Levine, B.A. (1987) Eur. J. Biochem. 164, 259–266.
[28] Yamamoto, K. and Sekine, T. (1983) J. Biochem. 94, 2075–2078.
[29] Lowey, S., Waller, G.S. and Trybus, K.M. (1993) J. Biol. Chem. 268, 20414–20418.
[30] Sweeney, H.L., Kushmerick, M.J., Mabuchi, K., Sreter, F.A. and Gergely, J. (1988) J. Biol. Chem. 263, 9034–9039.
[31] Greaser, M.L., Moss, R.L. and Reiser, P.J. (1988) J. Physiol. 406, 85–98.
[32] Bar, A., Simoneau, J.-A. and Pette, D. (1989) Eur. J. Biochem. 178, 591–594.
[33] Milligan, R.A., Whittaker, M. and Safer, D. (1990) Nature 348, 217–221.

[34] Schroeder, R.R., Manstein, D.J., Jahn, W., Holden, H. Rayment, I., Holmes, K.C. and Spudich, J. (1993) Nature 364, 171–174.
[35] Andreev, O.A. (1995) Biophys. J. 68, A141.
[36] Margossian, S.S., White, H.D., Lefford, J., Holt, J.C., Malhotra, A., Stafford, W.F. and Slayter, H.S. (1993) J. Muscle Res. Cell Motility 14, 3–14.
[37] Moczarska, A. and Kąkol, I. (1994) Abstracts of XXIII European Muscle Congress, Bochum, 19/13P.
[38] Stępkowski, D., Babiychuk, E.B., Danilova, V.M. and Kąkol, I. (1994) Biochim. Biophys. Acta 1209, 253–259.
[39] Boey, W., Everett, A.W., Sleep, J., Kendrick-Jones, J. and Dos Remedios, C.G. (1992) Biochemistry 31, 4090–4095.
[40] Morano, I. (1995) Biophys. J. 68, A168.
[41] Kishino, A. and Yanagida, T. (1988) Nature 334, 74–76.
[42] Trybus, K.M. (1994) J. Muscle Res. Cell Motility 15, 587–594.
[43] VanBuren, P., Waller, G.S., Harris, D.E., Trybus, K.M., Warshaw, D.M. and Lowey, S. (1994) Proc. Natl. Acad. Sci. USA 91, 12403–12407.
[44] Lowey, S., Waller, G.S. and Trybus, K.M. (1993) Nature 365, 454–456.
[45] Shimizu, T., Reinach, F.C., Masaki, T. and Fischman, D.A. (1985) J. Mol. Biol. 183, 271–282.
[46] Babiychuk, E.B., Stępkowski, D., Danilova, V.M. and Kąkol, I. (1991) FEBS Lett. 295, 55–58.
[47] Kąkol, I., Borovikov, Y.S., Szczęsna, D., Kirillina, V.P. and Levitsky, D.I. (1987) Biochim. Biophys. Acta 913, 1–9.
[48] Stępkowski, D., Szczęsna, D., Babiychuk, E.B., Borovikov, Y.S. and Kąkol, I. (1995) Biochem. Mol. Biol. Int. 35, 677–684.
[49] Wagner, P.D. and Stone, D.B. (1983) Biochemistry 22, 1334–1342.
[50] Ikebe, M., Ikebe, R., Kamisoyama, H., Reardon, S., Schwonek, J.P., Sanders, C.R. and Matsuura, M. (1994) J. Biol. Chem. 269, 28173–28180.
[51] Ikebe, M., Reardon, S., Schwonek, J., Sanders, C.R. and Ikebe, R. (1994) J. Biol. Chem. 269, 28165–28172.
[52] Zhi, G., Herring, B.P. and Stull, J.T. (1994) J. Biol. Chem. 269, 24723–24727.