The influence of salts on the phosphatase action of myosin.

by

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One of us has studied before the influence of salts on the phosphatase activity of myosin. It seemed desirable to repeat part of this work with crystallised myosin.

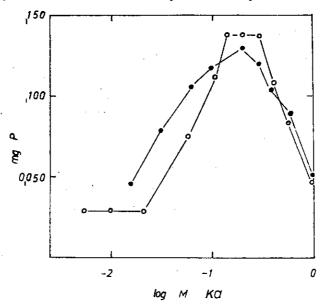


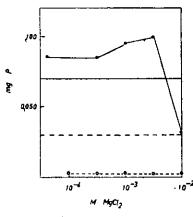
Fig. 1. Effect of KCI on the phosphatase activity of myosin and actomyosin. Formation of inorganic P determined by the method of *Fiske* and *Subbarow*. I mg myosin or 1 mg myosin plus 0.3 mg actin in 3 ml of water, 3,6 mg ATP as neutral K salt. Incubation for 5 min. at 38°.

Points = myosin, circles = actomyosin.

The influence of the KCl concentration on the phosphatase activity of myosin and actomyosin is given in Fig. 1.

It can be seen that there is no great difference between the KCl curve of myosin and acto-myosin. Both have a distinct KCl optimum at 0,2 M KCl.

The influence of MgCl₂ concentration at two different KCl concentrations is given in Fig. 2. (0,01 M KCl) and Fig. 3 (0.1 M KCl).



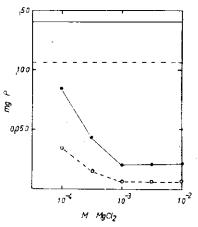


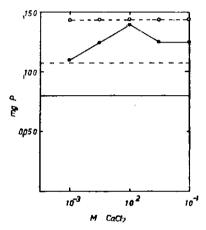
Fig. 2. Effect of MgCl₂ on the phosphatase activity of myosin in the presence of 0.01 M KCl. Broken line = myosin, full line = actomyosin. The straight lines indicate the phosphatase activity without MgCl₂.

Fig. 3. Effect of MgCl₂ on the phosphatase activity of myosin in the presence of 0.1 M KCl. Broken line = myosin, full line = actomyosin. The straight lines indicate the phosphatase activity without MgCl₂.

It can be seen that the phosphatase activity of myosin is completely inhibited even by the smallest MgCl² concentrations regardless of the KCl concentration. The influence of MgCl² on the activity of actomyosin depends on the KCl concentration. In the presence of very small KCl concentrations MgCl² enhances the phosphatase activity with a maximum between 10⁻³ and 5.10⁻³ M MgCl². Higher concentrations inhibit. In presence of 0,1 M KCl the activity of actomyosin is inhibited by all Mg concentrations, similar to the action of MgCl² on myosin. (The intermediary KCl concentrations of 0,025 and 0,045 had an intermediary effect: Up to 5.10⁻³ M Mg had no effect at all, higher concentrations inhibited.)

The question arises how this action of KCl should be

explained? The results of this laboratory obtained by visco-simetric methods² allow of a simple explanation. It has been shown that in the presence of KCl and ATP the actomyosin dissociates into actin and myosin. This dissociation depends on both the KCl and ATP concentrations. In our experiment this dissociation took place at 0,1 M KCl, hence Mg had at this KCl concentration on actomyosin the same inhibiting effect as on myosin.



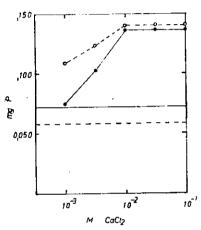


Fig. 4. Effect of CaCl₂ on the phosphatase activity of myosin in the presence of 0.01 M KCl. Broken line — myosin, full line — actomyosin. The straight lines indicate the phosphatase activity without CaCl₂

Fig. 5. Effect of CaCl₂ on the phosphatase activity of myosin in the presence of 0.1 M KCl. Broken line — myosin, full line — actomyosin. The straight lines indicate the phosphatase activity without CaCl₂.

Fig. 4 and 5 show the action of varied CaCl₂ concentrations in presence of 0,01 and 0,1 *M* KCl respectively. The experimental conditions are the same as in the experiments of Fig. 2. It will be seen from this curve that CaCl₂ has a strong enhancing influence at all concentrations between 10⁻¹ and 10⁻¹ *M*. The effect is identical at both KCl concentrations. Since myosin and actomyosin are equally activated by CaCl₂ dissociation will have no effect and identical curves will be obtained with myosin and actomyosin at both KCl concentrations.

In Fig. 6 is given the effect of varying concentrations of MgCl₂ in presence of 0,01 M CaCl₂ and 0,01 M KCl. It will be seen that the phosphatase activity which has been increased

by CaCl₂ is depressed by all concentrations of MgCl₂. The experiment gave in presence of 0,1 M KCl the same results. Mg and Ca, though in themselves both capable of increasing the phosphatase activity of actomyosin are, if given together, not synergetic but antagonistic.

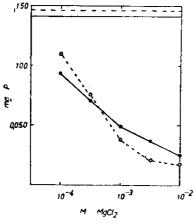


Fig. 6. Effect of MgCl₂ on the phosphatase activity of myosin and actomyosin in presence of 0.01 M CaCl₂ and 0.01 M KCl. Broken line = myosin, full line = actomyosin. The lines on the top give the phosphatase activity in absence of MgCl₂.

We want to use this occasion to correct an earlier statement. One of us (B.) has found before that quinine and nicotine strongly inhibited the phosphatase action of myosin. This was a rather important statement because these poisons do not inhibit the contraction of actomyosin. It was found since that quinine and nicotine do not inhibit the enzymic action of myosin. They only inhibit the detection of the phosphate hydrolysed. We could not corroborate either the earlier finding that oxalate inhibits the enzymic action of myosin. The question, thus, whether splitting of ATP is necessary for contraction, is still open.

^{1.} I. Banga. These studies 1, 27, 1941 - 42.

^{2.} F. Guba. This vol. p. 40.