

A CONTRACTILE ELEMENT CONTAINING TROPOMYOSIN (ACTOTROPOMYOSIN)

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The proteins derived from the contractile system of the skeletal muscle have attracted much interest, and our knowledge of the chemistry of the contractile system emanates chiefly from investigations on skeletal muscles. Our knowledge of the biochemistry of smooth muscles is, on the other hand, rather limited.

The muscle of uterus shows in its behaviour many features uncommon to other muscles. It is only working during a short period. During pregnancy it will increase in weight and after partus it rapidly decreases again.

Work performed on the contractility of threads prepared from homogenates of uterus muscle has shown that the contractility of the threads will increase during pregnancy and that threads prepared from reprecipitated material will show a poor contractility¹. In an electrophoretic study on homogenates² it has been shown that the component increasing during pregnancy is in many respects similar to actomyosins. This component increases in the corpus part of the uterus during pregnancy, but not in cervix and isthmus where the contractility does not increase.

It was, therefore, of considerable interest to see if the contractile proteins could be prepared in the same manner as from the skeletal muscle by using tissue from the corpus part of the uterus. The method of SZENT-GYÖRGYI³, successful in preparing myosin and actomyosin from skeletal muscle, has been used.

In the investigations sections from the corpus part of the human myometrium, taken in connection with uterus extirpations, have been used. In some cases the corpus part of cow uterus has been used in order to obtain a greater mass of material. No differences between these two kinds of muscle tissue have been observed.

Some experiments were first performed to obtain myosin from muscle of pregnant uterus by the method of SZENT-GYÖRGYI. No myosin precipitate formed. An analysis of the extracted solution showed that it contained less than five per cent myosin but it did contain actin and nearly sixty per cent tropomyosins. On account of the failure to obtain myosin, the investigations were concentrated on preparing actomyosin. It should be mentioned here that the contractile protein complex we extracted from the uterus muscle is not actomyosin even if in many respects it behaves as such and is obtained from the uterus muscle in the same manner as actomyosin from the skeletal muscle. We suggest the name actotropomyosin for the new complex.

The preparation of actotropomyosin

The muscle was thoroughly minced in the frozen state with a chop mincer. The

minced muscle was generally washed twice by stirring with ice cold, redistilled water for about one hour. After the washing, the minced muscle was immediately extracted with 0.5 *M* potassium chloride buffered with 0.1 *M* potassium phosphate (pH 7) for 18 hours at 0° C with gentle stirring. 300 ml of solution was used per 100 g muscle. The undissolved material was centrifuged off and the solution diluted with 4 volumes of redistilled water (0° C). A precipitate was formed during gentle stirring for 20–30 minutes. This was centrifuged off. Both the supernatant and the precipitate were saved for further processing.

The precipitate was dissolved in 0.5 *M* potassium chloride and 0.1 *M* potassium phosphate (pH 7) and dialysed for two days against the same solution. The solution so obtained has been called "once precipitated actotropomyosin". In some experiments, the proteins have been reprecipitated and redissolved in the same manner. The solution obtained has been called "twice precipitated actotropomyosin".

The pH of the supernatant has been readjusted with acetic acid to 4.6. A white flocculate precipitate formed. It was centrifuged off, resuspended and dialyzed against the original buffer solution. The dialyzed solution has then been analyzed.

Yields of actotropomyosin

The concentration in solutions of once precipitated substance has been determined by measurements of the increment of refractive index ($n = 186 \cdot 10^{-5}$ for one per cent solution). In Table I the mean values of several measurements are given. The variation was about ± 1 in the different measurements.

TABLE I

<i>Species</i>	<i>mg/g wet muscle</i>
human non-gravid	4
human gravid	1
cow non-gravid	4
cow gravid	1

Some comments concerning the preparations

In the cases when non-gravid muscle was investigated, a nucleoprotein was also precipitated. It was possible to separate partly the nucleoprotein from the solutions before the actotropomyosin since it precipitated at a water-dilution of 1.5–2 volumes. In some cases where the muscle was infiltrated with myoma, the nucleoprotein occurred in such amounts that no separation at all could be obtained. The nucleoprotein has not been investigated.

In the cases where muscle from pregnant uterus was used, no such precipitate was obtained.

Investigations on the solutions

Viscometric investigations have been performed on solutions of once and twice precipitated actotropomyosin. An Ostwald viscometer was used (110 sec., 22° C).

The concentration dependence of the specific viscosity in solutions of once precipitated actotropomyosin from non-gravid uterus was less than that from a synthetic actomyosin (crystallized myosin and actin 3:1) from skeletal muscle. A reprecipitation

of the actotropomyosin gave a more pronounced dependence of the viscosity (Fig. 1). The viscosity after ATP had been added dropped to about the same value as for the synthetic actomyosin with ATP. When measured again after 24 hours, the viscosity had risen nearly to the original value.

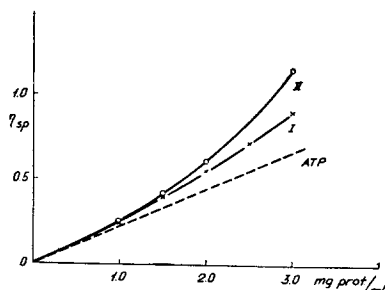


Fig. 1. The concentration dependence of the specific viscosity of actotropomyosin from non-gravid uterus. I. Once precipitated actotropomyosin. II. Twice precipitated actotropomyosin. The dotted curve shows the viscosity after addition of ATP.

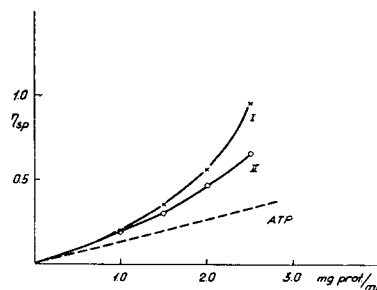


Fig. 2. The concentration dependence of the specific viscosity of actotropomyosin from gravid uterus. I. Once precipitated actotropomyosin. II. Twice precipitated actotropomyosin. The dotted curve shows the viscosity after addition of ATP.

The concentration dependence of the specific viscosity of actomyosin from pregnant uterus was often less than that of non-gravid uterus. It varied much in separate preparations. A reprecipitation of the actotropomyosin lowered the viscosity contrary to the non-gravid case. The viscosity after ATP had been added was much lower than in the other case (Fig. 2). Moreover, when treated with ATP, there is some difference in viscosity between once and twice precipitated actotropomyosin from pregnant uterus. Once precipitated uterus behaves as actotropomyosin from normal uterus. On addition of ATP, the viscosity shows a certain drop but then slowly increases and after some hours has reached its original value again. Twice precipitated actotropomyosin also shows this drop in viscosity when ATP is added, but the viscosity does not rise again with time.

TABLE II

"Actotropomyosin"	Precipitations	Conc. mg protein/ml	η_{sp}	ATP added	After 20 hours
non-gravid uterus	1	3	0.89	0.75	0.82
	2	3.5	1.05	0.89	0.97
gravid uterus	1	2.4	0.45	0.28	0.45
	2	2.6	0.47	0.35	0.35

In one experiment the actotropomyosin was precipitated four times. The solution then had a low viscosity and did not respond at all to the addition of ATP.

Some electrophoretic measurements were made on once and twice precipitated actotropomyosin from gravid uterus using 0.4 *M* KCl + 0.1 *M* K-phosphate, pH 7, as buffer. The solutions were somewhat polydisperse. In both cases three main components were visible with the mobilities $u = 7.7 \times 10^{-5}$, 4×10^{-5} , and 2.8×10^{-5} . The greatest peak was that with the mobility of about $4 \cdot 10^{-5}$. The mobility of this component was not quite constant in separate experiments.

References p. 208.

Salting-out analysis

To get a better definition of the protein composition of the solutions, some salting-out analyses have been carried out with the technique of DERRIEN⁴.

The protein solution was dialysed against a buffer solution (0.5 *M* KCl + 0.1 *M* K-phosphate, pH 6.9) for 2 days. The concentration of protein in the solution was measured and the solution was diluted so it contained about 3 mg per ml.

A saturated ammonium sulphate solution was prepared containing the buffer solution and the pH was adjusted to 6.9. From the ammonium sulphate and buffer solutions a series of solutions was prepared containing different amounts of ammonium sulphate. Every solution had a volume of 7 ml. To each one 3 ml of the protein solution was added and the mixtures were allowed to stand in ice water for about 24 hours. They were then filtered and to 5 ml of each solution 5 ml of buffer solution was added before the temperature was allowed to rise to room temperature. The extinction of the solutions was measured at 260 and 278 m μ .

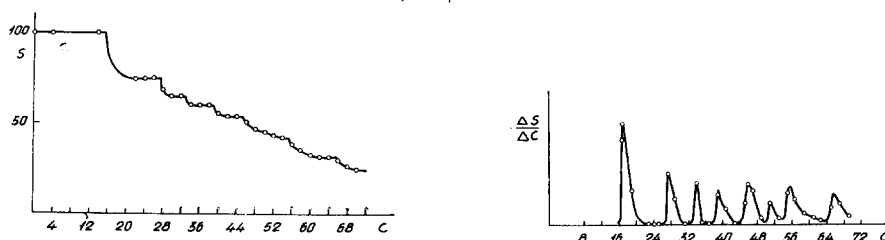


Fig. 3. Salting-out curve of the supernatant after precipitation of the actotropomyosin by dilution from the salt extract (non-gravid uterus). *S* = the extinction at 278 m μ . The initial extinction is put to 100. *C* = per cent in volume of saturated ammonium sulphate. Equilibrium temperature 0° C. Time for equilibrium 18 hours. $\Delta S/\Delta C$ = the derived salting-out curve.

In a diagram the extinction was plotted against the concentration of ammonium sulphate. Also the differential extinction as a function of concentration has been computed. The latter diagrams are also given in Figs. 3, 6 and 7.

The measurements have been performed in steps of two per cent of the saturated ammonium sulphate. It should be noticed that many more determinations are necessary to discover small components. The tedious work of finding such components has not been performed here; the work has been directed only towards the main components.

The precipitating ranges of the different structural main proteins found in other measurements on the different substances prepared are as follows: In the range 9–20% of saturated ammonium sulphate the actins precipitate. Between 28–32% actomyosin precipitates and in the range 33–45% the myosin components precipitate. Between 45–55% a protein precipitates which we have called the phosphate absorbing protein. In the next range, 55–64%, the tropomyosin components precipitate. Between 64–70% nucleotropomyosin precipitates and above 70% nucleoproteins and nucleic acid remain.

Salting-out curves (non-gravid uterus)

After precipitation by dilution of a salt extract prepared as described above, the supernatant was investigated. The remaining proteins in the solution were first precipitated at pH 4.6 and then dissolved again in the buffer and dialyzed against this before the salting-out analysis. A diagram of the analysis is given in Fig. 3. The diagram shows that the solution contains a relatively large amount of actin. Certain amounts of all the other structural proteins also appear. The ultraviolet absorption of the substances remaining in the supernatant above 70% saturated ammonium sulphate was fairly high. The difference between such a diagram and the one from an F-actin solution prepared

according to STRAUB is not great. However, a higher per cent of actin appears in the latter case and less material remains in the supernatant.

The precipitate obtained on dilution of the salt extract was reprecipitated to free it from most of the accompanying nucleoprotein and then dissolved, dialyzed and analyzed (Fig. 4). Two components are first found precipitating in the same range as G- and F-actin. At a somewhat higher ammonium sulphate level, an ultraviolet absorbing material goes into solution from the material already precipitated. In the range where this occurred the ultraviolet absorption at $260\text{ m}\mu$ was higher than that at $275\text{ m}\mu$, which points to nucleoproteins. These precipitated again in the range 45–52%. This type of salting-out curve seems to be characteristic for the actotropomyosin. More pronounced curves of the same type were obtained in the gravid case.

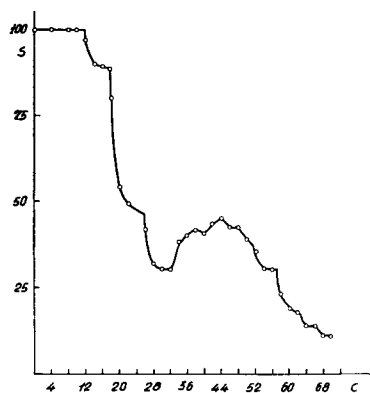


Fig. 4. Salting-out curve of the actotropomyosin twice precipitated (non-gravid uterus); conditions as in Fig. 3.

Investigations on gravid uterus muscle

After precipitation by dilution, the supernatant was investigated also in this case, after the same treatment previously described. From the curves it could be concluded that 30% of the extinction at $275\text{ m}\mu$ is due to the material precipitating as actin. The salting-out curve did not show any great differences from that obtained in the non-gravid case except that the material remaining in solution after 70% saturated ammonium sulphate was less than half of that in the non-gravid case.

The supernatant from twice precipitated actotropomyosin was investigated. Small amounts of actin and myosin were obtained and a great amount of tropomyosins. Also small amounts of the substance precipitating between 45–52% appeared (Fig. 5).

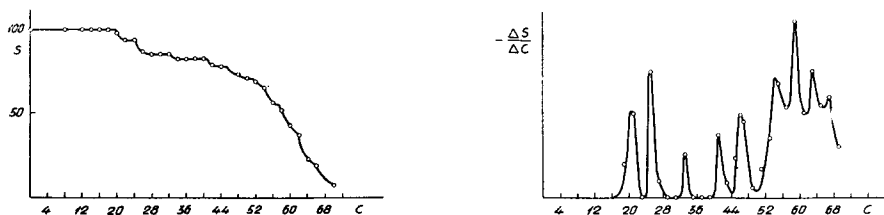


Fig. 5. Salting-out curve of the supernatant from twice precipitated actotropomyosin (gravid uterus); conditions as in Fig. 3.

The actotropomyosin twice precipitated and redissolved was treated with ATP, which was then dialyzed away. The salting-out curves showed certain irregularities as in the case of actotropomyosin from the non-gravid uterus. It was, however, clear that the solution contained actin, tropomyosins, and the phosphate-absorbing protein.

The viscosity of a solution from twice precipitated actotropomyosin was reinvestigated. The same phenomenon was observed as before. The viscosity dropped when ATP was added and did not rise again. But if, after the lapse of 24 hours, more ATP was added the viscosity dropped a little more. Evidently a small part of the substances was still able to reform the complex.

The phenomenon of a substance disintegrating during the salting-out experiments was also found in a solution of actotropomyosin once precipitated. The effect was here very pronounced. Small amounts of actomyosin and myosin precipitating during the rise of the curve can be distinguished as irregularities in a smooth curve where the precipitation of these substances can be expected (Fig. 6).

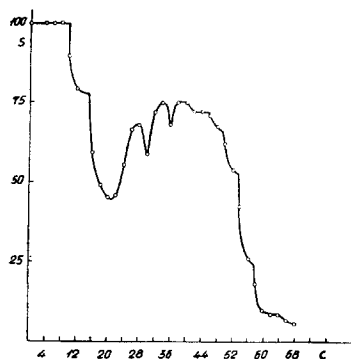


Fig. 6. Salting-out curve of actotropomyosin. Once precipitated (gravid uterus); conditions as in Fig. 3.

An extract of gravid uterus was also investigated where no substance precipitated on dilution. As seen from the salting-out curve, only a small rise of the curve was obtained. Evidently the solution contained only a minute amount of the complex. As seen from the curve, the solution contained much actin and a lot of the tropomyosins and also the phosphate-absorbing protein (Fig. 7).

An analysis has also been made of the constituent parts of the actotropomyosin. To a solution ATP was added and the actin was spun down in a "Spinco" centrifuge. The supernatant was dialyzed and a salting-out curve was determined. The curve showed similarities to that of the supernatant from twice precipitated actotropomyosin but more of the phosphate absorbing protein was found here.

Quantitative data for the different components cannot be obtained from the curves without knowledge of the specific extinction of the different components; only a comparison between the different curves can be made. Experiments are in progress to determine directly the amount of the different components and these will be reported later. Preliminarily it can be stated that about 40% of the actotropomyosin is actin and 20-25% tropomyosin. The remaining material consists of the phosphate-absorbing protein, nucleic acid and some myosin.

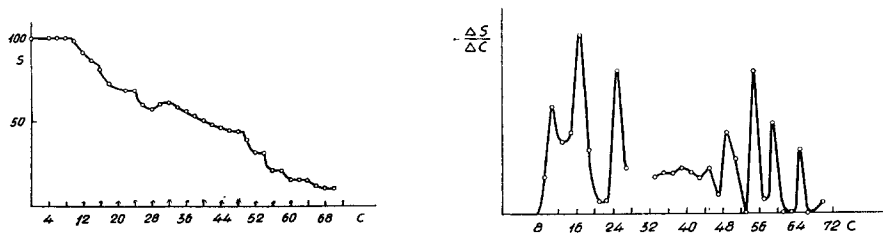


Fig. 7. Salting-out curve of salt extract from gravid uterus where no actotropomyosin precipitated; conditions as in Fig. 3.

DISCUSSION

Our investigations show that there must be great structural differences between the uterus and skeletal muscles (even if no new structural proteins occur). An interesting feature in the extraction experiments is that the actin in the uterus muscle can be dissolved so easily. In the gravid case, after an 18 hours salt extraction, no actin could be obtained from the insoluble residue by the method of STRAUB, indicating that all the actin had gone into solution.

If a short salt extraction is performed with skeletal muscle almost only myosin is obtained. In the uterus case, however, a solution was obtained containing about 35% material precipitating as actin before 24% saturation of ammonium sulphate and only a very small amount of myosin and actomyosin (5%). Very much of the tropomyosins appeared.

It is very difficult to decide if the amounts of myosin and actomyosin found in the experiments really belong to the contractile system of the uterus muscle. It may also be that they are contaminations deriving from other muscles.

Our investigations show that a complex of another quantitative composition of structural proteins than the usual actomyosin is extractable from uterus. In many respects it reacts as actomyosin and ought, therefore, to be considered to contain the contractile element of the uterus muscle. Since it contains large amounts of actin and even tropomyosins we have called it actotropomyosin.

The existence of actotropomyosin is supported by different investigations. In the ultracentrifuge⁵ it has been observed to sediment as rapidly as actomyosin. After the addition of ATP to a solution of the actotropomyosin, the ultracentrifuge diagrams show a polydisperse substance sedimenting with about the same speed as actin and a material with a lower sedimentation constant than myosin.

Electrophoretic investigations² performed on homogenates of gravid corpus demonstrate the presence of a dominating component with a mobility near $4.3 \cdot 10^{-5}$, thus higher than that of actomyosin. This component is still more pronounced in the electrophoresis diagram of the once precipitated substance. Most of the substances with higher mobilities than the complex disappears.

Our salting-out curves show that there is a substance precipitating before the actomyosins which gradually disintegrates at higher ionic strength releasing substances which precipitate again at still higher ammonium sulphate concentrations. This phenomenon does not occur in solutions where the complex is totally disintegrated before the start of the experiment, in which cases proper salting-out curves are obtained.

The actotropomyosin seems hitherto to have been confused with actomyosin because in so many respects it behaves similarly to this protein. We want, therefore, to stress a little the similarities and dissimilarities between the two substances. The following discussion refers to the actotropomyosin from the gravid corpus muscle, since this seems to be the purest preparation.

The actotropomyosin precipitates from salt solutions as actomyosin but at a slightly higher dilution and very often with a heavy loss of material.

It is similar to actomyosin in the possibility of making threads of it which can contract when ATP is added. Threads of actotropomyosin seem able to respond to ATP as effectively as those of actomyosin. Actotropomyosin responds to ATP in salt solutions with a drop of the viscosity which after a certain time increases again. A difference can, however, be observed. The viscosity drops to a lower value for actotropomyosin at the same protein content as in the case of the actomyosin.

The actotropomyosin is unstable when it is precipitated by dilution from a salt solution. Twice precipitated and redissolved it reacts with ATP with a drop of the viscosity but the viscosity does not rise appreciably again, indicating a loss of part of the structure. A smaller part seems, however, to recover since on addition of ATP again to such a solution after 20 hours, the viscosity drops still more.

Salting-out analyses of the material left in solution after reprecipitation of the

actotropomyosin indicated that the greatest part of the substances was tropomyosins, the phosphate-absorbing protein and ribonucleic acids. From the electrophoretic investigations it seems that these parts build up a complex which reacts with actin. When this complex disintegrates, it loses its ability to react with the actin.

The word tropomyosins has been used since three different kinds of tropomyosins have been distinguished: the tropomyosin of BAILEY⁶, the tropomyosin of HAMOIR⁷ and the nucleotropomyosin⁸. From the great amounts of nucleic acid occurring in the solutions and from the instability and insolubility of the actotropomyosin in dilute salt solutions, it can be concluded that nucleotropomyosin occurs in the complex. Investigations which will be reported later show that it is here connected with the phosphate-absorbing protein.

In investigations not reported here, using small amounts of organic solvents to prepare the phosphate-absorbing protein in a free state, it has been found that this precipitates between 45 and 52% saturated ammonium sulphate. It can take up phosphate from ATP but not from orthophosphate. It shows a rapidly declining phosphatase activity during the first hours after the preparation from the complex. Therefore, it is assumed that the phosphatase uptake is due to a residue of the phosphatase activity. It also takes part in a deaminating process, in these respects resembling myosin. Since nucleotropomyosin does not show any enzymic properties, it can be assumed that this protein probably holds the other protein in a certain state so that it can exhibit the phosphatase activity.

Finally, we will point to some differences of the non-gravid and gravid uterus muscle. In the electrophoretic work performed on homogenates of the uterus muscle it was shown that a very sharp peak related to the actotropomyosin only appeared in the homogenates from the gravid corpus part. A similar very sharp peak did not appear in the gravid isthmus case or in homogenates from non-gravid corpus or cervix. In these cases a broader peak appeared suggesting that either another substance overlapped the small amount of actotropomyosin or that there was a totally different substance present. The conclusion from these measurements is that the complex appears in great amounts only in that part which contracts effectively and strongly during pregnancy, during which state there is an increase in the content of actotropomyosin per gram wet muscle in that part.

In our investigations we have found that from the non-gravid corpus a nucleoprotein can be extracted in large amounts. The amount decreases during the gravidity and in the gravid case it can only be present in small quantities. This nucleoprotein does not precipitate in ammonium sulphate up to 73%. Analyses of it have not been made.

When the salt extracts are diluted this nucleoprotein precipitates but at a higher salt concentration than the actotropomyosin, so the actotropomyosin can be partly freed of it. It seems, however, nearly impossible to get rid of it in the non-gravid case. While actotropomyosin also occurs in this case a possible explanation of the greater stability of the actotropomyosin (compare the viscosimetric measurements) is that the nucleoprotein, in some manner, prevents the actotropomyosin from disintegrating.

In this connection it may also be of interest to compare our measurements of the actotropomyosin content per gram wet muscle performed by different methods. The results are summarized in Table III.

Even if these figures are very approximate, they clearly show certain discrepancies.

TABLE III

COMPARISON OF THE ACTOTROPOMYOSIN CONTENT IN CORPUS UTERI HOMOGENATES
MEASURED BY DIFFERENT METHODS

Electrophoretic yields (*u*). Viscosimetric yields (*v*). Amount of precipitated actotropomyosin (*p*).
All yields in mg protein per gram wet muscle.

	<i>u</i>	<i>v</i>	<i>p</i>
Non-gravid corpus	11.2	3.5	4
Gravid corpus	14.4	14.2	1

In the non-gravid case a little more actotropomyosin is obtained by precipitation than viscosimetrically calculated. This probably depends on the precipitated actomyosin not being pure. A much higher value is obtained from the electrophoretic calculations on whole homogenates. Evidently this depends on another substance (the nucleoprotein) interfering with the measurements. In the gravid case only a small part of the actotropomyosin precipitates. A reprecipitation of the complex causes also heavy losses of material so that, on account of the instability of the complex, such a small amount is obtained.

The tropomyosin occurs in both cross-striated and smooth muscles. Generally it comes out to a small extent when actin is prepared according to the method of STRAUB. One may draw the conclusion that the tropomyosin really is a part of the contractile system in all kinds of muscle. The fact that the reactivity with actin has not been observed before probably depends upon the complex being very labile and also in many respects behaving in the same manner as actomyosin. Thus small amounts of the complex described have been overlooked.

ACKNOWLEDGEMENT

This investigation has been financially supported by the Swedish Medical Research Council and the Swedish Natural Science Research Council. The authors want to express their gratitude to Professor J. NAESLUND for his interest.

SUMMARY

The contractile element of the uterus body is shown to have composition other than that of the skeletal muscle. Only small amounts of myosin and actomyosin can be extracted. On short salt extraction, much tropomyosin appears in the solution and also free actin is found.

On longer salt extraction, a complex called actotropomyosin can be obtained from the extract. The actotropomyosin reacts with ATP similarly to actomyosin. In its general behaviour it shows rough similarities with actomyosin but the differences between the two complexes are clear cut.

On reprecipitation, the complex partly disintegrates and gradually loses its ability to react with ATP.

Salting-out analyses of the complex show that it starts to precipitate at low salt concentrations but disintegrates. A part of it goes into solution and precipitates at higher ammonium sulphate concentrations.

Salting-out curves of the material left in the solution after reprecipitation show that the supernatant contains a protein, which has been called the phosphate-absorbing protein, tropomyosins, and ribonucleic acid. The same result is obtained by an investigation of the material disintegrating from the ammonium sulphate and precipitating at higher salt concentrations.

It is proposed that the actotropomyosin contains actin, the phosphate-absorbing protein and nucleic tropomyosin. The two latter components form a complex acting as myosin. The phosphate absorbing protein has enzymic properties when part of the complex.

Some difference between the extractions from normal and gravid uterus are discussed.

RÉSUMÉ

L'élément contractile de l'utérus n'a pas la même composition que celui des muscles squelettiques. On ne peut en extraire que de faibles quantités de myosine et d'actomyosine. La solution qui résulte d'une brève extraction saline, renferme beaucoup de tropomyosine et de l'actine libre. Après une extraction saline prolongée, l'extrait renferme un complexe que les auteurs désignent par actotropomyosine. L'actotropomyosine réagit comme l'actomyosine avec l'ATP. Son comportement général est en gros semblable à celui de l'actomyosine mais des différences très nettes existent entre les deux complexes.

Au cours de la reprécipitation, le complexe se dissocie en partie et ne peut plus réagir avec l'ATP.

L'étude du relargage montre que le complexe commence à précipiter à des concentrations salines faibles en se dissociant. Une partie se redissout et précipite à des concentrations en sulfate d'ammonium plus élevées.

Les courbes de relargage du produit resté en solution après reprécipitation montrent que la solution surnageante renferme une protéine, que les auteurs désignent par "phosphate-absorbing protein", de la tropomyosine et de l'acide ribonucléique. L'étude du produit qui se dissocie en présence de faibles concentrations de sulfate d'ammonium et reprécipite à des concentrations plus élevées conduit aux mêmes résultats.

Les auteurs supposent que l'actotropomyosine renferme de l'actine, la "phosphate-absorbing protéine" et de la tropomyosine nucléique. Ces deux derniers constituants forment un complexe fonctionnant comme la myosine. La "phosphate-absorbing protein" possède les propriétés enzymatiques quand elle fait partie du complexe.

Les extractions de l'utérus normal et de l'utérus gravide présentent quelques différences qui sont discutées.

ZUSAMMENFASSUNG

Es wurde gezeigt, dass die kontraktile Elemente des Uterus eine andere Zusammensetzung wie die der Skelettmuskeln haben. Es können nur kleine Mengen Myosin und Actomyosin extrahiert werden. Bei kurzen Salzextraktionen wird in der Lösung viel Tropomyosin neben freiem Actin gefunden.

Bei längeren Salzextraktionen wird ein Actotropomyosin genannter Komplex aus dem Extrakt erhalten. Actotropomyosin reagiert mit ATP wie Actomyosin. In seinem allgemeinen Verhalten zeigt es grobe Ähnlichkeit mit Actomyosin, aber es besteht ein deutlicher Unterschied zwischen beiden Komplexen. Bei der Wiederausfällung zerfällt der Komplex teilweise und verliert allmählich seine Fähigkeit mit ATP zu reagieren.

Aussalzanalysen des Komplexes zeigen, dass er bei niedrigen Salzkonzentrationen auszufallen beginnt, aber dabei zerfällt. Ein Teil davon geht in Lösung und fällt bei höheren Ammonsulfatkonzentrationen aus.

Aussalzkurven des in Lösung verbliebenen Materials zeigen, dass nach der Wiederausfällung die überstehende Flüssigkeit ein Protein, das Phosphat-absorbierendes Protein genannt wurde, Tropomyosine und Ribonucleinsäure enthält. Das gleiche Ergebnis wurde bei einer Untersuchung des mit Ammonsulfat zerfallenden und bei höheren Salzkonzentrationen ausfallenden Materials erhalten.

Es wird angenommen, dass das Actotropomyosin Actin, Phosphor-absorbierendes Protein und Nucleintropomyosin enthält. Die beiden letzten Bestandteile bilden einen Komplex, der wie Myosin wirkt. Das Phosphat-absorbierende Protein besitzt, wenn es ein Teil vom Komplex ist, die enzymatischen Eigenschaften.

Einige Unterschiede zwischen den Extraktionen aus normalen und graviden Uterus werden besprochen.

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Received August 24th, 1953