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## Dephosphorylation of Adenosine Triphosphate in Muscular Contraction

CONTEMPORARY views on the role of adenosine triphosphate in contractile activity include the assumption that the breakdown of this substance is the immediate chemical source for mechanical energy[1,2]. However, as has recently been emphasized by A. V. Hill[3], there is no proof that such a dephosphorylation occurs as an early event in contraction. Furthermore, it has been shown[4] that the effects of adenosine triphosphate upon certain physical properties of actomyosin do not primarily involve splitting of the nucleotide. The experiments reported here were undertaken in order to decide whether, indeed, any dephosphorylation of adenosine triphosphate occurs during the contractile phase of a twitch.

It appears unfavourable to attempt the detection of a diminution of adenosine triphosphate by analysis of the hydrolysable phosphate (compare Lundsgaard[5]), since one would have to evaluate small differences between large numbers, which might furthermore be distorted by the formation of other labile phosphate compounds. Instead, we have employed a chromatographic assay method, closely resembling that of Cohn[6], to determine adenosine mono-, di- and triphosphate separately by their ultra-violet absorption.

The experiments were carried out with the combined leg muscles of frogs. Muscles from the two legs were isolated, care being taken to subject the two sets to the same manipulations. Starting at a temperature of about 20° C., one set was immediately extracted with ice-cold 1 *N* perchloric acid, whereas the other set was fixed at the height of contraction by sudden submersion into liquid air (compare Lundsgaard[5]). The results are shown in the accompanying table.

It is seen that in contraction there is a pronounced decrease of adenosine triphosphate, and that its breakdown goes in part beyond the stage of the diphosphate, probably by the myokinase mechanism. The decrease of adenosine triphosphate averages about one-fifth of the total nucleotide, which is about twice the expected amount[7]. If confirmed, this would indicate that precipitous cooling mobilizes the contractile apparatus to a somewhat greater extent than occurs physiologically in a single twitch, or else that in this mode of stimulation part of the muscle twitches more than once. We intend to study various other ways of eliciting contraction in different types of muscles.

DISTRIBUTION OF ADENINE NUCLEOTIDES AS A PERCENTAGE OF THE TOTAL NUCLEOTIDE FRACTION, FOR FROG MUSCLE AND (LAST LINE) FOR RABBIT PSOAS MUSCLE

| Control | | | Contracted | | | Difference | | |
|---|---|---|---|---|---|---|---|---|
| AMP | ADP | ATP | AMP | ADP | ATP | AMP | ADP | ATP |
| 7·5 | 14·5 | 78·0 | 9·0 | 26·8 | 64·2 | +1·5 | +12·3 | −13·8 |
| 6·3 | 13·5 | 80·2 | 10·0 | 15·9 | 74·1 | +3·7 | +2·4 | −6·1 |
| 8·6 | 14·0 | 77·4 | 5·5 | 37·4 | 57·1 | −3·1 | +23·4 | −20·3 |
| 8·6 | 44·7 | 46·7 | 14·8 | 55·8 | 29·4 | +6·2 | +11·1 | −17·3 |
| 5·3 | 22·3 | 72·4 | 6·0 | 44·9 | 49·1 | +0·7 | +22·6 | −23·3 |
| 12·8 | 12·2 | 75·0 | 16·8 | 39·8 | 43·4 | +4·0 | +27·6 | −31·6 |
| 2·2 | 12·7 | 85·1 | 5·2 | 16·2 | 78·6 | +3·0 | +3·5 | − 6·5 |

In sustained muscular activity, and possibly also in a completely reversed twitch, a breakdown of phosphocreatine rather than of adenosine triphosphate is generally detected (see ref. 2). Our present results suggest, however, that it is primarily adenosine triphosphate which is dephosphorylated in the contractile phase. Apparently, a transfer of phosphate from phosphocreatine to adenosine di- and monophosphate by the Lohmann reaction[8] occurs only in later phases of the contractile cycle. We suggest that actually the Lohmann reaction is specifically connected with relaxation.

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## Transaminases in *Neurospora crassa*

THE presence of a wide range of transaminases in both animal tissues[1] and bacteria[2] has recently been reported. A rather similar array of enzymes appears to be present in the mould *Neurospora crassa*.

Mycelium of the wild-type strain *E5297a* was grown for three days at 25° in 'minimal' Fries No. 3 medium, and cell-free extracts were obtained by grinding with phosphate buffer and powdered glass followed by centrifugation as described in detail elsewhere[3]. The extracts, which were free of large