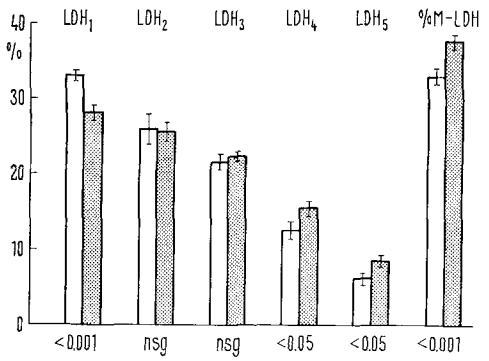
anoxia. Data in the literature[9] show that after the administration of isoproterenol there is an increase in the myocardium in the level of the energy substrate, glycogen, and also of Q_{O_2}[10] but not in correlation with resistance of the heart to acute anoxia.

Increased resistance both of the myocardium to anoxia in vitro, and to necrogenic doses of isoproterenol was also found in height adapted rats[11]. Anaemic rats also have a greater tolerance for anoxia[12]. One of the common factors in all 3 models (adaptation to isoproterenol, adaptation to height[13], anaemic rats[14] is a shift of LDH isoenzymes in favour of the anaerobic type of metabolism which is correlated with increased tolerance of the myocardium for acute anoxia in vitro. An increase in M-type LDH may be one of the mechanism by which the heart becomes adapted to an anaerobic environment. An increase in M-type subunits can maintain LDH activity, even in the presence of a greater amount of substrate and can thus maintain the heart in a state of greater efficiency during anoxic periods in experiments in vitro and on the isolated right ventricle[11].



Distribution of isoenzymes LDH and percent of M-type subunits LDH in the heart. □, controls; ////, isoproterenol.

Zusammenfassung. Die an kleine Dosen Isoproterenol gewöhnten Herzen von Ratten wiesen eine bedeutsame Vermehrung der LDHM-Untereinheiten auf. Die Zunahme der Fähigkeit zu anaërober Glykolyse ist wahrscheinlich einer der Faktoren, der für die grössere Widerstandsfähigkeit der isolierten rechten Herzkammer dieser Ratten gegen akute Anoxie verantwortlich ist.

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The Relationship Between Activation Heat and Calcium Transients in Frog Sartorius Muscle

The heat which antecedes tension development has been originally defined by Hill[1] as activation heat; this heat is still produced after work and tension development is eliminated. During the initial events preceding contraction, the activator calcium is released from the sarcoplasmic reticulum[2].

The relation between the kinetics of Ca^{2+} changes and the time course of heat liberation at 0 °C has been studied in freshly excised frog sartorius muscle, using the characteristic absorption of the Ca-murexide complex at 470 nm and 540 nm as a measure of the free Ca^{2+} concentration. The procedure was that of Jöbsis and O'Connor[3]. To eliminate any mechanical response, the muscle pairs were initially preshortened to about 75% of their standard length by previous stimuli, a method which has previously been used to measure activation heat[1]. The heat changes at 0 °C were followed with a Hill-type thermopile (90 chromel-constantan couples) connected to a sensitive galvanometer system. The light transmission was directly recorded in a cold room with the aid of a double-beam spectrophotometer, specially adapted for low-temperature measurements. In some experiments, in which the ATP content was additionally analyzed, the rephosphorylating reactions were inhibited by treating the muscles with 0.38 m*M* fluorodinitrobenzene according to Davies et al.[4]. This treatment had no effect on the heat and Ca^{2+} transients.

The results obtained for single stimuli (condenser discharges of 0.05 μF, 40 V) are presented in the Figure. If one compares the first time derivative of the activation heat with the kinetics of the Ca-murexide complex, it becomes obvious that, apart from the initial 25 msec, there exists a close relationship between the release and reabsorption of Ca^{2+} and the rate of heat production. To obtain some information as regards the changes in the level of ATP in the time course of activation, the muscles were frozen at 40 msec or at the end of the 370 msec period by immersing the experimental (as well as the unstimulated control) muscle rapidly in isopentane cooled with liquid nitrogen. As indicated by the voltage changes of the stimulus, later than 30 msec after the start of the freezing no longitudinal currents could pass along the muscle. Thus freezing must have been complete at 70 msec and 400 msec, respectively. Analyzing the ATP content of 32 muscle pairs fluorometrically, revealed that there was no detectable change in the ATP level which paralleled the

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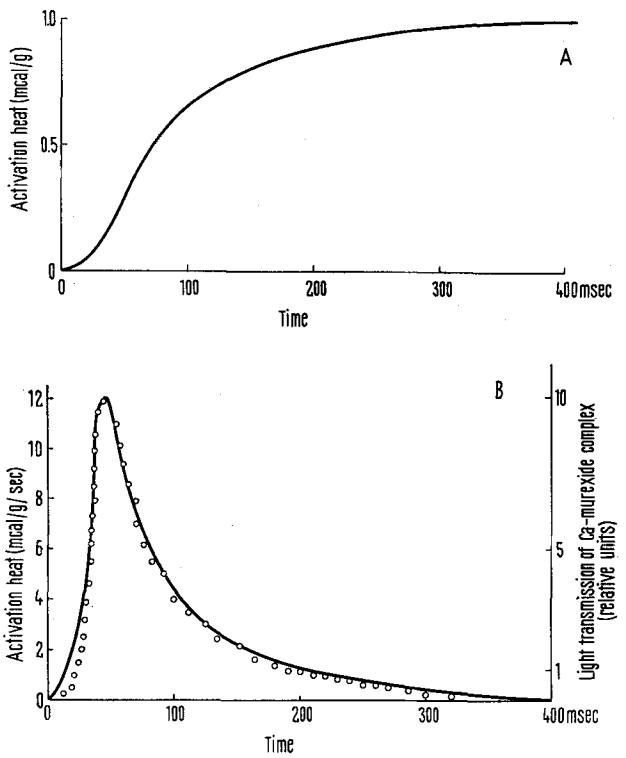
heat liberation during the initial 40–70 msec. However, over the 400 msec period 0.072 ± 0.008 µmole ATP/g muscle was hydrolyzed, a value which agrees with similar findings of DAVIES et al.[4].



Time course of activation heat (A) and the relationship between the first time derivative thereof and the release and reabsorption of calcium (B). Measurements of light transmission and heat production were obtained in parallel experiments. The solid line in Figure B is the time derivative of activation heat; the open circles represent the Ca^{2+} concentration at the various time intervals averaged from 10 successive twitches. The Ca-murexide concentration is given in relative units, all points are fractions of the light absorption value at 200 msec.

The difference in the time course of heat production and Ca^{2+} release over the initial 25 msec can readily be explained in terms of conformational changes accompanying the propagation of membrane depolarization[5]. As these changes are very similar to those observed upon nerve excitation[5], the value of 0.009 mcal/g obtained as heat per impulse in crab nerves[6] may be used for comparison. The heat value of about 0.015 mcal/g suggested from the difference in Figure B may reflect the greater membrane surface of muscle[7]. The finding that about 0.24 mcal heat/g liberated during the initial 50 msec are unaccompanied by any ATP splitting, suggests that all the ATP is split during the time in which Ca^{2+} is pumped back into the sarcoplasmic reticulum. SRETER[8] reports that for any molecule of ATP split, 8–10 Ca^{2+} ions are taken up by the sarcoplasmic vesicles. Therefore, on the basis of the observed ATP hydrolysis, the calcium level should be about $6 \times 10^{-4} M$ at its maximum in activated frog sartorius muscle at 0 °C.

As a working hypothesis it is suggested that the fraction of heat produced initially in parallel to the build-up of the free Ca^{2+} concentration reflects some conformational change as a result of Ca-binding to the myofilaments.

Zusammenfassung. Für den Froschmuskel wird ein enger Zusammenhang zwischen der zeitlichen Änderung der Aktivierungswärme und der Änderung der Kalziumkonzentration nachgewiesen.

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The Influence of Dimethyl Sulphoxide on Metabolic Activity of Ehrlich Ascites Carcinoma Cells and Microorganisms

When studying the mechanism of biological activity of isothiocyanates and their synthetic producers, we have so far constantly been up against the problem of appropriately dosing the above-mentioned substances in a wider range of concentrations related to cell suspensions in the culture medium. The possibility of choosing the solvent in which the required isothiocyanate concentrations can be obtained will naturally be limited by the fact that the solvent employed must not influence the metabolic processes of the organism under investigation. For the purposes mentioned we have for several years utilized methanol, ethanol, diethylene glycol, monoethylether of diethylene glycol and other solvents miscible with water. It was not until recent years that we started employing as a solvent dimethyl sulphoxide (DMSO), mainly for its high dissolving capacity in respect to a wide spectrum of inhibitors rather less soluble in the other organic solvents miscible with water. Its physico-chemical properties, biological effects, metabolism and utilization have already been described in a number of papers[1,2]. There has also been published a number of communications concerning the influence of DMSO on microorganisms[3–5]. Rather less, however, is known about the influence of low concentrations of DMSO on the metabolic processes of cells.

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