The Time Course of Plasma Enzyme Changes Accompanying Skeletal Muscle Stimulation

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Summary. 30 min electrical stimulation of hind limb skeletal muscle in nembutal anaesthetised dogs was accompanied by increases in arterial haematocrit, plasma GOT and plasma LDH, which were almost completed within the first 10 min of stimulation. This fast response indicated that a rapid change in either the entry and/or clearance of enzyme from the plasma must have occurred.

A variety of studies has demonstrated significant increases in the blood plasma levels of many different enzymes following physical exertion[2]. In most instances, samples of blood were obtained directly before and at various time intervals after the exercise; the time course of the changes in plasma enzyme levels during the exercise bout itself was not examined.

As part of an ongoing study into the mechanisms responsible for the exercise evoked plasma enzyme response, we have been looking at the effect of skeletal muscle stimulation on the plasma levels of three enzymes: creatine phosphokinase (CPK), glutamic-oxaloacetic transaminase (GOT), and lactic dehydrogenase (LDH). The methods of skeletal muscle stimulation and of determining the plasma activity of these enzymes has been described previously[3]. To examine the time course of plasma enzyme changes during muscle stimulation, arterial blood samples were obtained at 5 min intervals during 30 min of stimulation (Figure 1). It was apparent that the changes observed during stimulation were almost complete within the first 10 min of stimulation. Thus we saw parallel increases in arterial haematocrit, plasma LDH (PLDH), plasma GOT (PGOT), and plasma protein, with little change in plasma CPK (PCPK).

The similarity of the time course of the changes in PGOT, PLDH and arterial haematocrit was striking. Furthermore, we found that there was a significant correlation between the absolute increase in PGOT or PLDH, and the absolute change in arterial haematocrit, evoked by 30 min of muscle stimulation ($r = 0.79$, $p < 0.05$; $r = 0.87$, $p < 0.05$, respectively; Figure 2). Since the increase in arterial haematocrit during muscle stimulation was likely to have been the result of splenic contraction rather than a reduction in plasma volume, it was considered possible that GOT and LDH were in some way released into the circulating blood during splenic contraction. Initial experiments showed that levels of both GOT and LDH in splenic venous blood increased several fold during splenic contraction induced by topical application of norepinephrine. The total activity involved was too small to account for the plasma enzyme response to skeletal muscle stimulation. This conclusion was supported by results obtained during muscle stimulation in 2 acutely splenectomized dogs (Figure 3). The increases in arterial haematocrit were practically abolished, while sizeable increases in PGOT and PLDH persisted.

Thus the increases in PGOT and PLDH appear to occur early in stimulation and, while related to the extent

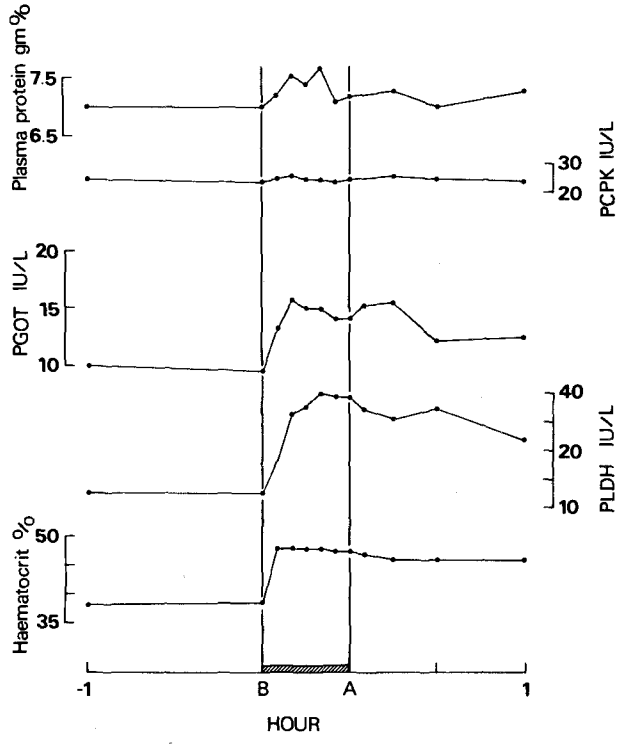

Fig. 1. An example of the changes in plasma enzyme activity, arterial haematocrit, and plasma protein concentration occurring during 30 min of skeletal muscle stimulation. Stimulus applied between B and A.

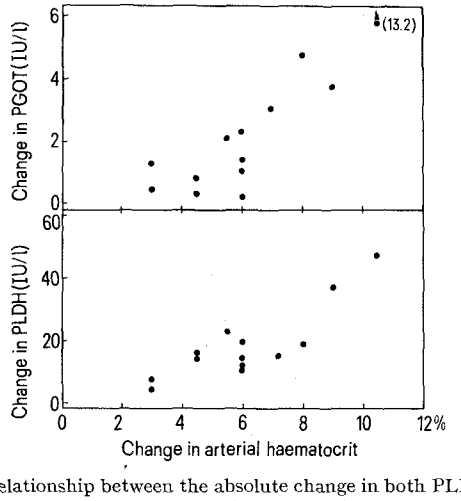

Fig. 2. Relationship between the absolute change in both PLDH and PGOT, and arterial haematocrit during 30 min of skeletal muscle stimulation ($N = 13$).

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[2] E. Schmidt and F. W. Schmidt, in *Biochemistry of Exercise* (Ed. J. R. Poortmans; Karger, Basel/New York 1969), p. 216.

[3] D. J. Loegering and J. B. Critz, Am. J. Physiol. *220*, 100 (1971).

of splenic contraction (haemoconcentration), are not the result of it.

Since both GOT and LDH have relatively short plasma halflives ($t_{1/2}$), whatever promotes their increase early in stimulation must continue until stimulation ceases[4,5]. The time course suggests a rapid step alteration in either the introduction (an increase) and/or the clearance (a reduction) of both enzymes within the plasma compartment upon commencing stimulation. Enzyme clearance is accomplished by several mechanisms[6]. One of these, uptake by the reticulo-endothelial system (RES), is known to clear both GOT and LDH from the extracellular fluid[5]. Since this clearance occurs primarily in the hepatic and splanchnic vascular beds it is conceivable that a reduction in the blood flow through these beds would result in a reduction in plasma enzyme clearance. Such a mechanism may have occurred during muscle stimulation. The degree of splanchnic vascular constriction and splenic contraction would likely be similar since they are both responses to increases in sympathetic activity. Further, the $t_{1/2}$s of GOT and LDH may be sufficiently short to account entirely for the muscle stimulation induced increases in the plasma levels of these 2 enzymes purely through a reduction in the rate of their clearance from the plasma. Data obtained in the dog have indicated a plasma $t_{1/2}$ of approximately 30 min for both the muscle-type and heart-type isoenzymes of LDH[5]. Estimates of the plasma $t_{1/2}$ for the mitochondrial and cytoplasmic isoenzymes of GOT in the dog have yielded values of 54 min and 4.0 h, respectively[4,7].

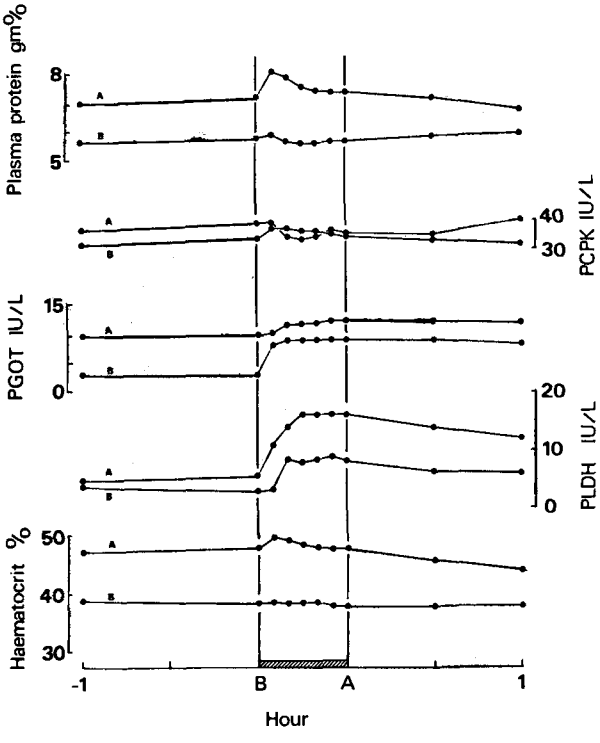

Fig. 3. Changes in plasma enzyme activity, arterial haematocrit, and plasma protein concentration during 30 min of stimulation ($N = 2$) following acute splenectomy.

On the basis of these observations we suggest that one mechanism involved in the plasma enzyme response to exercise is a reduction in the clearance of enzyme from the extracellular fluid in the face of either a normal or increased entry of enzyme into this compartment. One explanation of the reduction in the plasma enzyme response to exercise seen following physical training would therefore invoke a change in the distribution of blood blow during exercise in the trained individual. Such changes have been demonstrated and support this hypothesis[8].

[4] K. G. WAKIM and G. A. FLEISHER, J. Lab. clin. Med. *61*, 86 (1963).
[5] K. G. WAKIM and G. A. FLEISHER, J. Lab. clin. Med. *61*, 107 (1963).
[6] S. POSEN, Clin. Chem. *16*, 71 (1970).
[7] G. A. FLEISHER and K. G. WAKIM, J. Lab. clin. Med. *61*, 98 (1963).
[8] L. B. ROWELL, J. R. BLACKMAN and R. A. BRUCE, J. clin. Invest. *43*, 1677 (1964).

On the Role of the Decreased Renal Vascular Resistance in the Mechanism of Volume Natriuresis

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Summary. Acute hypohysectomy (AH) prevented the increase of the cardiac output, renal cortical blood flow (e.g. the decrease of the renal vascular resistance) and renal sodium excretion during the ECFV expansion. The non-occurrence of natriuresis in AH rats is suggested as being partly in causal relation to the inability to decrease the renal vascular resistance and thus to incrase the peritubular hydrostatic pressure.

Much effort has been concentrated on exploring a possibility of the existence of a humoral inhibitor of the sodium tubular transport, since DE WARDENER's et al.[2] evidence on the tubular nature of natriuresis due to the expansion of the extracellular fluid volume (ECFV) with saline ('saline' or 'volume' natriuresis). The prospective hormone has been called the 3rd factor or a natriuretic hormone[3,4]. However, it was latter found that volume natiuresis in dogs was accompanied by the increased renal blood flow even if the kidney was not connected with the organism by nerves and the blood perfusion pressure was constant[5]. So it was suggested that besides a humoral inhibitor of the sodium tubular transport also a vasodilatory humoral substance might circulate in plasma of dogs with the increased ECFV[5]. An alternative possibility is that a vasoconstrictory agent in plasma may disappear. The resulting vasodilation might then contribute to the inhibition of sodium transport as well. The results of the present experiments are considered to be consistent with these previous suggestions concerning the mechanism of volume natriuresis.

Material and methods. 54 male Wistar rats (Institute of Experimental Endocrinology, Slovak Academy of Sciences SPF breeding) were anaesthetized by Inactin Promonta, then the trachea was cannulated and in 24 rats an acute hypophysectomy was performed by parapharyn-