

GALVANIC INDUCTION OF CELL LYSIS AND ITS EFFECT ON PROTEIN CONTENT AND SUCCINIC DEHYDROGENASE, GLUTAMATE DEHYDROGENASE ACTIVITY LEVELS IN LIVER OF SHEEP

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ABSTRACT

The induction of cell lysis by voltage gradient and its effect on protein content, succinate dehydrogenase (SDH) and glutamate dehydrogenase (GDH) activity levels were studied in the liver of sheep. The cell lysis was found to occur when the liver was exposed to direct current. It is also observed that as the voltage gradient increased from 10 to 30 volts d.c/cm, the cell lysis also increased and the protein content, SDH and GDH activities showed a decline. The decline of SDH activity was higher when compared to that of GDH.

INTRODUCTION

WHEN gastrocnemius muscle and medulla oblongata which have elongated cells were subjected to a selected voltage gradient, earlier investigators¹⁷⁻¹⁹ have found the electromigration of subcellular components which were separated into acidic and basic proteins. The preliminary studies^{16,18} showed that enzyme activities were changed in cathodal and anodal halves in which acidic and basic protein environments predominate. The same work is extended to the liver tissue which is the key centre for most of the metabolic pathways.

MATERIALS AND METHODS

Sheep of similar age and body size group were decapitated and the liver was excised, cooled to 0° C and preserved in the deep freeze. The tissue was washed in Krebs-Henseleit Ringer salt solution (Krebs and Henseleit¹⁵, 1932) and the peritoneal membrane was carefully removed. The tissue was blotted gently between the folds of Whatmann No. 1 filter paper.

VOLTAGE GRADIENT AND PREPARATION OF AH, KH, C₁ AND C₂

The voltage gradient was applied to the tissue as suggested by Nandakumar and Swami¹² (1974) as follows.

The liver was washed in fresh cold Krebs-Henseleit bicarbonate Ringer solution and sliced into 4 approximately equal bits using stainless steel blade. One of the bits was taken in a clean small pyrex beaker cooled at 5° C and this served as the control. The other three bits were subjected to voltage gradients of 10, 20 and 30 volts per cm respectively, for 20 minutes. Temperature of about 5° C was maintained in the set up so that the residual metabolism is reduced to minimum (Swami and Krishnamoorthy¹¹, 1964). At the end of 20 minutes, the liver was quickly sliced transversely into two halves in the centre and these

were designated as the anode half and the cathode half depending on the proximity of the tissue to the electrode with which they were associated. The control bit was also sliced in a similar manner and each half was called C₁ and C₂.

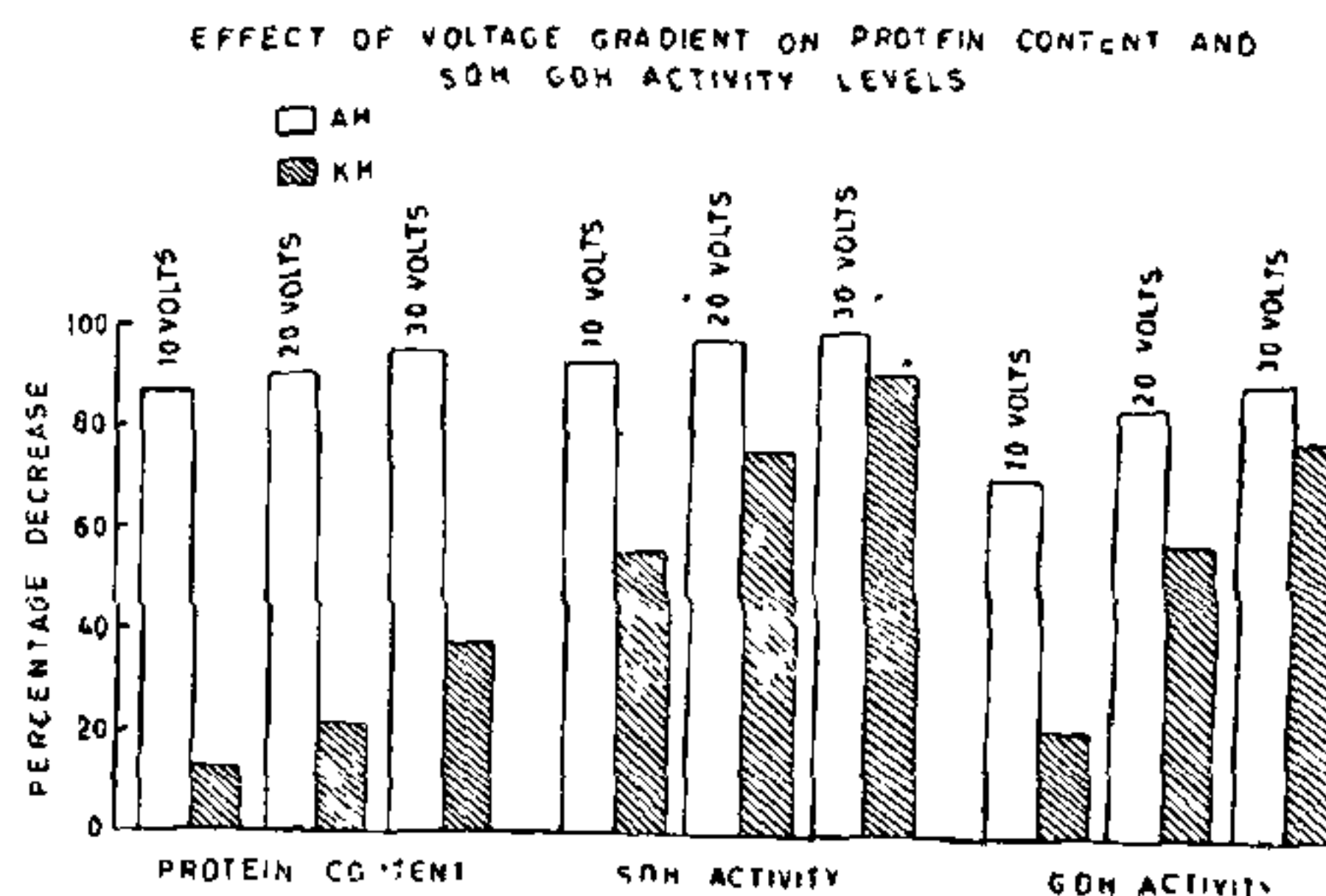


FIG. 1

The control and experimental tissue slices were homogenized in cold 0.25 M sucrose solution and also in distilled water separately with the help of YORCO tissue homogenizer at 2,500 rpm for two minutes. 5% (W/V) homogenates were prepared at 0° C to 5° C and were subjected to centrifugation at 2,500 rpm for 15 minutes. The supernatant fractions were used for further assay. The protein content was determined by the method of Lowry *et al*⁵ using bovine serum albumin as standard. The SDH (EC 1.3.99.1) activity was estimated by the method of Nachlas *et al*⁴ (1960) as modified by Pramamma *et al*⁹ (1975) and GDH (EC 1.4.1.3) activity was estimated by the method of Lee and Lardy (1965) with a slight modification as described by Pramamma and Swami⁹ (1975) and the enzyme activity is expressed in μ moles of formazan/gm wet weight of tissue/hr.

RESULTS AND DISCUSSION

The application of voltage gradient induced a general decrease in protein content in both the AH and KH compared to the control. However in the

experimental halves, the cathodal half showed minor decrement in the protein content while the AH showed a considerable decrement in protein content indicating that the destruction of the cellular protein was less in the KH as compared to AH as a consequence of application of voltage gradient. With the increase in voltage gradient from 10 to 20 and 30 volts/cm, the protein decrement or destruction was more in the anodal halves than in the cathodal halves.

This may be due to the fact that the tissue was exposed to the direct current, the galvanic field spreading from anodal (anelectronic field) to cathodal region (catelectronic field). When the tissue was exposed to such a field the cell lysis has occurred and it is also evident from the results that increase in the voltage gradient leads to the decrease in the protein content in both the AH and KH regions as compared to control.

Similarly, there is a decline in the activities of SDH, and GDH. The per cent inhibition in the case of SDH activity is more when compared to GDH activity. From this, we can infer that GDH is having a higher resistance than SDH to the applied voltage gradient. This may be due to the fact that the GDH is an ammonia detoxifying agent which is not as labile as SDH which is a parent enzyme in the oxidative metabolism.

From the above data it can be inferred that catelectronic field is relatively safer than the anelectronic field to induce cell-lysis. Such an application of selected voltage gradient may be helpful to arrest the tissue overgrowth which occurs in pathological conditions.

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