The ratio ΔG/ΔP is a useful indicator of insular function, being less affected than the Staub effect by malnutrition and other factors that impair carbohydrate storage or utilization. While 39.5% of the apparently normal dogs did not have the Staub effect, only 16.2% of the animals had a ΔG/ΔP ratio smaller than –100, the assumed normal limit. All of the diabetic dogs had a ratio smaller than –100.

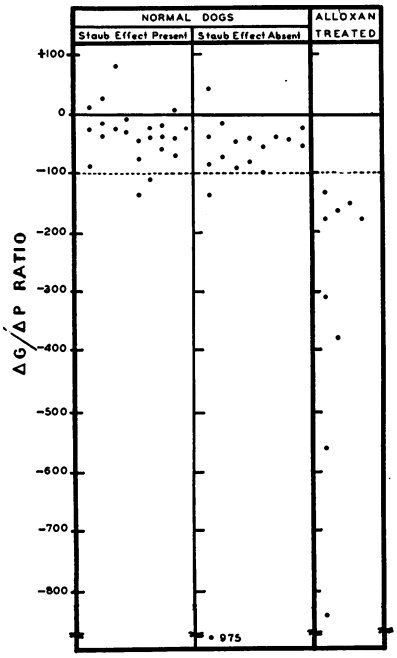

FIG. 1.

The positive values shown had a negative glucose difference, suggesting a faster rate of sugar removal from the blood. When the index is positive, because of a positive phosphorus difference, it means there is a defective production of insulin.

The results in diabetic patients and in normal human subjects will be reported elsewhere; they are in agreement with the experimental data obtained.

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Amino Acids in the Mitochondrial Fractions of Tissues as Determined by Paper Partition Chromatography

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The biochemical properties of intracellular structures identified as mitochondria have received considerable attention recently. However, little is known of the nature of the protein moiety of these important cell constituents. As a first step in the characterization of mitochondrial protein, the amino acids in acid hydrolyzates were studied by two-dimensional paper chromatography (*1*, *2*).

The method used for the isolation of the mitochondria was essentially that of Hogeboon *et al.* (*3*) with the exception that additional cycles of low and high speed centrifugation were employed to insure the attainment of maximal uniformity of the sedimented material. The isolated particles were the same size and shape as the structures identified as mitochondria in smears made from homogenates, and in free cells found in the sediment from the first low speed centrifugation. These particles possessed the same staining characteristics, with Janus green B before fixation, and with aniline-acid fuchsin after fixation with osmic acid, as do mitochondria in cells. The isolated mitochondria were hydrolyzed with 6 *N* HCl in sealed tubes for 24 hr and aliquots were subjected to chromatography. Samples were treated with H_2O_2 to enable detection of cystine as cysteic acid and methionine as methionine sulfone. Numerous chromatograms were

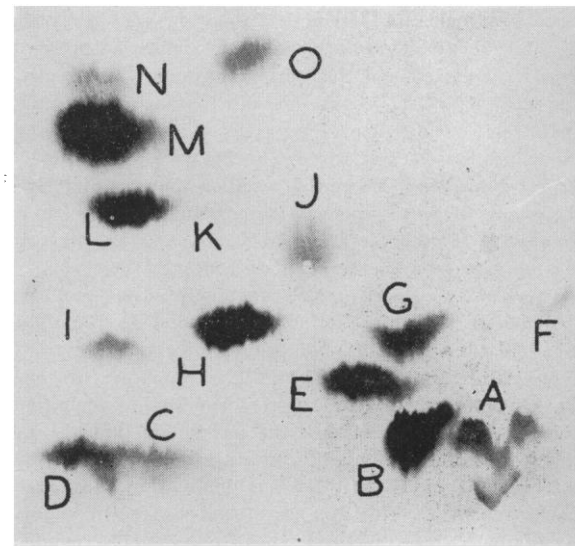

FIG. 1. Photograph of chromatogram from hydrolyzate of rat kidney mitochondria. (A) Aspartic acid, (B) glutamic acid, (C) lysine, (D) arginine, (E) glycine, (F) cystine (cysteic acid), (G) serine, (H) alanine, (I) histidine, (J) threonine, (K) methionine (methionine sulfone), (L) valine, (M) leucine and isoleucine, (N) phenylalanine, (O) tyrosine.

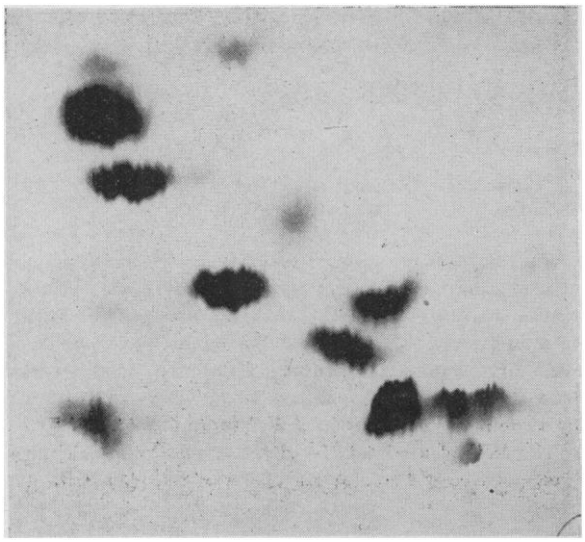

FIG. 2. Photograph of chromatogram from hydrolyzate of mouse pancreas mitochondria.

run on each sample, and comparisons between samples were made for chromatograms on which the amino acids occurring in the highest concentrations gave spots of approximately equal area and intensity, as shown in Figs. 1 and 2. Mitochondrial fractions of the liver and kidney of the mouse and rat, and of pancreas, mammary gland, hepatoma, squamous cell, and mammary carcinomas in the mouse were analyzed.

Photographs of two typical chromatograms are shown in Figs. 1 and 2. The visible constituents are identified on Fig. 1. The yellow spots given by proline and hydroxyproline are not visible on the photographs because of insufficient contrast, although they were visible on the original chromatograms. Eighteen amino acids were clearly identified on the chromatograms. Tryptophan was not observed because of destruction during acid hydrolysis. Glutamic acid, aspartic acid, glycine, alanine, serine, proline, the leucines, and valine were present in the largest quantities. Somewhat smaller amounts of lysine, arginine, threonine, phenylalanine, and tyrosine were found. Methionine, cystine, histidine, and hydroxyproline were present in the smallest concentrations.

There was a remarkable constancy in the relative amounts of the chief amino acids found in the mitochondria from the different samples of tissue examined. This is illustrated in Figs. 1 and 2, in which preparations from different organs of different species are compared. The chromatograms suggest the possibility that the rat kidney mitochondria might contain slightly more lysine and histidine and slightly less methionine than those from mouse pancreas. This will have to be checked by quantitative procedures. The patterns of amino acids found in the preparations from the malignant tissues examined were virtually identical with those obtained from normal mouse tissues.

The similarity of the amino acid patterns found in the mitochondrial fractions of the various tissues studied suggests that there is a characteristic protein, or combination of proteins, associated with these particles. The results also indicate that the quantity of the dicarboxylic amino acids exceeds that of the basic amino acids.

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On the Participation of the Reticulo-endothelial System in the Alarm Reaction

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We wish to report upon experiments which revealed a marked increas in phagocytic activity of the reticulo-endothelial system during the alarm reaction, that is, during the first phase of the general adaptation syndrome.

The reticulo-endothelial system probably participates in the defense of the organism through its phagocytic activity, and by production of antibodies, agglutinins, antitoxins, etc. (*1, 4*). A local increase in the activity of this system has been described in the thymus (*5*) after exposure to various acute nonspecific stresses, and in the spleens of rats subjected to chronic inanition (*3*) or injected with cortical extracts (*2*). Stimulated by these findings, we have performed two series of experiments in order to clarify the relationship between the reticulo-endothelial system and the hormonal and metabolic changes which occur during the alarm reaction.

In our first experiment, 44 piebald male rats (average body weight 150 g) were divided into four equal groups: group I served as untreated controls; groups II, III, and IV were fasted 48 hr, and during the last 24 hr were submitted to various stresses, such as cold (0 – 5° C), spinal cord transection (at the height of the 7th cervical vertebra), and repeated, exhaustive, forced exercise. All animals were injected intravenously with 2 ml of a dilute solution of Higgins India ink (1 part India ink to 5 parts physiologic NaCl solution) 1 hr before they were killed. At autopsy, naked eye inspection showed, in all the stressed animals, a markedly increased deposition of India ink in the lung, kidneys, adrenals, bone marrow, and the "hibernating gland." In the hibernating gland, this was accompanied by an acute discharge of lipid granules, hyperemia, and edema. These changes are characteristic of the alarm reaction and have been given special attention elsewhere (*6*). Compared to the controls, the India ink deposition in the liver of the alarmed rats did not seem to be significantly increased, while in the spleen there was a diminution of India ink deposition.

Subsequent histological examination confirmed and extended the autopsy findings. There was increased phagocytosis in the lung, kidneys, adrenals, bone marrow, hiber-