TOTP demonstrated a marked ability for time dependent inhibition of both Na^+, K^+-dependent and Mg^{++}-dependent ATPases (Figure 1). At the lowest concentrations used (0.25 m*M*) Mg^{++}-dependent ATPase activity was depressed to a much greater extent than the Na^+, K^+-dependent component. At higher concentrations, however, the Na^+, K^+-dependent enzyme activity was more susceptible.

Mevinphos uniformly inhibited Na^+, K^+-activated ATPase to a greater extent than Mg^{++}-dependent enzyme (Figure 2). In all cases after the addition of inhibitor there was at first a rapid decline of activity for a short period, followed by a gradual inhibition which appeared to be exponential with time.

The observed initial drop in ATPase activity produced by mevinphos may represent a rearrangement of the ATPase protein in the lipid matrix of the membrane. The extent of this initial depression of activity is greater for the Na^+, K^+-stimulated activity, which may coincide with the possible location of the Na^+, K^+-stimulated enzyme in the external plasma membrane and therefore its ready availablility to the medium[11].

In the light of recent findings it appears that several apparently unrelated reports may well involve ATPase inhibition. BULLOCK et al.[12] reported the inhibitory effects of organophosphates on axonal conduction to be either reversible or irreversible depending on the duration of exposure and concentration of inhibitor. HOSKIN et al.[13] used 3 potent organophosphate acetylcholinesterase inhibitors to examine nerve conduction, acetylcholinesterase inhibition, and inhibitor penetration. The authors noted, as observed previously[12], that irreversible inhibition of axonal conduction was concentration and time dependent, and that the concentrations of inhibitor needed were far above those needed for acetylcholinesterase inhibition. These workers proposed that the observed effects possibly occurred due to the binding of the compounds to an unspecified membrane component. The possibility of a mechanism of nerve conduction inhibition which is irreversible, slower, and not related to acetylcholinesterase inhibition was, therefore, broached.

Because of the role of Na^+, K^+-ATPase in the maintainance of transmembrane ionic gradients[14] and the secondary involvement of this gradient in amino acid and sugar transport[15], its inhibition by organophosphates could result in extensive neuronal damage. Also, the disruption of utilization of ATP by ATPase within the synaptic area could alter energy metabolism of the nerve terminal by secondarily altering the activities of other enzymes for which ATP or ADP may be allosteric effectors. The inhibition observed for the neurotoxic compound TOTP and previously reported for DFP, therefore, could conceivably provide a plausable explanation for the delayed neurotoxicity found in association with exposure to these compounds. The lack of such neurotoxicity of mevinphos may be explained on the basis that this compound does not accumulate in the nervous system[16]. TOTP, on the other hand has been shown to persist in the nervous system[17]. In our studies reported previously, TOTP accumulated in brain and spinal cord of chicken after a single effective dose to the extent of 90 µg/g tissue (ca. 0.25 m*M*), a concentration that has been shown to be inhibitory to ATPases in this report.

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The Glycocalyx of the Epithelial Cells of the Colon, Observed in Normal and Ulcerous Colitic Conditions

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Summary. Biopsies of subjects affected by ulcerous colitis were stained with ruthenium Red. Alterations of the cellular coat and glycocalyx of the epithelial cells of the colon were identified.

The importance of the 'cell coat' in many functions of cellular cycle has been well established[2-4]. JOHNSON[5] showed that maltase and invertase enzymes are located in the brush-border's glycocalyx of the small intestine's absorbing epithelium. This coat probably takes a prominent part during the process of absorption of several substances and is also considered to be the site of a number of antigenical cellular receptors. The digestive enzymes seem to be strictly related to the basal areas of the glycocalyx and represent the so-called 'coat strictly attached'. WILLIAMS and MCKENZIE[6] described significant cell coat's variations in the small and large intestine of mice. In fact, the epithelial cells of jejunum-ileum and colon regions showed a remarkable structural likeness, and a great difference was also noticed between the two superficial glucide-coats.

Further, MORGAN[7], DULBECCO and STOKER[8], MARTINEZ-PALOMO and WIRBR[9] observed significant variations of the polysaccharidic substance at the surface of cells infected by virus. These variations might be related to the phenomena showed from infected cells, as, for in-

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stance, those in which a cellular contact inhibition is described.

The purpose of this note is to describe the polysaccharidic coat (glycocalyx) in the colon's epithelium of some normal subjects, and of other ones affected by ulcerous colitis. In the present investigation, the whole course of the illness, from beginning till end, has been followed.

Material and method. Biopsies were reduced to small fragments and rapidly fixed in 2.5% glutaraldehyde in cacodilate buffer (0.2 *M*; pH 7.2) for 2 h. The fragments, washed in cacodilate buffer for 30 min, were postfixed for 3 h in 1.33% OsO_4. Ruthenium red, previously purified, was added to the solution of osmium in concentration of 1500 ppm according to LUFT's[10] method. The blocks, were then washed, dehydrated in alcohol and embedded in Epon 812, sectioned with a Porter Blum MT1 ultramicrotome and studied in a Zeiss E/M 9A

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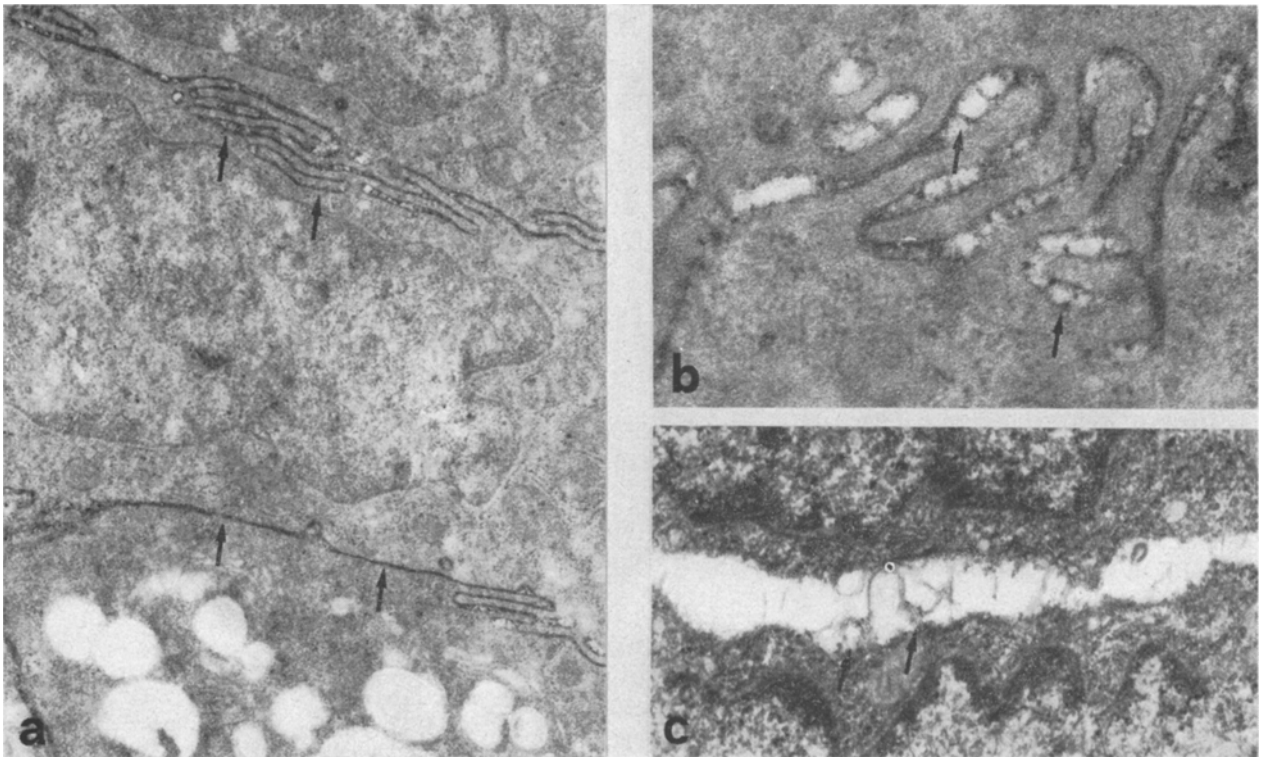

Fig. 1. a) Epithelial cells of normal human colon (× 16,000, unstained). b) and c) Ulcerative desease of the colon. b) × 43,000; c) × 18,000; unstained. Arrows: cell coat.

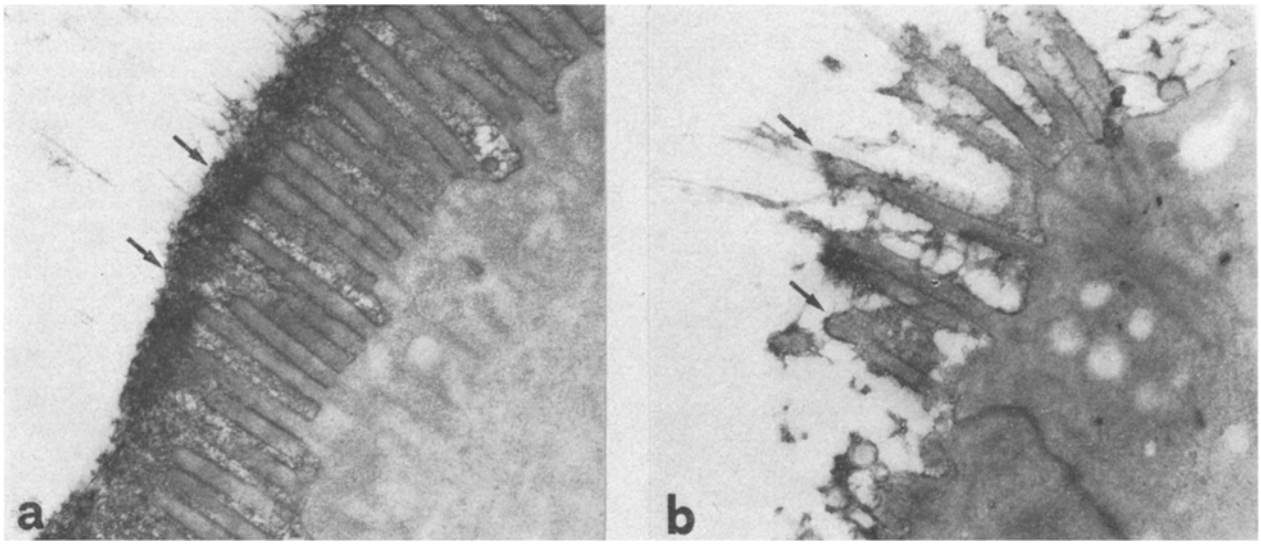

Fig. 2. a) Epithelial cells of normal human colon (× 7,000, unstained). b) Ulcerative desease of the colon. (× 7,000, unstained). Arrows: glycocalyx.

electron microscope. Histological sections stained with E.E. have been used as control in order to evaluate the stage of the illness.

Results. The cell surface of the colon epithelial cells in normal individuals is formed by a glycoproteinous substance rather sharp and apparently cementing the adjacent cells (Figure 1a). Colitis makes the cell coat thinner and the epithelial cells appeared to break off in the sites where the effects of the pathological phenomena were more patent. In these sites, fibrillar bridges of polysaccharidic-like material appeared to be stretched from one membrane to the other among the same population of cells (Figure 1b). In cells completely detached, their 'glycocalix' appeared clearly discontinuous and the plasma membrane showed a characteristically irregular and fibrillar-like outline (Figure 1c).

On the contrary in normal conditions, the glycocalyx appeared as a continuous coat coating the surface of the microvilli regular in number and disposition (Figure 2a). In pathological conditions, the glycocalyx was not so clearly evident and seems to form an extremely loose, irregular net which left unsheltered the most part of the microvilli. In these cases the microvilli were less numerous and showed a very irregular disposition (Figure 2b).

Discussion. The morphological alteration observed in the glycocalyx of the colon epithelium is probably due to the pathogenetic processes, and this, in turn, might be related to the action of lymphocytes. In fact, these lymphocytes, sensitized during the pathological process as antimucus-antibody-lymphocytes, might be responsable for the alteration of the glycocalyx, in all the above epithelial structures[11,12]. But it is also possible that the action of specific virus might alter not only the genetical patrimony of the cell, but also the whole mechanism of proteinuous synthesis. Then, in this case, the virus might be able to modify the biochemical and histochemical composition of both the cellular coat and glycocalyx.

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Evidence of Diurnal Fluctuation of Sensitivity to Noradrenaline in the Rat - the Role of the Thyroid

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Summary. The capacity for heat production, under the influence of the same amount of noradrenaline, in the rat was significantly higher in the evening (20.00 h) than in the morning (07.00 h). Thyroidectomy produces not only a lower level of heat production, but also a complete disappearance of the differences between the morning and the evening experiments.

In mammals and birds the existence of diurnal fluctuations was demonstrated in the general metabolism, body temperature, adrenocortical function, catecholamines, sodium and potassium excretion, urine volume excretion as well as in some other functions[2,3]. It was also found that oxygen consumption in the rat adapted to 29 °C was higher in the evening and during the night than in the morning. This difference disappeared completely after the thyroidectomy[4]. It is well established that noradrenaline produces an increase in the heat production in the rat and mouse adapted to cold or to thermoneutral zone[5–10]. However, until now no data have been available concerning the changes of the sensitivity to noradrenaline during the day and night.

Material and methods. Observation was made in 6 groups of albino male rats of Wistar strain, weighing 180–200 g each group consisting of 10 animals. Rats were adapted to room temperature (19–22 °C) for about 4 weeks, with daily illumination, food and water ad libitum. Noradrenaline (Galenika) was injected i.p. in doses of

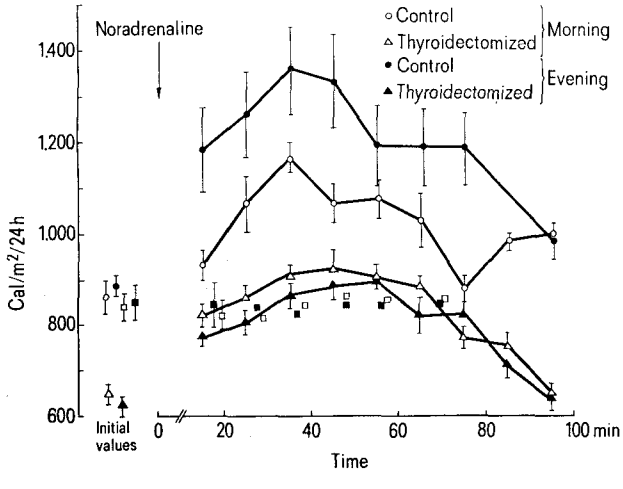

Fig. 1. The effect of noradrenaline (1.6 mg/kg) on the heat production in the rat adapted to 19–22 °C and measured at 30 °C.
Normal control: measurement made in the morning (07.00 h) ○ – ○; in the evening (20.00 h) ● – ●. Thyroidectomized: measurement made in the morning (07.00 h) △ – △; in the evening (20.00 h) ▲ – ▲. Physiological solution: measurement made in the morning □ – □; in the evening ■ – ■. Mean ± SEM of 10 animals.

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