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Prevention by Ascorbic Acid of Liver Glycogen Depletion in Endotoxin Intoxication

IT is well established that there is a striking depletion of liver glycogen in endotoxin intoxication, and insulin, glucose and lactate are relatively ineffective in increasing liver glycogen storage[1]. This depletion of liver glycogen is not necessarily due to epinephrine discharge or to circulatory disturbances attendant on intoxication, but it seems to be related to the accumulation of endotoxin in liver[2]. The characteristic prostration common to all animal species in the later stage of endotoxin intoxication seems to be related to this depletion of glycogen[2]. It is especially pronounced in adrenalectomized animals, and death results from hypoglycæmic deterioration with or without cramp. It is well known that pretreatment with cortisone prevents death[1]. Although the mechanism of glycogen depletion has been discussed in relation to inhibition of hepatic enzymes, especially succinic dehydrogenase[3], or to the effect on mitochondria leading to oxidative uncoupling[4], no conclusive evidence indicating special relations of endotoxin to these disturbances could be obtained. In this respect it has been shown that the effect of endotoxin in depleting liver glycogen in rabbits is much lessened in the hot summer season even in adrenalectomized animals[5]. It has therefore been suggested that the depletion of liver glycogen might be determined by the metabolic response of liver induced by the uptake of endotoxin in the hepatic reticuloendothelial cells. Here it was shown that saturation with ascorbic acid could ameliorate the depletion of liver glycogen in cold weather in which the susceptibility of the animal is markedly enhanced.

Albino rabbits weighing about 2·5 kg were used. They were maintained by 'Okara' (soy-beans curds waste 300 g with salt 0·5 g a day). Ascorbic acid (500 mg/day, mostly 'Hi-C' tablet of Takeda Chemical Industries, Ltd., Osaka) was mixed with 'Okara'. For attaining blood total ascorbic acid level at the saturation-level (about 1·2 mg/dl., DNP method), at least 4 days' administration was necessary. Experiments were performed both on intact and adrenalectomized animals in order to determine the effect of ascorbic acid saturation independent of glucocorticoids with which ascorbic acid has been claimed to be synergistic[6]. It has been repeatedly shown that adrenalectomized rabbits do not necessitate any replacement treatment and eventual existence of accessories can easily be checked by a water diuresis test[7]. Purified endotoxin (300 μg/kg of *Salmonella typhosa*, Difco) was administered intravenously after fasting overnight. Liver specimens for glycogen analysis were taken after 3 h under light 'Nombutal' anæsthesia (20 mg/kg, intravenously).

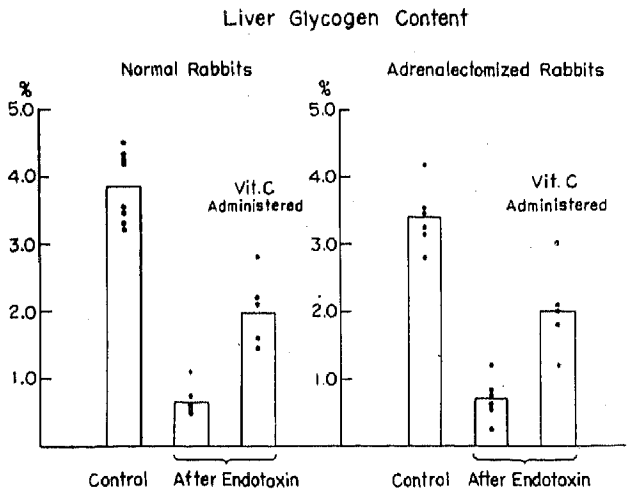

Fig. 1. Prevention of liver glycogen depletion due to endotoxin by ascorbic acid saturation (3 h after endotoxin)

As shown in Fig. 1, ascorbic acid saturation could markedly ameliorate the depletion of liver glycogen after endotoxin both in intact and adrenalectomized animals. With this the attendant hypoglycæmia was completely prevented and in intact animals the preceding initial hyperglycæmia due to epinephrine discharge[2] became marked and prolonged. Even adrenalectomized animals could tolerate the toxic dose used here (100 per cent lethal for adrenalectomized rabbits[2]). Thus the effectiveness of ascorbic acid in preserving liver glycogen independent of glucocorticoids could be substantiated. There was no indication that ascorbic acid consumption might be increased in endotoxin intoxication, since no appreciable changes in ascorbic acid content (total and reduced) in blood and liver occurred after endotoxin. Therefore, the effectiveness of ascorbic acid did not seem to be concerned with the supplement of ascorbic acid consumed. No gluconeogenetic effect[8] could be expected. Stabilization of hepatic metabolism by its antioxidative effect seemed to be the underlying mechanism. In cases of severe endotoxin intoxication the blood glutathione (GSH, after Brückmann and Wertheimer) became reduced (from about 30 to 20 mg/dl. 3 h after endotoxin inoculation). This was prevented with lessening of the intoxication by ascorbic acid saturation, just as the case with tolerant animals.

Saturation with ascorbic acid did not materially increase the ascorbic acid content of liver (13–18 mg per cent, total ascorbic acid) and yet could show the foregoing remarkable effect. In this respect, the observation by Fukuda and Horiuchi[9] in man that, with increasing saturation with ascorbic acid, the consumption of ascorbic acid estimated by the deficit in the daily urinary output compared with the amount ingested increased markedly might throw some light on this problem. In any event, significance of ascorbic acid saturation could be evaluated in endotoxin intoxication leading to hepatic glycogen depletion.

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