It would appear that these phenomena have previously been overlooked on account of the relatively narrow range of conditions under which they occur.

A full account of the completed work will be published elsewhere.

Protein solubilities were determined by Mr. J. I. M. Ironside and Mr. I. Robertson. Nucleic acids were determined by Miss Eleanor Brown. The work described in this communication is part of the programme of the Food Investigation Organization of the Department of Scientific and Industrial Research.

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Protective Action of Vitamin B_{12} in Experimental Liver Injury

There are several reports concerning the biochemical changes accompanying experimental liver damage by administration of carbon tetrachloride and other steatogenic poisons. However, the fundamental metabolic deviation causing fatty degeneration has not been well understood. Patwardhan *et al.*[1] have demonstrated that marked changes in the distribution and quantity of several constituents of the liver cells occur before structural damage to the organ. In the present communication, it is shown that a rapid rate of depletion of soluble sulphydryl compounds is among the major derangements in liver injury by carbon tetrachloride. The possibility has been indicated that the protection afforded by prior administration of vitamin B_{12} may be due to its known influence on sulphydryl metabolism[2].

Adult male rats (Wistar strain), 100–150 gm. in weight, reared on the laboratory stock diet, were given, intraperitoneally, vitamin B_{12} (10 μgm./animal) 3 hr. prior to carbon tetrachloride administration (2 c.c. per kgm. body-weight). It was ascertained from preliminary experiments that protection by the vitamin was best under these conditions and that animals not receiving the vitamin showed maximum liver fat accumulation at the end of about 48 hr., after which there was regeneration. Animals were killed at intervals after carbon tetrachloride injection and the livers quickly excised, chilled and homogenized in cold isotonic sucrose to a 20 per cent suspension. Determinations were made on aliquots of the whole liver and on centrifugally separated fractions. The values reported (Table 1) are averages for four animals from each group and are for the whole liver only, sampled at the end of 6 and 48 hr. after carbon tetrachloride poisoning and employing the analytical procedures cited.

It is seen that liver fat accumulation proceeds even after restoration of glutathione to normal levels and that vitamin B_{12} protects against rapid depletion of glutathione. A study of the distribution of glutathione in cell fractions has shown that while the mitochondrial fraction is completely devoid of it, nearly 80 per cent is present in the supernatant, the remainder being accounted for in the nuclei. Nuclear bound glutathione does not become depleted as a result of carbon tetrachloride injury. Mitochondrial damage in carbon tetrachloride injury can be inferred from alterations in pyridino-protein nucleotides[8] and lability of other acid-soluble nucleotides[9].

The protective action of vitamin B_{12} is more marked with respect to changes in ribonucleic acid and in phospholipids than in neutral lipids. It has apparently no effect on glycogen losses in the prenecrotic and necrotic stages. Deoxyribonucleic acid and methionine values remain unchanged throughout.

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Lability of Intramitochondrial Components in Experimental Liver Injury

The mitochondrial membrane has a limited ability to retain nucleotides and other co-factors like Mg^{++} which are essential for maintaining normal respiration and efficient coupled phosphorylation[1]. Exposure to hypotonic conditions, ageing, other environmental alterations and physiological states cause swelling of the mitochondria accompanied by biochemical modi-

Table 1. Changes in Liver Constituents in Injury by Carbon Tetrachloride

	Untreated control	Hours after administration of carbon tetrachloride			
		6		48	
		Without vitamin B_{12}	Vitamin B_{12}-protected	Without vitamin B_{12}	Vitamin B_{12}-protected
	(mgm./gm. fresh weight)				
Total lipids (ref. 3)	23·6 ± 1·5	29·1 ± 1·2	28·2 ± 1·1	48·8 ± 2·5	34·9 ± 1·5
Phospholipids (ref. 3)	15·1 ± 0·2	15·1 ± 0·3	15·5 ± 0·2	10·6 ± 0·2	14·6 ± 0·2
Ribonucleic acid (ref. 4)	10·6 ± 0·6	8·8 ± 0·6	9·4 ± 0·6	6·9 ± 0·9	10·3 ± 0·3
Deoxyribonucleic acid (ref. 4)	3·8 ± 0·3	3·6 ± 0·3	3·7 ± 0·3	3·6 ± 0·3	3·6 ± 0·3
Glutathione (ref. 5)	1·4 ± 0·2	0·4 ± 0·2	1·3 ± 0·2	1·2 ± 0·1	1·7 ± 0·1
Glycogen (ref. 6)	3·3 ± 0·5	1·0 ± 0·2	1·5 ± 0·3	0·7 ± 0·2	0·6 ± 0·4
Methionine (ref. 7)	5·0 ± 0·5	4·9 ± 0·5	5·0 ± 0·3	4·9 ± 0·3	5·0 ± 1·0