method of Aschaffenberg and Drewry[3] with minor modifications was employed. Preliminary electrophoretic experiments in a Tiselius apparatus, of crystalline buffalo β-lactoglobulin, indicated its isoelectric point to be approximately 5·25 and the electrophoretic patterns obtained near the isoelectric point with acetate buffers of 0·1 ionic strength were found to be asymmetrical or partially resolved on the descending side and hypersharp on the ascending side. Sedimentation experiments with a Spinco model *E* analytical ultracentrifuge with protein concentrations up to 6·8 per cent and at 20° C. and low temperatures (about 8° C.) gave symmetrical patterns. These results show that there is close similarity between physico-chemical properties of buffalo β-lactoglobulin and β-lactoglobulin *B* of cow's milk[5,6].

Further work is in progress and details will be published elsewhere.

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Intracellular Distribution of Ubiquinone in Rat Liver under Certain Stress Conditions

Observations on the effects of pantothenic acid deficiency on mitochondrial composition in relation to oxidative metabolism in rat liver were reported earlier[1]. In the present communication, observations on intracellular distribution of ubiquinone in the pantothenic acid-deficient rat liver are presented. The effects of vanadium administration and of vitamin A-deficiency have also been studied, since vanadium is known to cause decreased hepatic coenzyme *A* (ref. 2) and vitamin A-deficiency results in increased ubiquinone-level[3].

Dietary deficiencies of pantothenic acid and vitamin A were produced by feeding weanling rats (40–50 gm.), Wistar strain, a 10 per cent purified casein ration deficient in the respective vitamin and replete in all other respects, for a period of 8 weeks. To one group of rats, fed the casein ration with all the vitamins, ammonium vanadate was administered intraperitoneally (0·6 mgm./rat/week), throughout the experimental period. The animals were killed, livers quickly removed and chilled in cracked ice. Livers were homogenized in isotonic (0·25 *M*) sucrose, nuclear and mitochondrial fractions separated in a *PR*-2 International refrigerated centrifuge[4] and microsomes in a Spinco preparative centrifuge[5]. Determinations of coenzyme *A*, ubiquinone and mitochondrial succinoxidase activity were as described before[1]. The supernatant fraction was freeze-dried before saponification in the assay procedure for ubiquinone.

The results (Table 1) show normal rat liver ubiquinone to have an average distribution of 29·1 per cent in the nuclei, 37·8 per cent in the mitochondria, 14·9 per cent in the microsomes and 5·5 per cent in the supernatant fraction. Of the total coenzyme *Q*, 84–93 per cent was recovered in the various cytoplasmic fractions.

Vanadium administration, as also pantothenic acid-deficiency, result in marked and very similar decreases in total coenzyme *A*, mitochondrial ubiquinone and succinoxidase activity. This close parallelism suggests that the reduced hepatic coenzyme *A*-level is the primary cause of the decrease in ubiquinone; the involvement of coenzyme *A* in the biosynthesis of the isoprenoid side-chain of the ubiquinone molecule is known (cf. ref. 1). Vitamin A-deficiency results in elevated levels of ubiquinone in all the cell fractions, being particularly marked in the microsomal and supernatant fractions and is unattended by any change in succinoxidase activity.

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Extraction of Nucleic Acid-free Lipopolysaccharides from Gram-negative Bacteria

The extraction of Gram-negative bacteria by the hot phenol method of Westphal[1] yields a lipopolysaccharide along with considerable amounts of nucleic acid. The nucleic acid can be separated from the high molecular weight lipopolysaccharide by high-speed centrifugation, but to reduce the nucleic acid to a level of less than 2 per cent may require four or five sedimentations.

Table 1. Intracellular Distribution of Ubiquinone in Rat Liver

Group	Ubiquinone					Coenzyme *A* units	Succinoxidase activity (μl.oxygen/hr.)
	Whole liver (μgm.)	Nuclei (μgm.)	Mitochondria (μgm.)	Microsomes (μgm.)	Supernatant (μgm.)		
10 per cent casein	127 ± 7·6	37 ± 4·1	48 ± 2·7	19 ± 3·1	7 ± 1·6	164 ± 11	1,304 ± 32
10 per cent casein + vanadium	97 ± 9·9	33 ± 4·3	33 ± 1·9	13 ± 1·8	6 ± 0·9	89 ± 17	996 ± 51
10 per cent casein − pantothenic acid	103 ± 3·1	33 ± 5·6	36 ± 4·1	17 ± 2·4	6 ± 1·1	96 ± 6	1,031 ± 69
10 per cent casein − vitamin A	187 ± 11·6	46 ± 3·9	61 ± 6·2	34 ± 2·4	17 ± 4·1	—	1,361 ± 98

Results are averages of four independent determinations ± *S.E.*, and are expressed per gm. fresh liver weight.