capacity was barely measurable. That histidine was destroyed during photo-oxidation was demonstrated by thin-layer chromatography of acid hydrolysates and by estimation of histidine with bacterial histidine decarboxylase.

These results indicate that the binding of copper to protein in *M. trunculus* haemocyanin involves imidazole groups, although the possibility that other amino acid residues also participate cannot be completely excluded.

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The Effect of Isolation Conditions on the Polyamine Content of Rat-Liver Microsomes

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Recently evidence has accumulated to suggest that polyamines are associated with cellular RNA and its biosynthesis. Raina & Telaranta (1967) have reported the effect of various ions on the polyamine content of the microsomes isolated in a sucrose medium, and of the ribosomes isolated from them by disruption with deoxycholate. In the present work similar experiments have been carried out using microsomes and ribosomes isolated in an ionic medium.

When microsomes are isolated in an ionic medium (Zamecnik & Keller, 1954) and given a single wash in the same medium they contain approximately 15% of the total spermidine and 40% of the total spermine present in the homogenate. When sucrose medium (Hogeboom, 1955) is used a substantially higher proportion of both polyamines are present in the microsome fraction. The effect of dialysing the microsomes isolated in ionic medium against four changes of NaCl solutions of various concentrations was tested. When the concentration of NaCl was increased above 0·1M all the spermidine was removed, and increasing to 0·5M removed all the spermine. When NaCl was replaced by $MgCl_2$ the corresponding concentrations were 0·05M and 0·07M. Thus Mg^{2+} ions are more effective than Na^+ ions at removing polyamines, the effect appears to depend partly on the ionic strength of the cation.

The ability of microsomes isolated in ionic medium to exchange their polyamines with those of other cell fractions was tested. Liver microsomes were isolated from rats injected with [1,4-$^{14}C_2$]-putrescine. Unlabelled microsomes were isolated in a parallel experiment. The unlabelled microsomes were mixed with labelled nuclei, mitochondria, and cell sap and a homogenate reconstituted. The microsomes were then re-isolated. The specific activities of spermidine and spermine in the original labelled microsomes and the re-isolated microsomes showed that extensive exchange had occurred. When ribosomes were isolated according to Von der Decken & Campbell (1962) using deoxycholate and Lubrol W they contained less than 20% of the polyamines present in the microsome fraction.

It is concluded that the amounts of polyamines present in isolated microsomes depends largely on the isolation procedure and it is possible to prepare ribosomes which are active in protein synthesis and which contain only a small proportion of the total cellular polyamines.

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Removal of Ammonia by Formation of Amino Acids in Rat Liver

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When liver of fed rats was severed from the blood circulation (and the urea cycle ceased to function) the alanine content increased from 0·87 ± 0·64 μmoles/g. wet. wt (mean value 6 rats ± S.D.) at 0 min. to 1·53 ± 0·93 after 2 min. ischaemia and to 2·26 ± 0·60 after 5 min. The tissue content of ammonia, aspartate and glutamate did not alter significantly during this period. Inhibition of alanine aminotransferase by preinjection with L-cycloserine (Otto, 1965) prevented the accumulation of alanine. In these experiments rat liver was freeze-clamped, powdered and extracted with perchloric acid as described by Williamson, Lund & Krebs (1967) and the metabolites were estimated by enzymic methods (Williamson, Lopez-Vieira & Walker, 1967). There was also a rapid increase in the concentration of alanine when fed rats were given an intraperitoneal injection of 2·5 m-moles of NH_4Cl/kg. body wt. and their livers were analysed