partial exocytosis induced by the Ca^{2+} influx attending the arrival of an action potential at the varicosities (see Smith, 1972).

At present little is known about the mechanisms responsible for the translocation of dense-cored vesicles along the axon. The process is energy-dependent and independent of protein synthesis. Although the vesicles are formed in the cell body, their motion is generated locally within the axon, since they continue to move in a centrifugal direction within axons isolated from their cell bodies. Recently interest has focused on a possible involvement of axonal microtubules in translocation of noradrenaline-storage vesicles. Drugs such as colchicine and vinblastine sulphate, which are known to disrupt microtubular structure, are able to prevent the movement of storage vesicles within axons. The correlation between the dispersal of organized microtubules and inhibition of movement holds over a wide range of colchicine concentrations. Further, there is some morphological evidence, albeit incomplete, for an association between microtubules and dense-cored vesicles (Banks *et al.*, 1971, 1972). Better evidence for cross-bridges between microtubules and presumptive synaptic vesicles has been provided in electron micrographs of the central axons of the lamprey by Smith (1971). It is conceivable that a movement generating interaction between microtubules and vesicles could be mediated by an ATP-degrading enzyme similar to dyenin. The latter is attached to microtubules in

Tetrahymena cilia and has been characterized by Gibbons (1966).

A protein able to bind colchicine and probably identical with the microtubular subunit has been detected in extracts of postganglionic sympathetic-nerve trunks. Studies are being undertaken to discover whether it is able to bind, directly or indirectly, to dense-cored vesicles.

Vesicles are not the only organelles able to move within axons; mitochondria appear to have the ability to move both centrifugally and centripetally in a manner that is independent of axonal continuity with the cell body. Available evidence suggests that mitochondrial movement is not dependent on the structural integrity of the axonal microtubules.

The paper to be presented will discuss methods that have been used in the study of axonal transport and will review what is already known of the process and its mechanism in postganglionic sympathetic neurons.

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COMMUNICATIONS

The Exocytosis of Polymorphonuclear-Leucocyte Lysosomal Enzymes Induced by Cytochalasin B

By PHILIP DAVIES, A. C. ALLISON, R. I. FOX, M. POLYZONIS and A. D. HASWELL (*Clinical Research Centre, Watford Road, Harrow, Middlesex HA1 3UJ, U.K.*)

The exocytic release of polymorphonuclear-leucocyte lysosomal enzymes may be initiated in several ways (Woodin & Wieneke, 1964; Weissmann *et al.*, 1971). We have used two of these to investigate the effect of cytochalasin B on exocytosis of lysosomal enzymes from glycogen-induced rabbit peritoneal-exudate polymorphonuclear leucocytes: (a) chemical perturbation of the plasma membrane by vitamin A alcohol; (b) lysosomal enzyme release normally accompanying the phagocytosis of [^3H]uridine-labelled *Escherichia coli*.

Low doses of vitamin A ($10\text{--}50\text{ }\mu\text{g/ml}$) selectively release lysosomal enzymes, whereas higher doses ($50\text{--}400\text{ }\mu\text{g/ml}$) also release non-lysosomal enzymes. In the presence of cytochalasin B ($10\text{ }\mu\text{g/ml}$) lysosomal

enzyme release is potentiated in the absence and in the presence of all vitamin A concentrations examined. In contrast, cytochalasin B does not potentiate the release of non-lysosomal enzymes (lactate dehydrogenase and leucine 2-naphthylamidase) induced by higher doses of vitamin A.

We and others have previously reported the inhibition of bacterial phagocytosis by cytochalasin B (Allison *et al.*, 1971; Davis *et al.*, 1971). We have now shown that cytochalasin B inhibits the phagocytosis of [^3H]uridine-labelled *E. coli* by polymorphonuclear leucocytes. Since phagocytosis was accompanied by a dose-related release of lysosomal enzymes, the latter might also be expected to be inhibited by cytochalasin B. The opposite result was obtained experimentally, however, cytochalasin B potentiating lysosomal enzyme release compared with controls both in the absence and in the presence of bacteria. This phenomenon was specific in that lactate dehydrogenase release was never elevated above a minimal value.

Cytochalasin B interferes with several biological processes (Turner & Carter, 1972); strong circumstantial evidence suggests that interference with contractile-microfilament function explains these

effects (Wessels *et al.*, 1971), although this has been questioned by others (Turner & Carter, 1972; Haslam, 1972). We propose a model in which cytochalasin B induces exocytosis by interference with the contractile microfilaments of polymorphonuclear leucocytes; filaments normally restrict lysosome contact with the plasma membrane, but, when filament function is disturbed by cytochalasin, contact of granules with membrane takes place, and this results in release of lysosomal enzymes.

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The Role of Ketone Bodies in the Metabolism of the Adult Desert Locust

By E. BAILEY and J. A. HORNE (*Department of Biochemistry, University of Sheffield, Sheffield S10 2TN, U.K.*) and M. E. G. IZATT and L. HILL (*Department of Zoology, University of Sheffield, Sheffield S10 2TN, U.K.*)

Bailey *et al.* (1971) demonstrated the occurrence of the ketone bodies acetoacetate and D-3-hydroxybutyrate in the blood of the adult pigeon (*Columba livia*) and haemolymph of the adult desert locust (*Schistocerca gregaria*). In the pigeon the ketone bodies probably play a similar role in metabolism to that in mammalian systems (Bailey & Horne, 1972). The present study was undertaken to try to elucidate the role of ketone bodies in the metabolism of the desert locust.

Acetoacetate and D-3-hydroxybutyrate are found in the haemolymph, fat-body and flight muscle of adult locusts. Acetoacetate is the major ketone body in haemolymph and flight muscle, whereas D-3-hydroxybutyrate often predominates in fat-body. The haemolymph concentration of acetoacetate varies with age (being highest at ages when the fat-body lipid reserves are being broken down), and is increased by flight, starvation and injection of corpus cardiacum extracts but is unaffected by stress or sex difference. Ketone bodies are formed in the fat-body although rates of formation of acetoacetate and D-3-hydroxybutyrate *in vitro* are low. However, the addition of haemolymph from starved insects to

incubation mixtures containing fat-body and potassium palmitate causes a marked stimulation of ketone-body formation. Both [3-¹⁴C]acetoacetate and D-3-hydroxy[3-¹⁴C]butyrate are oxidized by fat-body, flight muscle and testes of adult male locusts, [3-¹⁴C]acetoacetate being much more readily oxidized than D-3-hydroxy[3-¹⁴C]butyrate [only low activities of D-3-hydroxybutyrate dehydrogenase (EC 1.1.1.30) have been found in the tissues]. Flight muscle is tenfold more active than testis or fat-body in oxidizing [3-¹⁴C]acetoacetate. Starvation causes a greatly decreased rate of oxidation of ketone bodies by fat-body and flight muscle, but has no effect on the ability of testis to degrade those compounds. It appears that in the fed insects flight muscle is the major user of ketone bodies, whereas in the starved male locust testis is a major user of these compounds.

The results obtained suggest that ketone bodies are formed in fat-body during mobilization of triglyceride food reserves and are then either oxidized by the fat-body or transported (mainly as acetoacetate) in the haemolymph to the flight muscle and other tissues to serve as a respiratory fuel.

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Measurement of the Specific Radioactivity of Glutamine and Glutamate in Plasma

By J. COSTELLO, DEIRDRE M. KEANE and E. BOURKE (*Department of Clinical Medicine, Trinity College, Dublin 2, Irish Republic*)

Studies on the kinetics of glutamine and glutamate metabolism both *in vivo* and *in vitro* are frequently complicated by the fact that under many conditions simultaneous synthesis and utilization occur. Thus measurement of the specific radioactivity of glutamine and glutamate in plasma and other biological fluids is of value in studies of turnover rates and turnover times of these amino acids, and also for investigation of control factors in their synthesis and degradation. A method is described for the simultaneous measurement of specific radioactivity of both amino acids in plasma and other biological fluids.

Total glutamate is measured enzymically by the method of Bernt & Bergmeyer (1968) and total glutamine by a method based on that of Hoek *et al.* (1969).

For the radioassay of glutamate and glutamine the following procedure is used. First, total [¹⁴C]glutamate and [¹⁴C]glutamine are measured as follows: glutamine is converted into glutamate and ammonia by incubation for 1 h at pH 5.0 with *Escherichia coli*