to be no sodium pump. Instead, it appears that Na^+ efflux occurs by exchange for H^+ , which is then extruded by the proton pump. Na^+-H^+ antiport was originally reported by Mitchell & Moyle (1969) to occur in mitochondria.

What role do these gradients of pH and of electrical potential play in the energy-linked transport of nutrients? The case of K^+ is best understood. Proton-conductors, which collapse the gradients by allowing protons to flow back into the cells, block net K^+ uptake. Inhibitors of the adenosine triphosphatase likewise inhibit K^+ uptake, but reagents such as valinomycin have little effect. The inference, that K^+ uptake is dependent on the electrical potential generated by proton extrusion, is supported by the characteristics of mutants defective in K^+ uptake. One class of mutants is deficient in proton extrusion, another lacks the Na^+-H^+ antiport system. It seems clear that generation of an electrical potential is necessary for the accumulation of K^+ , though probably not sufficient.

The relationship of proton gradients to the uptake of other nutrients is less clear. Uptake of phosphate by *S. faecalis* is strictly dependent on glycolysis, and experiments with inhibitors suggest that, as in mitochondria (Palmieri *et al.*, 1969; Klingenberg, 1970), a gradient of pH is required for this process. Studies on amino acid uptake, still in a preliminary stage, point in the same direction. It now appears that substantial accumulation of threonine, glycine and related amino acids can be achieved by means of a pH gradient even in the absence of a source of metabolic energy. If this proves to be correct, it will lend strong support to the concept (Mitchell, 1970) that the circulation of protons between cytoplasm and medium provides the driving force for the active transport of many nutrients into bacterial cells.

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The Transport of Methyl α -Glucoside by *Pseudomonas aeruginosa*

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Glucose metabolism in *Pseudomonas aeruginosa* occurs via four pathways, namely the Entner & Doudoroff (1952) sequence, the pentose phosphate cycle, the phosphorylated oxidative route and the direct oxidative pathway leading to 2-oxogluconate. The enzymes of glucose catabolism are repressed by growth on organic acids such as succinate and citrate (Hamilton & Dawes, 1960; Lessie & Neidhardt, 1967). Experiments conducted in a chemostat, operated under conditions of nitrogen limitation, have demonstrated that citrate, or a metabolic derivative, inhibits both glucose utilization and the induction of the glucose-metabolizing enzymes by glucose when cells metabolizing citrate are presented with glucose as an alternative carbon source (Ng & Dawes, 1967; Hamlin *et al.*, 1967). Similar investigations on the competition between carbon sources for metabolism, for example the glucose-lactose diauxie of *Escherichia coli* (Loomis & Magasanik, 1967) and the succinate-glucose diauxie of *Arthrobacter crystallopoietes* (Krulwich & Ensign, 1969), have demonstrated that permeation can be a major control site. We have investigated the methyl α -glucoside transport system in *Ps. aeruginosa* in order that its role in the control of glucose utilization by organic acids may be assessed.

Uptake of ^{14}C -labelled methyl α -glucoside was measured by using a rapid membrane-filtration technique to separate the cells from the incubation medium, maintained at 37°C . The extracellular methyl α -glucoside on the filter was removed by washing. The extent of accumulation detected was critically dependent on the composition and temperature of the wash fluid employed. Washing with ice-cold phosphate buffer, supplemented with MgSO_4 , resulted in complete loss of accumulated material, whereas washing at temperatures in the range $18-22^\circ\text{C}$ resulted in a 60–80% loss. These effects could be overcome by the addition of NaCl to the basal wash fluid, the amount required increasing with decreasing temperature of the solution.

Incubation of glucose-grown cells with methyl α -glucoside results in an accumulation above the concentration in the medium. This accumulation is sensitive to the addition of NaN_3 , and extraction of the intracellular material revealed only unchanged methyl α -glucoside. Studies on the initial rate of uptake indicate that a component capable of saturation with methyl α -glucoside (K_m 2.5 mM) is involved, and exit studies with cells loaded with methyl α -glucoside indicate that exit is a first-order process (k 0.34) and is dependent on the concentration of

external methyl α -glucoside. Specificity studies, as measured in competition experiments, indicate that glucose, 2-deoxyglucose, 6-deoxyglucose and galactose are transported by the same system. Independent experiments, based on the fact that 2-deoxyglucose, 6-deoxyglucose and galactose are substrates for glucose dehydrogenase, confirm that these substrates are taken up by whole cells. These findings support the claim by Romano *et al.* (1970) that the phosphoenolpyruvate-dependent phosphotransferase system is not involved in glucose transport in this organism. Isotopic experiments with both 2-deoxyglucose and galactose have shown that these compounds are actively transported into glucose-grown cells. In both cases the major intracellular material was the unaltered substrate, although their hexonic acid derivatives could be detected extracellularly. Our results are contradictory to those of Eagon (1971), who has proposed that 2-deoxyglucose is not actively transported but enters by passive diffusion, the concentration gradient being maintained by the intracellular activity of glucose dehydrogenase. However, we have also shown that mutants defective in glucose dehydrogenase exhibit unimpaired 2-deoxyglucose transport. These discrepancies can, we believe, be explained on the basis of the cold-shock phenomena described by Farrell & Rose (1968), since previous investigations have not taken this into account.

Both the concentration and the activity of the methyl α -glucoside transport system have been shown to be under metabolic control. Succinate and gluconate inhibit the activity of the transport system, the nature of the inhibition suggesting that it is due to some metabolic product. Uptake is severely repressed in citrate- and succinate-grown cells.

These results will be further discussed.

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Transport of Non-Electrolytes in Yeast

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In spite of the fact that various yeast species have been exploited for thousands of years in generally beneficial fermentation processes and that some have recently proved their economic worth as producers of protein from waste water and even from petroleum, our exact knowledge of the ways they take up their carbon and nitrogen substrates is rather meagre. The greatest amount of information by far is available from baker's yeast, *Saccharomyces cerevisiae*, but some heuristically significant findings have been made in other species, like *Saccharomyces carlsbergensis* and *Rhodotorula glutinis*.

The uptake of materials by yeasts does not proceed to any significant extent by simple diffusion, there being in all the cases examined specific receptors residing in the outer membrane, the plasmalemma, which, either themselves or in concert with other membrane components, translocate the substrate into the cytoplasm. This is so for the uptake of both monosaccharides and amino acids, the two classes of non-electrolytes studied in some detail. However, other features of the transport process differ rather fundamentally, and they are dealt with separately below.

Monosaccharides are transported apparently by systems of rather broad specificity (Cirillo, 1961), although, even so, two or more may exist in a given species (Kotyk, 1967). The systems may be either constitutive or inducible by growth or incubation with a suitable inducer (Cirillo, 1968; Kotyk & Haškovec, 1968). Whereas in the genus *Saccharomyces* monosaccharides are taken up by a process of facilitated diffusion (although some phosphorylation during the membrane passage may take place in the case of D-glucose and other, closely related, sugars; van Steveninck, 1969), in other yeast species the process is a typical primary active transport, proceeding apparently in coupling with an exergonic metabolic reaction, but without interaction with an active transport, say, of ions (Kotyk & Höfer, 1965; Deák & Kotyk, 1968). All these uphill transports are distinguished from the 'permease' transport systems in bacteria by their extremely tight coupling with the energy-producing reaction. In fact, there is no reliable way of suppressing the 'uphill' part of the process without blocking transport altogether (Höfer & Kotyk, 1968). It may be envisaged that the movement itself of the carrier is metabolically linked. Another distinguishing feature, even with active transports, are the relatively high half-saturation constants (at least 10^{-4} M), implying relatively low affinities for substrate. This drawback is somewhat