The increase in isocitrate lyase activity also closely corresponded with the increase in O_2-uptake rate where this had been delayed by the presence of ammonium ion. Oxidation rate therefore is closely related to the increase in isocitrate lyase activity in unstarved, starved and ammonium-inhibited yeast.

From these results it appears that the glyoxylate cycle may be induced at the same time as the increase in O_2-uptake rate in baker's yeast adapting to acetate oxidation. Starvation depletes the endogenous stores of the yeast and so delays enzyme induction. In the presence of ammonium ion adaptation is delayed, possibly because of a stimulation of glycolytic activity with resulting elevated levels of phosphoenolpyruvate, a repressor of isocitrate lyase (Kornberg, 1965).

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Ultramicrochemical Studies on the Site of Formation of Dehydroepiandrosterone Sulphate in the Adrenal Cortex

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Biochemical studies have shown that cortisol and 11β-hydroxyandrostenedione are synthesized by the fascicular and reticular zones of the human adrenal cortex (Griffiths, Grant & Symington, 1963). Little is known, however, about the site of formation of dehydroepiandrosterone sulphate (DHAS). The work now reported shows that the reticular zone of the cortex is mainly responsible for the sulphation of dehydroepiandrosterone (DHA) both in the guinea pig and in the human.

Fresh adrenal tissue was frozen in solid carbon dioxide, transferred to a cryostat where cylinders of tissue were bored out and fresh frozen sections (2·5mm. diameter, 16μ thick) prepared (Grunbaum, Geary & Glick, 1956). One section was placed on a microscope slide for histological studies and the next three were placed in a reaction tube for enzyme assay. This sequence was followed through the entire cortex into the medulla. The tissue in the tubes was incubated with 0·1μc of [^{3}H]DHA (48mμg.) in 25μl. of a medium containing phosphate buffer, pH 7·0, ATP, $MgSO_4$ and K_2SO_4. After incubation, 100μl. of ethyl acetate containing 100μg. of DHAS was added, followed by 50μl. of 25% (w/v) NaCl. After mixing, the organic phase was removed and the aqueous phase was extracted twice more with ethyl acetate (100μl.). The residues from each extraction were chromatographed on silica gel in the solvent system 2-methylpropan-2-ol–ethyl acetate–aq. 5N-NH_4OH (41:50:20, by vol.) to obtain the sulphate fractions. Solvolysis of the sulphates yielded free extracts of steroid; these were acetylated and their specific radioactivities measured. Specific radioactivity measurements were also carried out on the free steroid obtained by hydrolysis of the acetates. The percentage conversion of DHA into DHAS in each incubation was then calculated from the mean value of the specific radioactivities.

The DHA-sulphating activity was based on the protein-nitrogen content of the tissue remaining in the reaction tube, which was determined by the bromsulphalein method of Nayyer & Glick (1954).

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The Absorption of Vitamin E in Man

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Low plasma vitamin E concentrations have been reported in patients with steatorrhoea (Losowsky & Leonard, 1967) and this has been confirmed in the present study. The cause of this deficiency is presumed to be diminished absorption of the vitamin, but little is known about the absorption in man. In the few reported studies, large unphysiological doses have been used and absorption indirectly assessed from plasma concentrations, which depend also on other factors. Rosenkrantz, Milhorat & Farber (1953) and Klatskin & Molander (1952) measured faecal quantities after large doses, but the methods used were non-specific.

In the present study fasting subjects were given oral doses of DL-α-[5-*methyl*-^{3}H]tocopherol in oily solution containing approximately 0·2mg. of total α-tocopherol. Radioactivity in urine, faeces, plasma and erythrocytes was followed for the next 14 days. A total of 39 subjects were studied, 20 without steatorrhoea (group A) and 19 with steatorrhoea (group B). Three deficient subjects in group B were also studied after repletion with

vitamin E. Twelve subjects were studied by chemical balance techniques when taking 200mg. of vitamin E acetate orally/day.

Urine radioactivity never exceeded 6% and will not be discussed further.

The overall absorption in group A varied from 61% to 90% (mean 75%) and in group B from 31% to 84% (mean 61%). The difference between the two groups was significant ($P < 0.001$). There was a significant correlation between the amount absorbed and the degree of steatorrhoea expressed as g. of fat/day ($P < 0.001$). Greater than 70% of the faecal activity was present as unchanged α-tocopherol and excreted over the first three days; thereafter less than 1%/day was excreted.

The plasma radioactivity curves were significantly different for the two groups but there was an overlap; the peak of radioactivity for both groups was at 8hr. Erythrocyte radioactivity curves paralleled the corresponding plasma curves but the amount of radioactivity was only approximately 10% of the plasma concentration.

Repletion with the vitamin did not alter its absorption.

Absorption is dose-dependent, as judged by comparison with therapeutic doses of vitamin E (200mg./day) in some of the same subjects.

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The Absorption and Distribution of Tocochromanols in the Rat

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The eight naturally occurring tocochromanols fall into two series, α-, β-, γ- and δ-tocopherol and the related compounds, α-, β-, γ- and δ-tocotrienol, differing only in having three double bonds in the isoprenoid side-chain. α-Tocopherol has been shown to have the greatest biological potency of the compounds so far studied; the order of potency is α-tocopherol > β-tocopherol > α-tocotrienol > γ-tocopherol > β-tocotrienol > δ-tocopherol (Bunyan, McHale, Green & Marcinkiewicz, 1961). The order of biological potency of the tocopherols is reflected in the tissue concentrations after oral dosing (e.g. Griffiths, 1959). The absorption and distribution of all the naturally occurring tocochromanols has been studied to compare tissue concentrations further with biological potency, more especially for α- and β-tocotrienol, and to study the differences in uptake brought about by the small changes in chemical structure.

Groups of female albino rats were given a dose of 12–20mg. of tocochromanol. The rats were killed 2, 4, 8, 16 or 24hr. after dosing, and the small intestine, liver, spleen, heart, kidney, skeletal muscle and brain were removed. Tocochromanols were isolated from the tissues and estimated by the method of Emmerie & Engel (1938).

Maximum concentrations of tocochromanol were found in the liver 4hr. after administration and the concentration declined continuously thereafter. The relative order of uptake in the liver, based on tissue concentrations 4hr. after dosing, was α-tocopherol (8·9% of the dose) > α-tocotrienol (7·6%) > β-tocopherol (6·4%) > γ-tocopherol (2·3%) > δ-tocopherol (1·6%) > β-tocotrienol (1·5%) > γ-tocotrienol (0·7%) > δ-tocotrienol (0·3%). Each tocopherol was absorbed more effectively than the corresponding tocotrienol. The relative order of uptake in both series of compounds was α-(5,7,8-trimethyl) > β-(5,8-dimethyl) > γ-(7,8-dimethyl) > δ-(8-methyl). The effect of three double bonds in the side chain was to decrease considerably the maximum concentration found in the liver and to increase the rate of decline from the maximum. This order of uptake was not the same in all tissues. In heart, muscle and kidney, the tocotrienols were found in amounts equal to or greater than those of the corresponding tocopherols. No tocotrienol was detected in the spleen after 8hr. after dosing; the tocopherols were detectable after 24hr. No tocochromanol was found in the brain over the 24hr. period. The pattern of maximum uptake shows α-tocotrienol to be absorbed more efficiently than β-tocopherol, although its biological potency is less, whereas β-tocotrienol is absorbed less efficiently than δ-tocopherol, although its biological potency is probably greater. The results demonstrate considerable variation in ratio between tissue concentrations at different times after dosing. For example, the ratio of α-tocopherol to α-tocotrienol in the liver was 100:85 4hr. after dosing, but 24hr. after dosing the ratio was 100:7, the tocopherol being more effectively retained. Suggested ratios for biological potency derived from tissue-storage tests therefore depend very much on the time-interval between final dosing and subsequent tissue analysis.

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