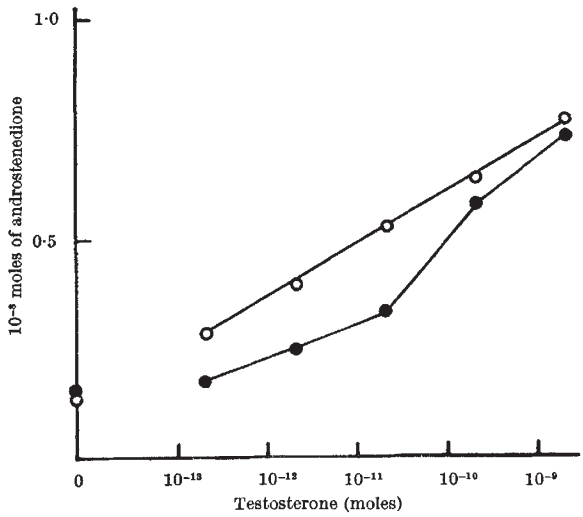

Fig. 1. Cycling reaction with testosterone. The steroid was dissolved in 50 μl. of propylene glycol with or without 5 μl. of methanol. Inhibition of methanol is shown. ○, No methanol; ●, methanol.

ably advantageous to decrease these concentrations further. Methanol in reaction (1) was also found to be inhibitory. The enzyme concentrations used in this reaction gave 94 per cent oxidation of $5{\cdot}8 \times 10^{-7}$ molar testosterone after 30 min of incubation at 38° C. A greater enzyme concentration, or incubation for up to 90 min, did not increase the percentage of oxidation.

After cycling for 60 min at 30° C the reaction was stopped and the steroids were extracted by the addition of dichloromethane. Testosterone and Δ^4-androstene-3,17-dione were then separated on purified Whatman No. 1 filter paper[4]. Aliquots were counted on planchets in a windowless gas flow counter and the oxidation of testosterone to Δ^4-androstene-3,17-dione was calculated from the relative conversion. Special precautions were necessary to keep all glassware and reagents free from contamination by nicotinamide nucleotides. Blank values corresponded to about 10^{-14} moles of testosterone. On a logarithmic scale a linear relationship was obtained between $3{\cdot}5 \times 10^{-13}$ and $3{\cdot}5 \times 10^{-9}$ moles of testosterone in the original sample (Fig. 1). This relationship is obtained because the substrate concentration in reaction (3) must be kept relatively low and decreases progressively during the course of the reaction. The cycling yield at the lowest initial concentration of testosterone was about 4,400, but it rapidly decreased when the initial testosterone concentrations were increased. The sensitivity obtained is from two to five times that reported for the electron capture detector, used in conjunction with gas chromatography after formation of the chloroacetate derivative of testosterone[5].

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Tyrosine Metabolism in Human Scurvy

TYROSINE is catabolized in the liver. One of the steps involved is the oxidation of *p*-hydroxyphenyl pyruvic acid to 2 : 5-dihydroxyphenyl pyruvic acid. The enzyme system which catalyses this reaction, *p*-hydroxyphenyl-pyruvic hydroxylase, is believed to require ascorbic acid as a cofactor. If the reaction is blocked, *p*-hydroxyphenyl pyruvic acid accumulates and is excreted in increased amounts in the urine. Other pathways for the removal of *p*-hydroxyphenyl pyruvic acid are utilized, for example, conversion to *p*-hydroxyphenyl acetic and *p*-hydroxyphenyl lactic acids, and these compounds can also be excreted in increased amounts in the urine. The excretion of excessive amounts of these substances in urine is known as tyrosyluria.

This condition occurs in premature infants fed on a diet which is deficient in ascorbic acid, but which contains large amounts of protein[1]. Tyrosyluria has been found in idiopathic steatorrhoea, in thyrotoxicosis, and in rheumatoid arthritis[2]. The condition was corrected in all the patients after they had been treated with large doses of ascorbic acid. It was therefore assumed that the tyrosyluria was caused by an inadequacy of dietary ascorbic acid. One of us (R. R.), however, has observed that tyrosyluria occurs in children with fibrocystic disease of the pancreas and that it is not corrected by saturation of the patients with ascorbic acid. This casts doubt on the view that lack of dietary ascorbic acid may inhibit liver *p*-hydroxyphenylpyruvic hydroxylase activity and so cause tyrosyluria. We have, therefore, investigated the excretion of tyrosine metabolites and tyrosine itself by patients with scurvy.

Twenty-four hour specimens of urine were collected from five adults and one 6 year old child who were suffering from frank scurvy. The clinical diagnosis in every case was subsequently confirmed by carrying out ascorbic acid saturation tests. The specimens were examined chromatographically for tyrosine metabolites by the method of Robinson *et al.*[3], and for tyrosine using the method of Chadwick *et al.*[4] None of the patients excreted excessive amounts of tyrosine and none showed signs of tyrosyluria.

Further urine specimens were examined after the patients had been saturated with ascorbic acid. There was no significant difference in the excretion of tyrosine or its metabolites before and after saturating the patients with ascorbic acid.

We conclude that a deficiency of dietary ascorbic acid does not cause tyrosyluria. Our results do not preclude the possibility that scorbutic patients may metabolize abnormally a loading dose of tyrosine.

Tyrosyluria may occur in patients with parenchymatous liver damage[5]. Boscott and Cooke's work was carried out on patients with conditions in which liver damage can occur and this could account, at least in part, for the tyrosyluria they observed[2].

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Effect of an Antihistamine on the Increased Vascular Permeability induced by Leucocyte Lysosome Fractions

IN previous publications[1–3] I reported the production of immediate increases in vascular permeability in rat tissues treated with a basic protein fraction extracted from lysosomes of exudate polymorphonuclear (PMN) leucocytes. It was suggested that the permeability effect was caused by the release of vasotropic amines from tissue mast cells