two antigenic determinants, both of the egg high-density-lipoprotein type, one of which is common to the very-low-density lipoprotein. In native high-density lipoprotein only the unique determinant is sufficiently exposed to react. On delipidation high-density lipoprotein also reacts with anti-(very-low-density lipoprotein). The difference between very-low-density lipoprotein and high-density lipoprotein is apparently due to the lipid moiety of the yolk high-density lipoprotein.

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An Alkaline Proteinase in Muscle Homogenates

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Homogenates of mouse or rat skeletal muscle in 50mM-potassium chloride degraded casein to perchloric-acid-soluble products at pH 8.3. The products were measured by extinction at 224nm, and at 260nm to correct for nucleic acid breakdown (which was relatively small). After centrifuging of the homogenates at 100000g for 30min nearly all of this proteolytic activity was usually found in the sediment fraction. The sedimented activity, when measured over the range pH 7.5–12.1, was maximum at pH 11; it was increased by 0.3M-sodium chloride, slightly at pH 10 but four- to five-fold at pH 7.5. The activity was lost when the fraction was heated at 80°C for 10min. Soya-bean trypsin inhibitor (1µg/0.3ml) produced a 77% inhibition of activity; there was no inhibition by hen-ovomucoid trypsin inhibitor (1µg) or by the supernatant (0.05ml) obtained by centrifuging a dialysed muscle homogenate at 100000g for 30min. Mouse serum (diluted 1:200; 0.05ml) caused a slight (10%) inhibition.

Attempts were made to solubilize the enzyme by extracting the fraction for 15min at room

temperature with various reagents followed by centrifuging at 30000g for 30min. Little or no enzyme was extracted with tricine buffer, pH 8.0 (30mM), acetate buffer, pH 5.0 (30mM), Triton X-100 (0.3%) or urea (8M)+2-mercaptoethanol (10mM). Sodium dodecyl sulphate (10mM) or guanidine hydrochloride (5M)+2-mercaptoethanol (10mM) completely inactivated the enzyme. The enzyme was extracted with 0.75M-sodium chloride or -potassium thiocyanate after removal of a large part of the inactive protein (mainly myofibril protein) with 8M-urea. The enzyme was slowly precipitated on standing in the cold and could be redissolved in 0.75M-potassium thiocyanate resulting in a tenfold purification (460-fold compared with the original homogenate). The extract hydrolysed *N*-benzyloxycarbonyl-L-tyrosine *p*-nitrophenyl ester but not *N*-toluene-*p*-sulphonyl-L-arginine methyl ester.

Huston & Krebs (1968) purified a Ca²⁺-dependent proteolytic enzyme from muscle extracts, with maximum activity at pH 7.5. Brush & Cheng (1968) purified an insulin-degrading protein from rat muscle, also with optimum activity at pH 7.5. Neither of these enzymes appears to be identical with the one studied here. The autolytic breakdown of a muscle myofibril fraction reported by Noguchi & Kandatsu (1969, 1970) also appears to be due to a different enzyme, since it is strongly inhibited by the supernatant fraction.

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The Effect of Chlorambucil on Nuclear Protein Synthesis in Yoshida Ascites-Sarcoma Cells

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Increases have been reported in the nucleic acids and protein content of a drug-sensitive strain of Yoshida ascites cells after treatment of the host animals with alkylating drugs; comparable changes have not been observed in a drug-resistant strain of the tumour (Harrap & Hill, 1969a,b). After the administration of a therapeuti-