Table 1. DHU-6-^{14}C and BUP-3-^{14}C incubated for 1 h at 30° C with aliquots of liver preparations representing 0.17—0.19 g of fresh tissue in a final volume of 3 ml of 0.25 M sucrose. Gas phase, air. Uracil-6-^{14}C was incubated at 37° C.

Substrate	Liver preparation	Amounts of radioactive compounds detected after incubation *				Radioactivity recovered after incubation ** (in per cent)
		Uracil	DHU	BUP	β-alanine	
8 μmoles BUP-3-^{14}C	Homogenate	0	0	77	23	
	Mitochondria + microsomes + supernatant	0	0	75	25	
	Microsomes + supernatant	0	0	75	25	
	Supernatant	0	0	77	23	96
8 μmoles DHU-6-^{14}C	Homogenate	0	72	6	22	
	Supernatant	0	73	10	17	102
1 μmole uracil-6-^{14}C	Supernatant	78	0	0	17	96

* Expressed in per cent of the total radioactivity on the paper strip after chromatography.
** Determined by plating TCA-treated aliquots of the incubation mixture.

Additional data from the work of Rutman *et al.*[7] indicate that the ureido carbon of uracil is released as carbon dioxide in the supernatant fluid from rat liver homogenates. The particulate fraction was found to inhibit the enzymatic activity. These results were confirmed by the recent work of Canellakis [5], and it was demonstrated that the enzymatic activity of rat liver slices approximated that of the supernatant fluid. On the basis of these findings it can be concluded that the enzyme catalyzing the reduction of uracil to DHU is present mainly or entirely in the part of the cytoplasm represented by the nonparticulate system.

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On the Molecular Weights of Human and Ox Pituitary Growth Hormones

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A growth-hormone-like principle in human pituitaries has recently been demonstrated by Gemzell and Heijkenskjöld[1]. The human pituitary growth hormone has now been isolated in an electrophoretically pure form [2], and investigations are being carried out concerning its chemical and physical nature. Some preliminary experiments on the sedimentation behaviour and molecular weight of this hormone are re-

Table 1. Sedimentation constants of human pituitary growth hormones under various conditions.

Buffer	pH	Protein mg/ml	S^0_{20}
Borate	10.0	7	1.99
»	9.9	8	2.12
»	9.9	15	2.30
»	9.9	15	2.20
Acetate	4.1	9	1.6
Phthalate	4.0	5	1.3

ported here and comparison is made with the ox hormone.

Experimental. The sedimentation coefficients were determined in a Spinco ultracentrifuge at 59 780 r.p.m. The molecular weight determinations were carried out in the same centrifuge at lower speeds as will be described elsewhere by Ehrenberg [3].

The human growth hormone was prepared by Heijkenskjöld [2] and the ox hormone was a gift from Dr. C. H. Li [4] to Dr. C. A. Gemzell, who kindly put it at our disposal.

At pH 9.9 a 0.028 M borate buffer was used, and at pH 4 either 0.125 M acetate or 0.045 phthalate. In all cases 1 % sodium chloride was added.

Results and discussion. The sedimentation coefficients from the experiments on the human growth hormone are listed in Table 1. The hormone protein appeared to be homogeneous in the experiments at pH 9.9, whilst the presence of small amounts of both faster and slower components is likely at pH 4. A single experiment on the ox hormone at pH 9.9 gave a sedimentation coefficient of 3.51 S in agreement with previous data [5,6]. As is seen the values for the human hormone are appreciably lower at this pH. The same differences persist in more acid buffers where the ox hormone still shows values above 3 S[6]. At the rather high pH 11.5, however, the sedimentation coefficients of ox hormone [6] are comparable to those of human hormone at pH 4 and 10. These data suggest that the human hormone has a lower molecular weight than the ox hormone at pH 4 and 10.

The molecular weights were determined on both hormones at pH 9.9. Two runs were made on the human hormone (10 and 5 mg/ml), both giving a molecular weight close to 30 000 in the first part of the experiment. In these calculations use was made of the value 0.76 of the partial specific volume of the ox hormone [7]. A decrease in the calculated molecular weight was observed in the later part of the experiment. This behaviour can be explained either by an inhomogeneous sample or by a concentration dependent equilibrium between molecules of different sizes. One experiment with ox hormone, also at pH 9.9 gave in the early part a molecular weight of about 75 000 and in the later part a decrease similar to that just described. Li and Pedersen [6] observed at this H an increase of S^0_{20} with protein concentration, which was explained by the assumption that the ox hormone protein behaves like a mixture of monomeric and dimeric forms. A similar concentration dependence of S^0_{20} of human hormone is indicated by the data of Table 1. There are thus good reasons to assume that this hormone protein also behaves like a mixture of molecules of different sizes at pH around 10.

The molecular weight of the ox hormone at pH 10 calculated by Li and Pedersen [6] from separate sedimentation and diffusion experiments was about 50 000; the sedimentation coefficient was extrapolated to zero protein concentration and the diffusion coefficient was determined at the low concentration of 2.5 mg/ml. This value is likely to correspond to the monomeric ox hormone, and our value of 75 000 to a sample of more or less aggregated molecules. In the later part of the experiment the observed molecular weight decreased towards the value for the monomeric form. By analogy herewith the value of the molecular weight of the human growth hormone of about 30 000 at pH 10 would be a mean for molecules of different sizes. The smallest hormone molecules present at this pH might have molecular weights as low as 15 000 – 20 000.

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