this way was 12320 ± 170 (S.E.M.). Each measurement requires only 0.4 ml of solution; equilibrium is reached within 20 min.

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Quaternary Structure of Helical Proteins with Globular Subunits

By E. J. MILNER-WHITE and P. G. SPAIN (*Department of Biochemistry and Department of Mathematics, University of Glasgow, Glasgow G12 8QQ, U.K.*)

The theory proposed (Milner-White, 1971, 1972) for studying the possible range of structures of oligomers assuming equivalence of the protomers also applies to helical proteins with globular protomers [e.g. actin filaments, bacterial pili or some rod-shaped virus coats; some enzymes (Olsen *et al.*, 1970) form enzymically active helical polymers]. For a summary of helix symmetries consult Klug *et al.* (1958).

Such proteins may be divided into two classes (heterologous-type and isologous-type). In one there is only heterologous binding between the protomers forming a helix. In this case the opposite ends of a single helix are different; such helices have polarity. In the other class each protomer has a minimum of two isologous site-pairs. The ends of such a helix are identical because of the dyad axes perpendicular to the helical axis; these helices lack polarity. (Sometimes two or more heterologous-type helices may lie antiparallel; in this case the protein will lack polarity).

In order to describe heterologous-type helices in terms of their binding-site structure it is necessary to define the angle ϕ (see Milner-White, 1972). ϕ is the angle between the planes of θ of two adjacent protomers measured in the plane perpendicular to the line joining their 'centres'. ϕ is defined as the clockwise rotation of the plane of θ of one protomer relative to that of the other protomer (as seen from the first protomer).

Parameters of the helix used are: $2\pi/\alpha$, the number of protomers per turn; r , the radius; h , the axial distance per protomer. ψ is another helix parameter. d is the distance between the centres of two adjacent protomers. These are related to ϕ and θ by the equations below (they apply for one heterologous site-pair; others may also be present). This approach will, we believe, be useful for the study of quaternary structure rather as the work of Ramachandran & Sasiekhara (1968) is useful for studying secondary structure.

$$h = d \cdot \sin \psi$$

$$d \cdot \cos \psi = 2r \cdot \sin \left(\frac{\alpha}{2} \right)$$

$$\sin \left(\frac{\alpha}{2} \right) = \cos \left(\frac{\theta}{2} \right) \cdot \sec \psi$$

$$\tan \psi = \tan \left(\frac{\theta}{2} \right) \cdot \sin \left(\frac{\phi}{2} \right)$$

Isologous-type helices with the same dimensions may be built if all the distances d are equal and if $\zeta_A = \zeta_B$. For such helices ϵ and $(\pi + 2\zeta)$ should be substituted for θ and ϕ in the above equations. With different geometry isologous binding may also generate more complex helicoid structures.

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Differences in the Metabolism of Glucose between Normal and Dystrophic Human Muscle

By DAVID A. ELLIS and JENNIFER M. STRICKLAND (*Midland Centre for Neurosurgery and Neurology, Holly Lane, Smethwick, Warley, Worcs., U.K.*)

Fibre-bundle preparations of muscle biopsies were incubated with [^{14}C]glucose. HClO_4 extracts were made and chromatographed on paper, and the radioactivities of radioactive spots were counted. Metabolites were identified by co-chromatography with authentic substances; fructose was identified as its 1-methylphenylhydrazine (Neuberg & Mandl, 1946).

Fructose contained significantly more label in muscle extracts from patients affected with Duchenne-type dystrophy than in those from normal subjects. Radioactivity of fructose, expressed as a percentage of total incorporated ^{14}C , was $14.4 \pm 10.1\%$ (9) for normal subjects, compared with $43.2 \pm 14.7\%$ (8) for dystrophic subjects ($P < 0.001$ in the t test). The same parameter in other control groups had the following values: neurogenic muscle disease, $11.0 \pm 11.0\%$ (3); adult muscular dystrophy including myotonic dystrophy, $15.9 \pm 18.1\%$ (8); polymyositis, $25.0 \pm 21.8\%$ (6); normal foetal muscle (12–18 weeks' gestation), $21.2 \pm 17.8\%$ (6); female carriers of Duchenne-type dystrophy, $48.7 \pm 21.2\%$ (7). The value in Duchenne dystrophy is significantly raised compared with those in neurogenic muscle disease ($0.001 < P < 0.01$), foetal muscle ($0.02 < P < 0.05$) and adult muscular dystrophy ($0.001 < P < 0.01$). The value

for female carriers of the disease is also significantly raised compared with those in normal muscle ($P < 0.001$), neurogenic disease ($0.01 < P < 0.02$), foetal muscle ($0.02 < P < 0.05$) and adult muscular dystrophy ($0.001 < P < 0.01$).

D-Glucitol was identified as a probable intermediate in the conversion of glucose into fructose by the appearance of [^{14}C]glucose and [^{14}C]fructose as the major labelled components in extracts of muscle fibre-bundles incubated with [^{14}C]glucitol. Further evidence for this route of formation came from measurement of the activities of polyol-NADP oxidoreductase (EC 1.1.1.21) and L-iditol-NAD oxidoreductase (EC 1.1.1.14). In muscle extracts polyol oxidoreductase activity (μmol of substrate transformed/min per g fresh wt. at 25°C) was 2.17 ± 2.13 (14) for Duchenne-type dystrophy, compared with 0.17 ± 0.53 (13) for normal subjects ($0.001 < P < 0.01$) and 0.71 ± 1.45 (14) for a group of subjects with either polymyositis or neurogenic muscle disease ($0.02 < P < 0.05$). L-Iditol oxidoreductase activity (same units) for the same three groups was 1.64 ± 2.14 (15), 0.45 ± 0.91 (14) and 0.54 ± 0.81 (14) respectively. The differences between the 'dystrophy' group and the other two are significant at the level $0.05 < P < 0.1$.

These results indicate the presence in human muscle of a subsidiary pathway of glucose metabolism, whose importance is increased in Duchenne-type muscular dystrophy and carriers of the disease.

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Activation of Liver Glycogen Synthetase *in vitro*

By H. M. PATEL and BRENDA E. RYMAN (*Department of Biochemistry, Royal Free Hospital School of Medicine, 8 Hunter Street, London WC1N 1BP, U.K.*)

Glycogen synthetase (EC 2.4.1.11) exists in a non-phosphorylated (*a*) form and a phosphorylated (*b*) form. In a fresh liver extract glycogen synthetase is predominantly in the *b* form, and it is activated to the *a* form on incubation at 20°C (Gold & Segal, 1967; Merseman & Segal, 1967). Activation is preceded by a lag period, which Hers and co-workers (Stalmans *et al.*, 1971) suggest represents the time taken for phosphorylase *a* activity to diminish and thus remove the inhibition it normally exerts on synthetase *b* phosphatase (the enzyme involved in the conversion synthetase *b* $\rightarrow$ synthetase *a*). The coupling of activation of synthetase (synthetase *b* $\rightarrow$ synthetase *a*) with deactivation of phosphorylase (phosphorylase *a* $\rightarrow$

phosphorylase *b*) is believed to be a regulatory mechanism of glycogen metabolism (Stalmans *et al.*, 1971; DeWulf *et al.*, 1970).

Synthetase activation is stimulated by several inorganic salts (Hers *et al.*, 1969) that do not influence phosphorylase *a* phosphatase (Stalmans *et al.*, 1970) (catalysing the conversion phosphorylase *a* $\rightarrow$ phosphorylase *b*). We have confirmed that the activation synthetase *b* $\rightarrow$ synthetase *a* in a liver preparation under the conditions reported by DeWulf *et al.* (1970) occurs after a well-marked lag period in the presence of 5 mM-sulphate. In the presence of chlorides (5 mM), however, this lag is not evident. Added phosphorylase *a* increases the lag period in the presence of sulphate (Stalmans *et al.*, 1971), but when MgCl_2 (5 mM) is used to replace sulphate little inhibition is observed, and in the presence of both salts the lag period is further prolonged. The glucose and AMP effect on synthetase activation (DeWulf *et al.*, 1970) is not observed in the presence of 5 mM- MgCl_2 . Further evidence suggests that the lag period is not exclusively regulated by phosphorylase *a* concentration but is partly a sulphate-dependent effect.

The immediate activation of synthetase by 5 mM- MgCl_2 is modified by addition of ATP (3 mM) and the lag period is reinstated. The duration of the lag period is dependent on the $\text{Mg}^{2+}/\text{ATP}$ ratio but independent of phosphorylase *a* concentration, which could be manipulated by the presence of 5 mM-caffeine. Activation of synthetase beings as soon as ATP is hydrolysed, and EDTA inhibits at this point. The observed effects of ATP and Mg^{2+} on the activation are not explicable in terms of their effects on synthetase *a*.

These results suggest that ATP concentrations may contribute to the regulation of synthetase phosphatase activity and hence synthetase activation by controlling the availability of Mg^{2+} . The possibility that changes in the conformational state of the substrate (i.e. synthetase *b*) may be involved in these studies *in vitro* rather than direct effects on the enzyme (i.e. synthetase phosphatase) is worth considering.

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