Intramolecular Rotation and the Structure of High Polymers. I. The Structure of Polypeptide Chain.

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I. Potential Curve of Internal Rotation. The results of experiments of Raman effect, infra-red absorption, dipole moment and electron diffraction made for ethylene dihalides by Mizushima, Morino, Watanabe, Simanouti, and others[1] in our laboratory have shown that there are three potential minima in one complete rotation about a carbon single bond as axis (see Fig. 1).

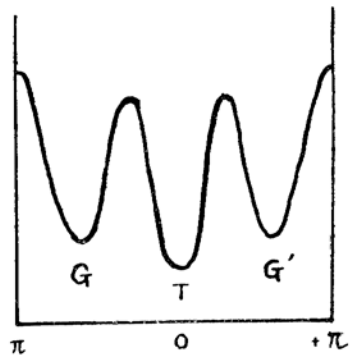

Fig. 1. Potential curve of ethylene dihalide.

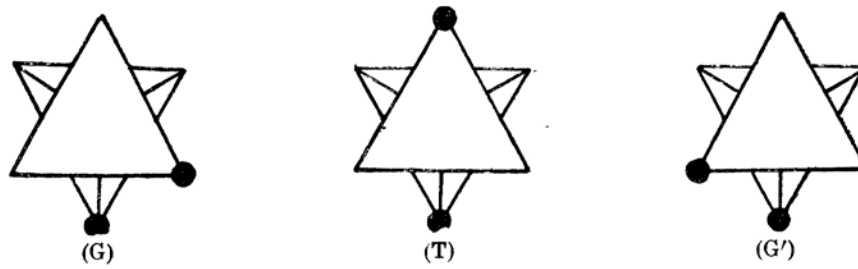

Fig. 2. Molecular forms corresponding to the three potential minima.

These three potential minima correspond to one trans form (T) in which two halogen atoms are at the farthest distance apart and two gauche forms (G and G′) obtainable from the trans form by internal rotation

(1) Mizushima, Morino, and others: *Physik. Z.*, 35 (1934), 905; 38 (1937), 459; *J. Chem. Phys.*, 9 (1941), 826; Mizushima, Morino, Watanabe, Simanouti, and others; *Sci. Pap. I.P.C.R.* (Tokyo), 39 (1942), 396, 401; 40 (1942), 87, 100, 417, 425; 42 (1944), *Chem.*, 1, 5, 27, 51.

of ±120° (see Fig. 2). The energy difference between the trans and the gauche minima amounts to 1 kcal/mol (1.2 kcal for ethylene dichloride and 1.3 kcal for ethylene dibromide). The height of potential barrier lying between these two kinds of minima amounts to about 10 kcal/mol, which is much smaller than the activation energy of ordinary chemical reactions so that "rotational isomers" of ethylene dihalides cannot be isolated under ordinary conditions. The experimental results obtained in our laboratory for ethylene chlorhydrin, chloral hydrate, normal paraffins,[2] and cyclohexane can also be explained by considering intramolecular potential of similar type.

II. The Basic Structure of Polypeptide Chain. As a stable configuration of polypeptide chain an extended form (Fig. 3) has already been proposed[3] which corresponds to the structure of silk fibroin or of β-keratin. However, from our experimental results stated in I we can propose another stable chain configuration (bent form) shown in Fig. 4. These two forms do not differ much from each other in internal energy and the bent form may be the more stable of the two because of the existence of intramolecular hydrogen bond (denoted by the dotted lines in Fig. 4).

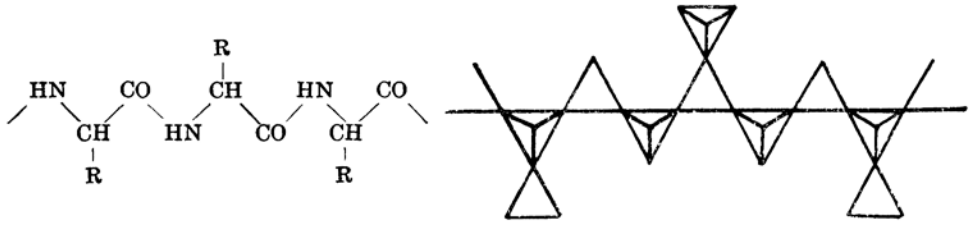

Fig. 3. Extended form.

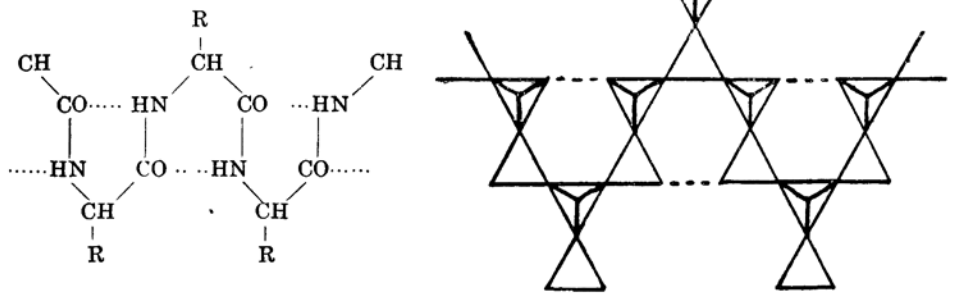

Fig. 4. Bent from.

Other stable forms are obtained by the suitable combination of these two basic structurs. Let us denote the unit structures of the extended

(2) Mizushima and Simanouti: *Proc. Imp. Acad. Tokyo*, 20 (1944), 86. The other results will be published before long.

(3) Meyer-Mark: *Hochpolymere Chemie* II, (1940).

and the bent forms by E and B respectively and hence represent the chain form of Fig. 3 by EEE ---- and that of Fig. 4 by BBB -----. Other conceivable structures are then denoted by EBEB ----, EEBEEB ----, EEEBEEEB ----, etc., among which the ring structures $(EB)_3$, $(EEEB)_3$, $(EBBB)_3$, etc. characterized by trigonal symmetry are included. Let us try to explain the experimental data obtained for proteins by these structures.

1) From the bond radii the period along the chain is calculated as 5.1 A for BBB ---- and as 10.2 A for BBEBBE -----. The experiment of Astbury made for keratin can be explained by these predicted values.[4] The mechanical properties of keratin fiber can also be accounted for by these structures as shown in next section.

2) The X-ray investigations show that the crystal structure of insulin,[5] excelsin,[6] etc. has trigonal symmetry. This is readily understood, if we consider that the molecule of such proteins is made of the said ring such as $(EEE ---- B)_3$, or by the suitable superposition of these plane forms. (The superposition may be caused through the hydrogen bond or the covalency of side chain).

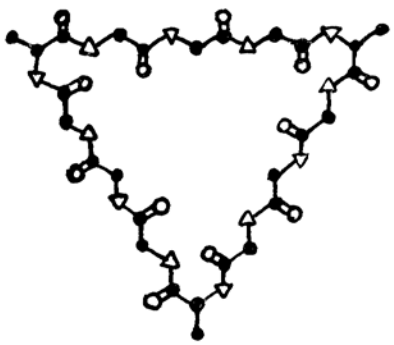

Fig. 5. The trigonal structure $(EEEB)_3$.

3) Whether an amino acid residue in the polypepetide chain takes E-form or B-form depends upon the nature of this residue. In the case of glycine residue which has no side chain E-form can be considered to be the more stable. This is compatible with the fact that silk fibroin molecule containing considerable number of glycine residues has a structure of β-keratin type or EEEEE -----.

In the case of a residue with the side chain which exerts large steric effect to the main chain or which strengthen the intramolecular hydrogen bond (denoted by the dotted line of Fig. 4), B-form becomes the more stable.

4) In any case these two forms E and B do not differ much in

(4) Astbury and Street: *Phil. Trans. Roy. Soc.* (London), A 230 (1931), 75.

(5) Crowfoot: *Proc. Roy. Soc.* (London), A 164 (1938), 580.

(6) Astbury, Dickinson, and Baily: *Biochem. J.*, 24 (1935), 2351.

their energy and the polypeptide chain tends to change its configuration by a slight change in the external condition. We can thus understand why some proteins are denatured easily and also why some proteins such as antibodies show quite specific properties.

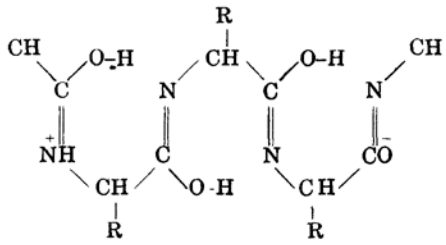

Fig. 6.

5) For BBB ---- structure a resonance form shown in Fig. 6 can be considered. This means that in some case the polypeptide chain may form an extended oscillator just as a molecular chain with conjugated double bond and thus we may explain by a field of forces resulting from this oscillator the mechanism of the combination of dyes with proteins, etc.

III. The Structure of α-keratin. No satisfactory explanation for the structure of α-keratin has hitherto been advanced. However, we consider that BBB ---- or BBEBBE ---- can represent this structure by which we can explain reasonably the experimental data obtained for α-keratin as follows:

1) The period along the fiber axis observed for α-keratin amounts to 5.15 A, which is in good agreement with that predicted from the structure BBB ----. The side chain spacing is found to be 9.8 A in β-keratin as well as in α-keratin. This is comprehensible, if we consider that cystine bond remains unchanged in both keratins (see Fig. 7 and 8).

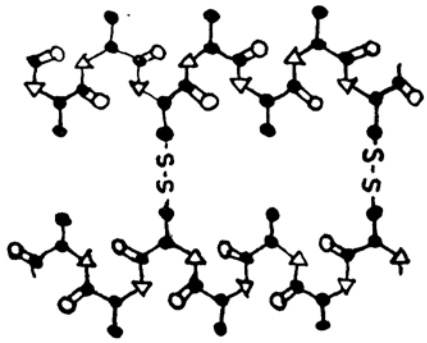

Fig. 7. Cystine bond in α-keratin.

The backbone spacing 4.65 A of β-keratin is not found in α-keratin. This will be due to the disconnection of intermolecular hydrogen bond of β-keratin which in turn forms the intramolecular hydrogen bond of

α-keratin denoted by the dotted lines of Fig. 4.

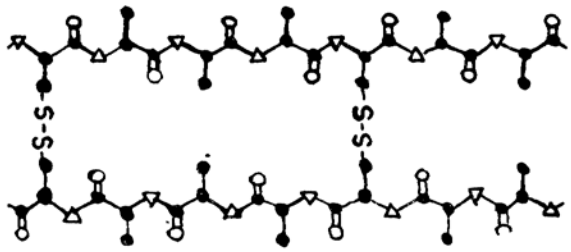

Fig. 8. Cystine bond in β-keratin.

The supercontraction of wool may also be explained, if, for example, we consider a structure such as shown in Fig. 9, which can be obtained by the disconnection of cystine chain.

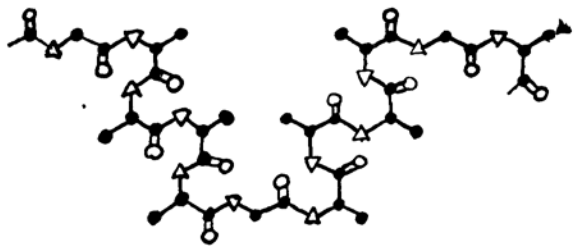

Fig. 9. An example of the structure of supercontracted wool.

2) Let us next discuss the mechanical properties of wool. For an elongation Δl within 2% Hooke's law is found to hold. We have, therefore,

$$E = \frac{1}{2}k(\Delta l)^2 \tag{1}$$

where E is the change in energy and k the force constant. Let y be Young's modulus refered to a single molecule and l_0 be an equilibrium length of an amino acid residue along the chain. Then y is defined as:

$$y = \frac{\partial E}{\partial \Delta l} \Big/ \frac{\Delta l}{l_0} \tag{2}$$

From Eq. (1) and (2) we have

$$y = kl_0 \tag{3}$$

The value of Young's modulus Y in its ordinary sense can be obtained from the experimental relation[7] between the tension K and the elongation $\Delta L/L_0$:

(7) Astbury and Coworkers: *Phil. Trans. Roy. Soc.* (London), A 230 (1931), 75; 232 (1933), 333; *Trans. Farad. Soc.*, 29 (1933), 193; *Proc. Roy. Soc.* (London), A 150 (1935), 533.

$$Y = K \Big/ \frac{\Delta L}{L_0} = \frac{10 \times 10^5}{0.02} = 5 \times 10^7 \text{ g/cm}^2 \qquad (4)$$

We can now put

$$\frac{\Delta l}{l_0} = \frac{\Delta L}{L_0}$$

$$y = YA ,$$

where A denotes the molecular cross section. This can be calculated from the observed side chain spacing 9.8 A of α-keratin and the backbone spacing which is assumed reasonably to be 6 A. We have then

$$k = \frac{y}{l_0} = \frac{YA}{l_0} = \frac{(5 \times 10^7 \times 980)(9.8 \times 6 \times 10^{-16})}{2.57 \times 10^{-8}} = 1.1 \times 10^4 \text{ dyne/cm} \qquad (5)$$

This value of k is found quite reasonable when compared with the force constant of hydrogen bond.[8)]

Let us next discuss larger elongation. The X-ray diagram shows that in this case the structure of α-keratin changes into that of β-keratin. Let E_2 be the energy of E-form refered to B-form, l_1 and l_2 be the length of an amino acid residue in B- and E-forms, and n_1 and n_2 be the number of residues in B- and E-forms, respectively, we have for the length L and energy E of a polypeptide chain

$$L = n_1 l_1 + n_2 l_2 = n l_1 + n_2 (l_2 - l_1) \qquad (6)$$

$$E = n_2 E_2 \qquad (7)$$

where $n = n_1 + n_2$

In the case of the coexistence of both forms the tension per unit chain K is calculated as

$$KA = \frac{\partial E}{\partial L} = \frac{\partial E}{\partial n_2} \frac{\partial n_2}{\partial L} = \frac{E_2}{l_2 - l_1} \qquad (8)$$

From this relation we see that all B-forms change into E-forms for a certain value of tension, at which the tension-elongation curve becomes parallel to the elongation axis. That such is not actually the case is due to the neglection of entropy in the foregoing discussion. The value of entropy will be very small in both extreme cases where all amino acid residues take B- or E-form, but in the intermediate case its value will be considerable, so that the said curve inclines to the elongation axis to some extent. If we put the value of tension 6×10^5 g/cm^2 (corresponding to the middle point of the slope of tension-elongation curve observed in

(8) Halford: *J. Chem. Phys.*, 14 (1946), 395.

the measurement for the relative humidity of 100%) into K of Eq. (8) and put

$$l_2 - l_1 = 3.32 - \frac{5.14}{2} = 0.75\,\text{A}\,,$$

we can calculate the energy difference between B- and E-forms as:

$$E_2 = 370\ \text{cal/mol}$$

The structure of α-keratin may also be considered as BBEBBE ----[9] (see Fig. 10), for which the period along the main chain is calculated as 10.3 A. This is also compatible with the experiment.[10]

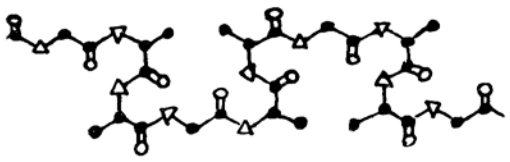

Fig. 10. α-keratin.

In this case the side chain (cystine bond) is directed upwards or downwards from the plane of Fig. 10, and therefore, the corresponding spacing (9.8 A) remains constant, when the chain is stretched out. If, therefore, we assume the backbone spacing as 9 A, we can calculate the force constant k and the energy difference E_2 just as in Eq. (5) and (8):

$$k = \frac{(5\times10^7\times980)\,(9.8\times9\times10^{-16})}{5.15\times10^{-8}} = 0.84\times10^4\ \text{dyne/cm},$$

$$E_2 = KA(l_2 - l_1) = 1800\ \text{cal/mol (residue)}.$$

The elongation of wool will not be explained by a single mechanism, as the experiment of Bull[11] shows. (The electron microscope experiment[12] shows that keratin fiber has specific fine structure, and, therefore, it may consist of complex micelles.) Hence the foregoing discussion will not cover all the elongation steps. However, since X-ray diagram shows the structural change (α to β change) after elongation, it will be difficult to explain the mechanical property of keratin fiber without taking into account the intramolecular rotation as stated above.

(9) This can be considered as a structure in the state of supercontraction, if the structure of keratin fiber in its ordinary state is represented by the configuration shown in Fig. 4.

(10) The intensity relation of X-ray scattering may more reasonably be explained by the structure of Fig. 10 than that shown in Fig 4. We are indebted to Dr. N. Tanaka for the discussion on this structure.

(11) Bull: *J. Am. Chem. Soc.*, **66** (1944), 1253; **67** (1945), 533.

(12) Mercer: *Nature*, **159** (1947), 535. See also Bear: *J. Am. Chem. Soc.*, **65** (1943)' 1784; **66** (1944), 2043.

The discussion of globular proteins based on the structure proposed by us will be given in a later communication.

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