The inhomogeneous stretching process of collagen

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1. Introduction

In an earlier paper (Nemetschek and Hosemann, 1973) a kink model was discussed, which can explain the observed splitting of the small angle reflections of native collagen fibers under stress. The protofibrils consist of paracrystalline regions of at least 300 Å lateral size and a length H in the directions of the fiber, which is somewhat smaller than 135 Å. These domains have a rhomboedric structure with lattice constants \( a = 13.4 \text{ Å} \) and \( b = 12.8 \text{ Å} \). The netplanes 100 build up grates of protofibrils. The protofibrils of each grate are hold together by a kind of "backbone-linkage". Under stress one can observe, that these grates are tilted by 3.8° along the (010)-direction. After 135 Å they are shifted by nearly \( \frac{3}{4} \) of the diameter of a protofibril in the (100)-direction

\[
135 \cdot \sin 3.8° = 9.2 \approx \frac{3}{4} \cdot 12.8 \text{ Å} = 9.6 \text{ Å}.
\]  

All 135 Å so-called "amorphous" layers orthogonal to the fiber axis exist. In our previous paper it was discussed that the protein chains here are looser packed and dislocations, chain-foldings and special smaller side chains are concentrated, which give rise to the cross stripes in the electronmicrograph (Nemetschek, 1967, 1971). A comparison with the kink model of Pechhold and Blasenbrey (1970) was made (Nemetschek and Hosemann, 1973) and shortly mentioned, that the structure in this "amorphous" region has some similarity with structures, discussed for melt crystallized polyethylene. In this paper another explanation for the kinked protofibrils is given, which is derived from the experience with synthetic polymers with elastomeric properties.

2. Comparison with synthetic elastomers

Segmented elastomers crosslinked by secondary valences consist of less disordered "hard" domains alternating with "soft" segments. As an example in polyurethane the stretching takes place exclusively in the soft segments. With increasing elongation the snarled chain molecules have become more and more elongated, whilst the molecules in the hard segments are well parallel adjusted as at the beginning. Similar conditions exist in partially crystalline homopolymer thermoplasts. Here the chains in the hard segments are bonded physically by van der Waals forces and now are not absolutely stiff. In both cases the expansion under stress is inhomogeneous. The soft segments are much more expanded than the hard ones. As shown below, similar conditions are discussed in native collagen too.

If the molecules are stretched in a different way in the hard and soft segments, the packing density changes in a different way too, and the problem is how the densities of the two segments fit together. Bonart (1964) pointed out that in stretched synthetic polymers anomalous orientations of the hard segments occur under strain. Examples are given in polyethylene terephthalate (Daubeney et al., 1954), in aromatic polyamide (Vogelsong, 1962) and in elastomers of urethane (Turner-Jones et al., 1962). Taking into account that the 135 Å periodicity in collagen in our earlier paper was discussed as a domain of higher disorder, there exist obviously similarities between the tertiary structure of these elastomers and the native collagen fiber. On the other hand remarkable differences between these biological and synthetic structures make it neces-
sary to find out a new type of kinked structure. Contrary to elastomers for instance the 670 Å period does not change within the error of experiment if native collagen is stretched. Randall (1954) found an increase of the long period for dried and stretched collagen. In dried collagen the subfibrils certainly are collapsed totally and the tertiary structure is damaged so much that there we meet with quite other properties of the structure which shall not be discussed here.

The question if in native collagen the tilting of paracrystallites exists still before stretching or if it is created only by the stretching process remains open. Some experimental data are given in the next section.

3. Materials and techniques

The fibers used in this study and the X-ray techniques previously described (Nemetschek and Hosemann, 1973).

4. Microdensitometric traces of unstretched and stretched native collagen

In figs. 1 and 2 microdensitometric traces of both families of reflections are given; i.e. the splitting occurs both for a part of the equatorial reflections lying on the a-axis and for the meridional reflections. In table 1 data are collected of the distances $D$ between the splitted centres and their distance $B$ from the zero reflection, all measured in mm on the film (diffraction diagram). Specimen-to-film distance was 201.5 mm in a Kiessig camera with CuKα radiation.

<table>
<thead>
<tr>
<th>Equator</th>
<th>Meridian</th>
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<tbody>
<tr>
<td>$B$</td>
<td>$D$</td>
</tr>
<tr>
<td>9</td>
<td>1.2</td>
</tr>
<tr>
<td>18</td>
<td>2.3</td>
</tr>
<tr>
<td>24</td>
<td>3.2</td>
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<td>23</td>
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The data of this table indicate that the model of the triple helix must be corrected at least in the stretched state. It is not the aim of this paper to discuss this in detail.

The data of table 1 prove that the splitting never can be explained according to Miller and Parry (1973) by layer lines of a superhelicoidal structure (supercoiled microfibrils) but only by tilting of the paracrystallites around the b-axis by 3.8°. A superhelix would produce splittings $D$, which are not proportional to $B$ and $\varepsilon$ therefore is not a constant. From the data of figs. 1 and 2 moreover we learn that by stretching the half width $D$ and $W$ of the reflections in the splitting direction become smaller than the half width of the unstretched samples marked by blackprints. This means that before stretching the fibers statistically are crooked and bent. Hereby the splitting, if existing before stretching, is covered.