The role of topological constraints in the kinetics of collapse of macromolecules

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Résumé. — Une chaîne polymérique formée de N segments s'effondre à la suite d'un abaissement brutal de température. On montre que la cinétique de cet effondrement est un processus en deux étapes lorsque N est beaucoup plus grand que la longueur caractéristique de reptation N_e. La première étape dure un temps $\sim N^2$ et amène à un globule de structure froissée, ou fractale. Dans cet état, toute partie de la chaîne forme elle-même un globule de ce type ; ces parties sont isolées les unes des autres parce que la chaîne ne forme pas un réseau fantôme. Les contours de la chaîne dans le globule froissé forment une ligne fractale dont la dimension fractale est 3, égale à la dimension d'espace. La seconde étape accroît la densité des globules par pénétration des extrémités de la chaîne à travers les globules fractals ; elle est réalisée par reptation, dure un temps $\sim N^3$, et produit des nœuds dans le parcours de la chaîne.

Abstract. — It is shown that the kinetics of collapse of a polymer coil consisting of N segments after an abrupt decrease of temperature is a two-stage process if $N \gg N_e$. The first stage takes a time $\sim N^2$ and leads to the peculiar state — crumpled, or fractal, globule. Any part of a chain of any scale is itself a globule in this state ; these parts are segregated from each other in space due to the non-phantomness of a chain. The chain fold in the crumpled globule is a fractal line with fractal dimension 3, equal to the space dimension. The second stage is a chain knotting ; it is realized by means of reptation-like mechanism of motion, takes a time $\sim N^3$ and is accompanied by an increase of globule density.

Introduction.

Kinetics of collapse of a polymer coil after the abrupt decrease of temperature or solvent quality was investigated in [1-3]. According to the theory proposed by de Gennes [1] the process of collapse leads to the formation of crumples of successively growing scale along the chain (Fig. 1). Just this process was observed independently in numeric experiments [2, 3]. In fact, the chain remains linear after the formation of crumples on a minimal scale, and a « new » chain appears to be shorter and thicker (Fig. 1b) ; this « new » chain forms crumples again, it leads to its further shortening and thickening (Fig. 1c) and so on until the spherical globule is formed (Fig. 1d). The time of this process was estimated in [1] and the result obtained in [1] is :

$$t_{\text{collapse}}^{(0)} \simeq \eta a^3 \frac{\Delta T}{\theta^2} N^2 \qquad (0.1)$$

where N is the number of chain links, θ is the θ-temperature which is considered to be the initial temperature before the temperature jump, $\Delta T = \theta - T$; ($T < \theta$) is the temperature jump, a is the size of a link, and η is the viscosity of the solvent.

The topological constraints due to the non-phantomness of the chain were not considered in [1]. It was argued in [1] that Gaussian coil of N links has of order $N^{1/2}$ contacts between the chain segments, but topological entanglements correspond to a small fraction of these contacts of order $N^{1/2}/N_e$, where N_e is the well-known parameter of the reptation model [4, 5]. It follows from the condition $N_e \gg 1$ that for the realistic chain lengths $N^{1/2}/N_e < 1$. Therefore the topological constraints are not essential for a coil.

But in reality, during the collapse, the number of intrachain contacts grows rapidly ; it is of order N in the globule ; hence, the arguments cited above
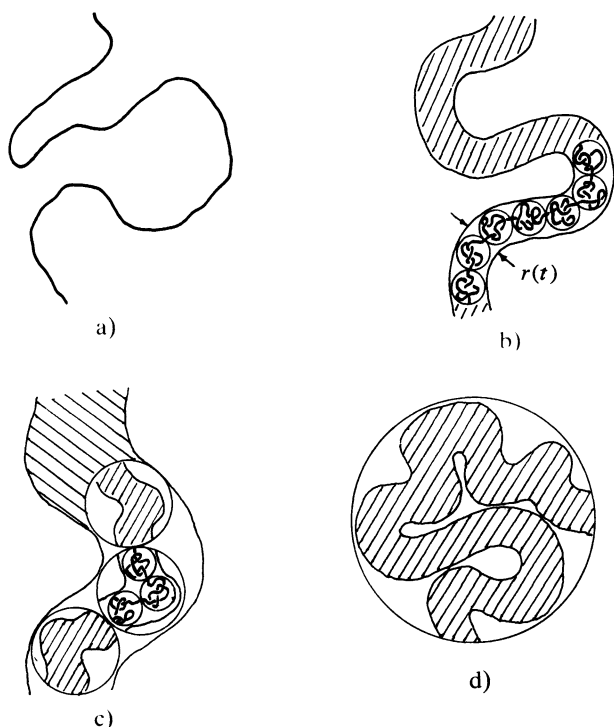
Fig. 1. — The sequential stages of collapse. (a) Initial state ; (b), (c) Intermediate stages ; (d) Final state — crumpled globule.

become inapplicable in the case $N \gg N_e$ and the topological constraints become essential. (The case $N < N_e$ will be briefly discussed in the next section.)

We shall argue in this work that crumples of any scale larger than N_e are impenetrable for each other due to the topological constraints. As a result, the self-similar process of crumpling shown in figure 1 leads to the long-living peculiar state which can be called « crumpled globule ». The striking feature of this state is that the chain parts of any scale larger than N_e are segregated from each other in space. This crumpled globule is fractal, i.e. the line presenting the chain trajectory on the scales corresponding to $N > N_e$ is fractal with fractal dimension $d_f = 3$, equal to the space dimension.

The subsequent relaxation of the crumpled globule to the equilibrium is realized by means of reptation of chain and consists of formation of quasiknots on a chain. This process can be defined as topological relaxation.

1. Fractal properties of the crumpled globule.

Let us consider the process of fast collapse of a polymer chain. The regime of fast collapse is realized when the temperature jump $\Delta T = \theta - T$ ($\Delta T > 0$) is large enough to form a globule, i.e. $\Delta T N^{1/2} \gg \theta$ and it is fast enough to take a time $t_0 \ll t_c$ where $t_c \simeq \eta \dfrac{a^3}{T} N^z$ is the largest relaxation time of the

chain and z is the dynamical critical index for a coil ($z \doteq 2 \div 3$, see for example [5]). This means that in this regime of collapse the chain ends do not take part in the process of collapse during the temperature jump. Therefore each finite part of a chain can be considered as a part of an effectively infinite (or closed) molecule without ends. Hence the topological state of any finite part of a chain becomes well defined : the whole chain is effectively infinite for these time scales. Since the chains ends have not enough time to take part in the collapse, the topological state of the chain becomes frozen on this stage of the process.

The process of collapse is shown in figure 1. At an arbitrary time moment t, the chain is presented as the chain of blobs — globules of the size $r(t)$ (Fig. 1b) ; these globules can be considered as new « monomers » for the subsequent stage. These « monomers » come into contact with each other and stick together.

If blobs coming into contact are the usual globulized linear chains, they could mix (without the density and volume interactions changes but with the entropy advantage) due to the interpenetrations of their free ends. Such a situation is realized in a melt of linear chains. But in our case blobs, being the internal parts of a very long chain, do not have free ends. Therefore the topological state of such blobs is fixed during the time scale under consideration. Let us now show that this circumstance leads to the mutual segregation of blobs (in contrast to the blobs formed from the linear chains with free ends). To explain this segregation the following analogy is very helpful : since the topological state of a certain blob is fixed it can be presented as a polymer ring without knots ([1]).

Due to the non-phantomness of a macromolecule, other parts of the chain (or other blobs) form the effective lattice of obstacles for this ring (Fig. 2b). However it was shown in [6] that the polymer ring without the volume interactions consisting of M links and not entangled with the obstacles of the lattice belongs to the same universality class as the randomly branched polymer chain and has the size

$$R^{(0)}(M) \simeq a M^{1/4} . \tag{1.1}$$

([1]) Generally speaking the topological state of a blob can differ from the trivial knot but it is equivalent to the initial topological state of a chain before the collapse starts. It makes sense to suggest here that any finite part of the chain in θ-conditions (before the collapse) do not contain knots. This suggestion is justified by the results of numeric experiments [7, 9] as well as empiric equation [8] which estimate the probability of knot formation in the course of chain cyclisation in θ-conditions. Thus it is natural to suppose that the initial topological state of the chain is trivial (without knots) on any finite scale.
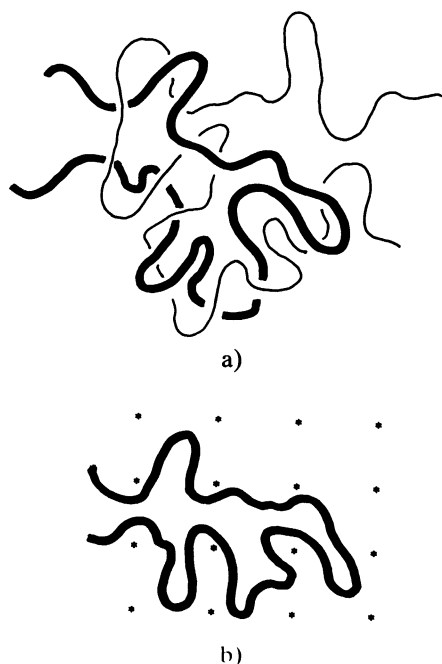
a)

b)

Fig. 2. — (a) The interpenetration of two different parts of a chain in the crumpled globule ; (b) The chain plays role of an array of obstacles for its part denoted by thick line.

Since $R^{(0)}$ is the equilibrium size of a ring chain in the lattice of obstacles, the entropy of this ring plotted as a function of the chain size R reaches its maximum when $R \simeq R^{(0)}$. At the same time the excluded volume effects lead to the relation

$$R(M) \gtrsim aM^{1/3} . \qquad (1.2)$$

Because of the inequality $R(M) > R^{(0)}(M)$ for large enough M further swelling of the chain will lead to further decrease of entropy. This means that the penetration of loops stretched from one blob into another one becomes entropically unfavourable and the segregation of blobs occurs.

For short enough chains the topological segregation of blobs does not take place because of the small number of topological constraints. Let us determine the crossover scale on which the segregation of blobs begins.

First of all we define the average number of chain monomers g between two neighbouring contacts of one part of the chain with another ones. The scale g depends on the globule density n. For a compact globule (which is similar to the melt) $g \simeq 1$; but if the density of the globule, n, is small $(na^3 \ll 1)$ then the chain part of g links in such a globule is Gaussian-like and its size can be estimated as $R_g \simeq ag^{1/2}$. Thus we obtain :

$$\frac{g}{R_g^3} \simeq n ; \quad \text{i.e.} \quad g \simeq \frac{1}{(na^3)^2} . \qquad (1.3)$$

We can represent the globule (as well as the polymer melt or a semidilute solution) as a densely packed system of g-link blobs [11, 12]. The average number of contacts for each blob is of order of unity but only a small fraction of order $\frac{1}{N_e}$ of the total number of contacts leads to the topological constraints. Therefore we can conclude that the chain part of $g^* \simeq gN_e$ links leads to one entanglement.

On the scales (along the chain) less than g^* the topological constraints are negligible and the chain parts of lengths less than g^* form the usual equilibrium globules.

Let us introduce B and C-the two- and three-body interaction constants respectively. As usual, slightly below θ-point we have

$$B \simeq -6 \frac{\Delta T}{\theta} < 0 ; \quad C = \text{const} > 0 . \qquad (1.4)$$

The density of the equilibrium globule is determined as follows [12]

$$n \simeq n_{eq} \simeq -\frac{B}{C} . \qquad (1.5)$$

The volume of the globule of g^* links is of order $\frac{g^*}{n_{eq}}$ and its size is $R_{g^*} \simeq \left(\frac{g^*}{n_{eq}} \right)^{1/3} \simeq N_e^{1/3} \left(\frac{g}{n_{eq}} \right)^{1/3}$. Substituting the estimate (1.3) into the latter formula we obtain the relation which is valid slightly below the θ-point

$$R_{g^*} \simeq N_e^{1/3}/na^2 . \qquad (1.6)$$

For the kinetics of collapse of macromolecules the topological constraints are negligible up to the scales on which the thickness of effective chain $r(t)$ (see Fig. 2b) is less than R_{g^*}. Therefore if the chain length is less than g^*, i.e. if the inequality

$$N < g^* \simeq N_e g \qquad (1.7)$$

is valid then the topological constraints do not play a role neither in the kinetics of collapse of macromolecule nor in the formation of globule structure. In this case the results of de Gennes [1] can be applied without any change.

Let us now consider long enough chains, i.e.

$$N > g^* \simeq N_e g . \qquad (1.8)$$

We define the « topological blob », as the equilibrium globule of g^* links (having the size R_{g^*}). Actually when the process of collapse reaches scales greater than g^* each g^*-link part of the chain produces approximately one entanglement, i.e. each topological blob fills one cell of the effective lattice of obstacles (Fig. 3).

If we now consider the subchain of s topological blobs (i.e. the chain consisting of sg^* monomers)
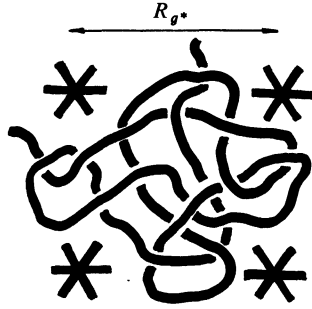
Fig. 3. — Topological blob consisting of $g^* = N_e\, g$ monomers in an effective lattice of obstacles with the size of the cell of order $R_{g^*} \simeq \left(\dfrac{g^*}{n_{eq}} \right)^{1/3}$.

then we can apply the arguments connected with equations (1.1) and (1.2). Actually, because the density of the system cannot exceed n the size of the chain of sg^* monomers is in any case larger than

$$R_{sg^*} \simeq \left(\frac{sg^*}{n_{eq}} \right)^{1/3} \simeq s^{1/3} R_{g^*}. \qquad (1.9)$$

The swelling of such subchain due to the interpenetration of loops in the effective lattice of obstacles is entropically unfavourable if $R_{sg^*} > s^{1/4} R_{g^*}$, i.e. for any $s > 1$.

Thus the chain part of length sg^* (for any $s > 1$) will not mix with other chain parts and will be segregated in space from other such blobs.

All these conjectures hold true if we consider two neighbouring (along the chain) topological blobs.

A part of the chain cannot be transferred from one blob to another because this will lead to density fluctuations [2].

So we come to the conclusion that during the collapse all chain parts on scales larger than g^* remain segregated and preserve their structure which was formed previously. As a result the process of collapse of long enough chains ($N \gg g^*$) leads to the folding of the chain into the self-similar structure which can be represented as a scale invariant system of mutually segregated for any scale (larger than g^*) crumples.

It is interesting to note that the trajectory of chain on scales $g^* < \tilde{g} < N$ is analogous to Peano curve [10] which fills the square and has crumples on any scale. The fractal (Hausdorff) dimension d_f of this line can be estimated as follows. Every part of the chain having $m > g^*$ segments is a globule itself since it became compact after the process of crump-

(2) If one of the blobs contains a knot in the initial state (before the collapse) then they may mix making this knot common, i.e. enlarging the scale of this knot. But as was mentioned above it is natural to consider the initial state of the chain to be trivial.

ling of neighbouring parts of the chain of scale of order m is completed. The size of this part is $R_m = \left(\dfrac{m}{n} \right)^{1/3}$. The fractal dimension of a line is determined via the relation $(R_m)^{d_f} = m$; hence :

$$d_f = 3 . \qquad (1.10)$$

This result for the crumpled globule should be compared with analogous estimates for equilibrium globules. It is well known (see, for example, [11, 12]) that from the local point of view (i.e. for scales less than the size of the whole equilibrium globule), the equilibrium globule is analogous to the polymer melt and the Flory theorem is valid. It means that the chain statistics of that globule is Gaussian up to scales equal to the size of the whole globule. Thus the structures of the crumpled globule and the equilibrium globule differ drastically for scales $g > g^*$.

Now we can define the scale g^* as the crossover scale where the fractal dimension changes :

$$\begin{cases} d_f = 2 & g < g^* \\ d_f = 3 & g > g^* . \end{cases} \qquad (1.11)$$

2. The density of the crumpled globule.

The time scales at which the crumpled globule state exists is restricted by the time during which the topological structure (trivial in our case) remains without changes. On these time scales the chain ends have not enough time to take part in the collapse. Therefore the crumpled globule formed from the linear chain is a *partially equilibrium system* with a fixed topology. (This state would be equilibrium for the ring or infinite polymer chain where the free ends are absent).

The density of the crumpled globule n and its volume $V \simeq \dfrac{N}{n}$ can be obtained from the minimisation of the free energy of a globule. The total free energy $F = F_{int} - TS$ includes the volume interaction term F_{int} as well as the entropy loss S due to the formation of crumples on any scales greater than g^*.

The free energy of volume interactions slightly below the θ-point ($na^3 \ll 1$) can be written in the usual form :

$$F_{int} = V\,(Bn^2 + Cn^3) . \qquad (2.1)$$

The entropy loss due to the formation of crumples can be roughly estimated as follows

$$S \simeq - \frac{N}{g^*} \qquad (2.2)$$

($\sim$ one entropy unit per crumple of minimal scale g^*).

Substituting equation (1.3) into equation (2.2) and minimising the free energy of the system under the condition $nV = N$ we obtain the following estimation for the globule density n.

$$n \simeq -\frac{B}{C}\frac{1}{1+\frac{1}{N_e}\frac{a^6}{C}} . \qquad (2.3)$$

This result must be compared with equation (1.5) which determines the density of the equilibrium large globule [12].

It can be easily seen that

$$n < n_{eq} . \qquad (2.4)$$

It should be noted that according to [13] the ratio $\frac{C}{a^6}$ (which determines in some sense the stiffness of the chain) for all experimentally available chains lies in an interval $25 < \frac{a^6}{C} < 500$ and thus the difference in densities of the crumpled globule and the equilibrium one is essential especially for stiff enough chains.

Now we can make a correction to the estimation of the time of collapse which was made in [1]. This correction is due to the fact that the process of fast collapse leads to the fractal crumpled globule state instead of equilibrium one. Thus

$$t_{collapse}(N) \simeq t_{collapse}^{(0)}(N) \cdot \frac{C}{C+\frac{a^6}{N_e}} \qquad (2.5)$$

(we use here the estimation of $t_{collapse}^{(0)}$ in the form

$$t_{collapse}^{(0)}(N) \simeq \eta a^6 \frac{|B|N^2}{C\theta} \simeq \eta a^3 \frac{\Delta T}{\theta^2}\frac{6a^3}{C}N^2 .$$

In the work [1] and in equation (0.1) the relation $\frac{6a^3}{C} \simeq 1$ was implied).

3. Topological relaxation of the crumpled globule.

In the case $N \gg N_e$ (more exactly in the case $N \gg g*$) the subsequent relaxation of the crumpled globule to the equilibrium one is realized due to the penetration of the ends of the chain through the fractal crumples. Such a mechanism of motion resembles the reptation-like motion of the chain consisting of « topological blobs » which takes place in an effective lattice of obstacles or in a « tube » formed by the chain itself.

The time of topological relaxation can be estimated as the reptation time of the chain of blobs on

its total length $\frac{N}{g*}$. The relaxation time of one topological blob can be estimated as $t_{collapse}^{(0)}(g*)$ and for a chain of $\frac{N}{g*}$ segments in the lattice of obstacles (one entanglement per blob) the relaxation time is of order $\left(\frac{N}{g*}\right)^3$. Therefore we obtain the total topological relaxation time t_{top} :

$$t_{top}(N) \simeq t_{collapse}^{(0)}(g*)\left(\frac{N}{g*}\right)^3 ; \quad (N \gg g*). \qquad (3.1)$$

It should be stressed that $t_{top} \sim N^3$ while $t_{collapse} \sim N^2$. This fact makes meaningful the consideration of collapse and topological relaxation as two processes separate in time (in case $N \gg g*$). (It is easy to verify that if $N \sim g*$, then $t_{top} \sim t_{collapse}^{(0)}$, which means that in this case there is no reason to consider the kinetics of coil-globule transition as a two-stage process).

Conclusion.

Predictions concerning the process of collapse of long enough chains ($N \gg N_e$) as well as properties of transient structures can be formulated now :

1) the kinetics of the coil-globule transition which takes place after abrupt decrease of temperature in the case $N \gg N_e$ must include two stages : at first the crumpled globule is formed, then the topological relaxation to the equilibrium state occurs ;

2) the density of the crumpled globule (especially — for stiff enough chains) is less then the density of the equilibrium globule ;

3) the fractal properties of the line describing the fold of a chain in the crumpled globule are not trivial and differ drastically from those of an equilibrium globule ;

4) the collapse of a ring polymer without knots is a one-stage process ; in the case $N \gg N_e$ the final structure is the crumpled globule. (This point will be discussed in detail in a forthcoming article).

The justification of these predictions in real or numeric experiments seems to be, from our point of view, an important step to estimate the adequacy of the concept of a crumpled globule suggested in this article.

It should be stressed that the concept of crumpled globule may be especially useful in the investigation of the structure of biopolymers (in particular DNA and proteins) where the hierarchy of structural levels is a well-known fact (e.g. nucleosome structure of chromatin and secondary, supersecondary and tertiary structures in proteins [14]).

References

[1] DE GENNES, P. G., *J. Phys. Lett.* **46** (1985) L-639.

[2] BIRSTEIN, T. M., GRIDNEV, V. N., SKVORTSOV, A. M., *Mol. Biol. (USSR)* **15** (1981) 394.

[3] BIRSTEIN T. M., GRIDNEV V. N., SKVORTSOV A. M., *Vysocomolek. Soed. (USSR)* **23A** (1981) 297.

[4] DOI, M., EDWARDS, S. F., *J. Chem. Soc. Faraday II* **74** (1978) 1789, 1802, 1818.

[5] DE GENNES, P. G., Scaling Concepts in Polymer Physics, (Cornell Univ. Press, Ithaca, N.Y.) 1979.

[6] KHOKHLOV, A. R., NECHAEV, S. K., *Phys. Lett.* **112A** (1985) 156 ;
NECHAEV, S. K., SEMENOV, A. N., KOLEVA, M. K., *Physica* **140A** (1987) 506.

[7] FRANK-KAMENETSKII, M. D., VOLOGODSKII, A. V., *Usp. Fiz. Nauk (USSR)* **134** (1981) 641.

[8] LE BRET, M., *Biopolymers* **19** (1980) 619.

[9] VOLOGODSKII, A. V., Private communication.

[10] MANDELBROT, B., Fractals : Form, Chance, Dimension, (W. H. Freeman & Co. S.F.) 1977.

[11] LIFSHITZ, I. M., GROSBERG, A. Yu., KHOKHLOV, A. R., *Rev. Mod. Phys.* **50** (1978) 683.

[12] GROSBERG, A. Yu., KHOKHLOV, A. R., *Sov. Scient. Rev. Phys. Rev.,* I. M. Khalatnikov Ed., (Harword Academic Publishers) **8** (1987) 147.

[13] GROSBERG, A. Yu., ZHESTKOV, A. V., KUZNETSOV, D. V., *Vysokomolek. Soed. (USSR)* **28A** (1986) 1397.

[14] KREIGHTON, T. E., Proteins : Their Structure and Molecular Properties (W. H. Freeman & Co. S.F.) 1984.