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A model of Engineering Materials Inspired by Biological Tissues

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Abstract

The perfect ability of living tissues to control and adapt their mechanical properties to varying external conditions may be an inspiration for designing engineering materials. An interesting example is the smooth muscle tissue since this “material” is able to change its global mechanical properties considerably by a subtle mechanism within individual muscle cells. Multi-scale continuum models may be useful in designing essentially simpler engineering materials having similar properties. As an illustration we present the model of an incompressible material whose microscopic structure is formed by flexible, soft but incompressible balls connected mutually by linear springs. This simple model, however, shows a nontrivial nonlinear behavior caused by the incompressibility of balls and is very sensitive on some microscopic parameters. It may elucidate the way by which “small” changes in biopolymer networks within individual muscular cells may control the stiffness of the biological tissue, which outlines a way of designing similar engineering materials. The ‘balls and springs’ material presents also prestress-induced stiffening and allows elucidating a contribution of extracellular fluids into the tissue’s viscous properties.

Keywords: living tissues, smooth muscles, prestressed cytoskeleton, tissues viscoelasticity, control of stiffness

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1. Introduction

Living organisms are far more complex than the most complicated engineering structures. Namely they provide many different functions such as metabolism, growth, remodeling, reproduction as well as control, sensing and communication allowing them to react to a wide spectrum of changing physical conditions. The fact that the structure of living tissues has been “designed” during hundred millions of years of evolution indicates that it must be optimized in a way that cannot be expected in engineering structures being evolved for hundreds or even tens of years. May we learn some “tricks” that has been used by Nature in construction of such effective structures? The problem is that the detailer study of the living tissues is done, the more complicated details and connections are found out. The picture becomes very complex and some features that may be important for our engineering inspiration remain unnoticed. Here, the role of oversimplified physical and mechanical models may be very important. Namely, they may discover some “right” views that neglect many complications and details and highlight only certain features that may be an inspiration how designing novel engineering solutions.

In this contribution we look for an inspiration in the structure of smooth muscle tissues that have remarkable mechanical properties. Their ability to adapt to the whole scale of external conditions by rapid changes of their mechanical properties is enormous. They are able to undergo extremely large deformations without damaging their structure and return back. Special mechanisms in the cytoskeleton (a protein fiber network that spans living cells) are able

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to change mechanical properties of this tissue in several orders. There is no doubt about the usefulness of materials with such properties in the industry. How are they designed?

From a strictly chemical viewpoint, the basic structure of living tissues is formed from complex conglomerates of various polymer fibers saturated by several kinds of liquid solutions [1]. Mechanical properties of this kind of condensed matter are given both by mechanical behavior of arrangements of elaborately linked and entanglement fibers as well as the presence of liquids. Generally, such structures are soft and practically *incompressible*. The fact differentiating living tissues from other kinds of such soft matter structures is their *special arrangement* in the form of living cells (at sizes of tens or hundreds micrometers) delimited by thin ($\sim$ 7 nm) *membranes* that are highly sophisticated structure of phospholipid and protein molecules – see Fig. 1. There are many reasons why living tissues are designed in such a manner. The cell structure indeed allows organisms to perform many tasks very effectively (e.g. metabolism or reproduction). We notice, above all, how such arrangement may be important in explaining special mechanical properties of smooth muscle tissues.

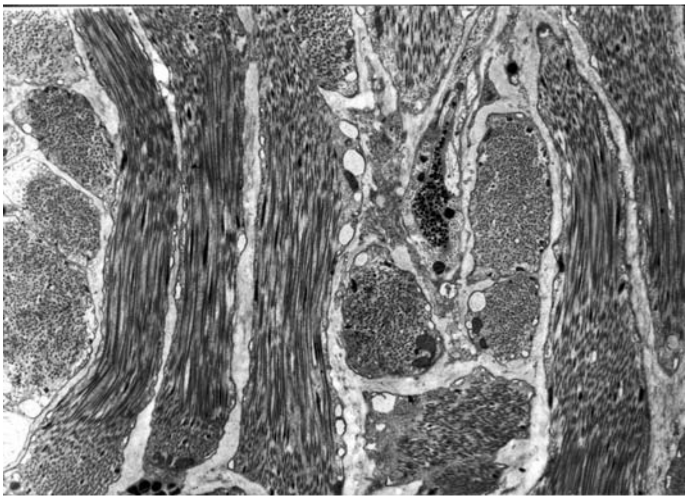

Fig. 1. Section through the smooth muscle cells of a gastropod (scale 5 μm), [8]

At macroscopic scales, living tissues may be understood as viscoelastic materials. Their viscoelasticity is, however, determined by other mechanisms than that of pure polymeric materials. Again the special structure of living tissues plays the important role. Namely, the polymeric viscosity is supplemented by effects connected with the flow of extracellular fluids around individual cells. We study this effect in our simplified model. The results may be also interesting when designing special engineering materials with a similar structure.

The paper is organized as follows. In the second paragraph we present a simple model material, motivated by basic features of a tissue structure, consisting of incompressible but flexible balls connected mutually by linear linkages. We show briefly some interesting properties of such a structure that is highly nonlinear, especially when the stiffness of balls is essentially smaller than that of their links. The third paragraph is devoted to the situation when the balls are prestressed. We show how the effect of prestress-induced stiffening, that occurs in living tissues, may be easily explained by this simple model material. The fourth paragraph deals with involving the viscoelasticity of the structure caused by movement of the fluid around balls during its deformations. Paper finishes with concluding remarks in the fifth paragraph.

2. Balls and springs

Let us imagine an incompressible structure consisting of identical flexible (very soft) balls filled with a fluid and joined mutually by elastic linkages. Because the balls are very soft, the deformation of the structure is realized predominantly by changes of their shapes so that the extracellular springs deform only a little. As a result, the material is very soft too as evidenced by a simple estimation of the stiffness

$$k_{est} \approx \left(k_s^{-1} + k_b^{-1}\right)^{-1} , \tag{1}$$

where k_s, k_b is the linkage and the ball stiffness respectively, see also Fig. 2. If k_b is very small in comparison with k_s, the structure stiffness k_{est} is near to k_b, i.e. $k_{est} \sim k_b$. It means that if balls become perfectly soft, i.e. $k_b \sim 0$, the structure is infinitely soft too. It, however, cannot happen if there exist some constraints on the balls' deformation preventing cases where none of all extracellular springs is deformed. Such a constraint adds some correction to the formula (1) and the structure becomes stiffer.

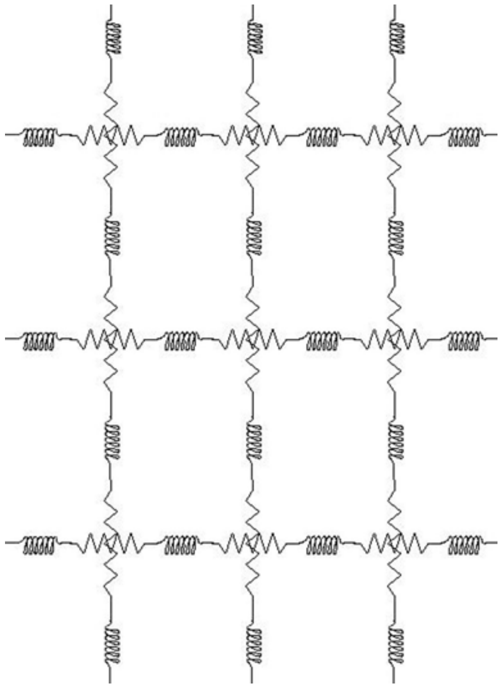

Fig. 2. The idealized 'balls and springs' material (without constraint on the balls) behaves as an arrangement of springs (with no coupling between the different space directions)

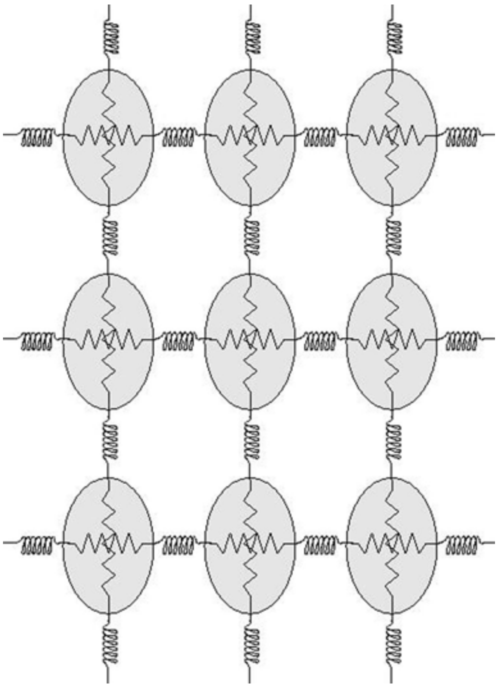

Fig. 3. The studied 'balls and springs' material includes a constraint on the balls' volume

A typical constraint of this kind is realized when the balls keep their volume to be *constant*, i.e. the balls are incompressible. Then, the formula (1) ceases to be valid if k_b is small regarding to k_s. Some nonlinear "structural effects" appear that correct the effective stiffness of the material. To estimate them, we define the simplest form of the *'balls and springs' material* (a continuum limit of the structure presented in Fig. 3) [2]: the structure is regular, the springs connecting the balls are linear and the balls are reinforced by some linear springs that define the balls stiffness. The strain-energy function of this material, W_{BS}, consists of two parts,

$$W_{BS} = W_{spring} + W' , \tag{2}$$

where W_{spring} is the strain-energy function of a structure in which the balls are fully compressible (i.e. the arrangement of linear springs shown in Fig. 2 which stiffness is expressed by (1)). The part W' corresponds to the structural effect caused by the incompressibility of balls. It is a nonlinear function that has to be determined numerically [2]. If the stiffness of balls is comparable with the stiffness of linkages or larger, the term W' is negligible. If the balls are essentially softer, the term W' is dominant and defines the mechanical properties of the structure (see Fig. 4).

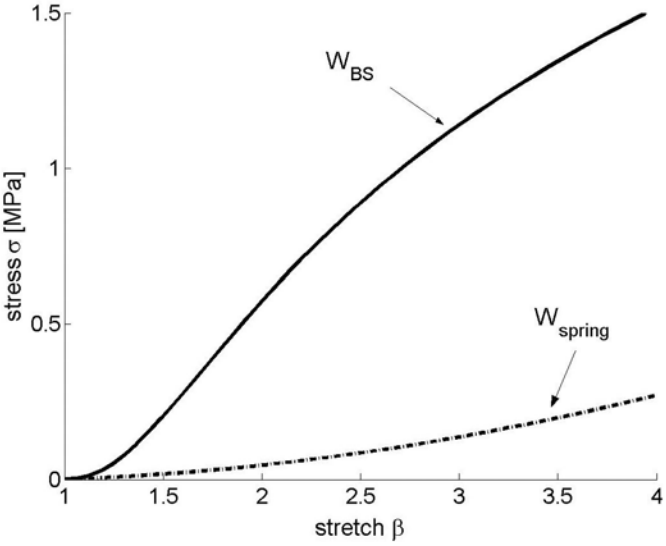

Fig. 4. Stresses resulting from the energies W_{BS} and W_{spring} in dependence on the stretch if the balls are essentially softer than the linkages ($k_i \approx 10^{-4}$), [2]

The most important result coming from the analysis of the 'balls and springs' material is as follows. Let β_i represent *principal stretches* at a point in a deformed state. The incompressibility of the material guarantees that

$$\beta_1\beta_2\beta_3 = 1\,. \tag{3}$$

We define some "effective stretches" as

$$\beta_i^{eff} = \frac{1+\delta_i}{1+k_i}(\beta_i - 1) + 1\,, \tag{4}$$

where k_i represents the ratio of stiffness of balls to stiffness of linkages in the *i*-th direction and δ_i is the ratio of the size of gap between balls to the ball dimension in the *i*-th direction. (Notice that $\beta_i^{eff} = 1$ whenever $\beta_i = 1$.) The study of the 'balls and springs' material shows [2] that if

$$\beta_1^{eff}\beta_2^{eff}\beta_3^{eff} = 1\,, \tag{5}$$

the energy W' vanishes. At the point where (5) is valid the strain-energy function has a deep local minimum (see Fig. 5).The effective stretches (4) depend also on parameters of the structure, k_i and δ_i. When changing slightly these parameters, we leave the minimum and the stiffness of the material may considerable increase.

Might it be the essence of a "trick" of Nature allowing smooth muscle to change very effectively its overall stiffness by certain processes within the cellular cytoskeleton? The smooth muscle cells are usually (in normal conditions) considerably softer than the extracellular matrix,

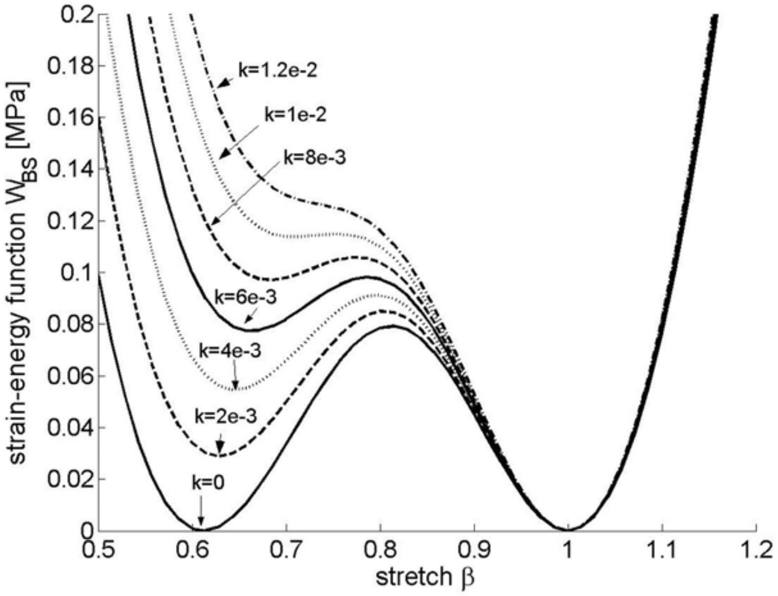

Fig. 5. The occurrence of a deep energy minimum in a (numerical) compression test of the 'balls and springs' material in dependence on the stretch ($\beta = 1$ represents the reference configuration), [2]

i.e. k_i are very small. Similarly, the gap between cells is very small compared to their size (in Fig. 1, the gap is about 250 nm) [9]. It means that β_i^{eff} are near to β_i that implies that the local state of the tissue is not far from the local minimum defined by (5) (since $\beta_1\beta_2\beta_3 = 1$). Small changes of cell stiffness (varying k_i) or their spatial dimensions (varying δ_i) thus may drive the material from/to the minimum and hence cause essential changes in its stiffness. In real smooth muscle cells, the molecular motors causes changes in strength of protein fibers that influence both the cells stiffness and their dimensions (i.e. all parameters k_i and δ_i may be varied in various manners).

3. The constant volume of cells as a control principle?

But smooth muscle cells are *not* balls with an impermeable surface filled with a constant amount of a fluid guaranteeing constancy of their volumes. The cell is an *open* system and there is a permanent exchange of the matter with its surrounding (a variable permeability of membrane lipid bilayers allows water to move across the plasma membrane if the osmotic strength of the extracellular space changes even by a small amount). Nevertheless, cells employ both short- and long-term strategies to *maintain a constant volume* by some mechanisms that compensate volume changes caused by flows through the membrane (e.g. cells respond to loss of water by activating ion pumps that bring ions into the cell and water follows, returning the cell to its original volume in minutes). These mechanisms are well defined, but how cells sense volume changes or trigger these responses is less clear [6].

Another question is *why* do cells sustain complicated mechanisms to keep their volumes constant upon any terms? The results obtained by studying a toy model of cellular structure – the 'balls and spring' material – might outline a partial answer. Namely, the maintenance of a constant volume may be explained as a simple *control mechanisms* allowing cells to tune and govern their mechanical properties. In a highly complicated structure of complex protein fibers, many deformable bodies, fluids carrying many chemical components, etc., the constant volume may serve as a “fix point” enabling cells to use complicated chemical mechanisms on polymer networks to produce *effectively* various macroscopically meaningful effects (forces, change of stiffness, movement etc.).

To support this idea we notice an interesting phenomena well-known in cellular mechanics. Namely, it is an experimental evidence that the cytoskeleton carries the pre-existing tension, the so-called prestress. Even at states with no outer load, the fibers are not relaxed. Since the cytoskeleton is responsible for the mechanical behavior of the whole cell, the prestress consequently plays an important role.

At the cellular level, the effect called prestress-induced stiffening is experimentally observed [13]. It means that with increasing prestress within cytoskeleton, the stiffness of the cell increases. This dependence is almost linear, as depicted in Fig. 6. The prestress is thus an important mechanism how cell controls its mechanical response. By increasing prestress, the stiffness increases and thus the cell actively resists deformation.

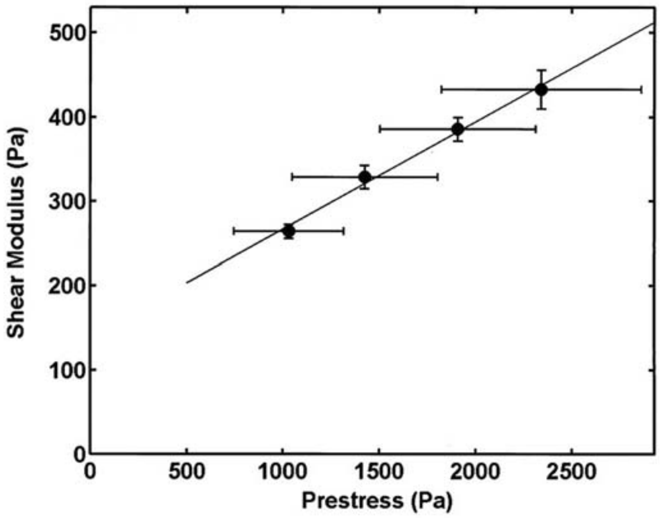

Fig. 6. The relation between prestress and shear modulus in smooth muscle cells [13]

Nevertheless, the prestress of a structure (e.g. a guitar with a tight string) may or may not mean the increase of the structure stiffness. If the stress in the guitar string depends linearly on its strain, for instance, the stiffening cannot occur. To find this effect in linear structures we have to suppose the existence of some elements – “bars” being able to carry a compression, that are combined with other elements in a special arrangement. It is popular to “sight” such structures (called the tensegrity, see e.g. [13]) as formed within the cellular cytoskeleton. Nevertheless, it is less clear how such complicatedly designed structures could be formed by spontaneous processes in cellular polymer networks, how their stability could be guaranteed, and so on.

When accepting the idea that cells control their volume to be constant, we can define the prestress simply by the assumption that the value of the volume maintained by a cell differs from that in which the cell might reach the minimum of energy [12]. In the case of the ‘balls and springs’ structure, the constant cell’s volume is proportional to

$$V \equiv c_1^{ref} c_2^{ref} c_3^{ref} \,, \tag{6}$$

where c_i^{ref} is the size of an individual ball in the i-th direction at the reference state (state with no external load). On the other hand, the volume vanishing the cell’s energy is proportional to

$$V_0 \equiv c_1^{(0)} c_2^{(0)} c_3^{(0)} \,, \tag{7}$$

where $c_i^{(0)}$ are the rest lengths of the cell's reinforcement (linear springs). The important assumption allowing us to incorporate the prestress into the 'balls and springs' structure is that V differs from V_0. For simplicity, let us suppose that

$$V \geq V_0 \,. \tag{8}$$

In the case of equality, there is no prestress within the structure. When the sharp inequality holds, the inner springs must be stretched to $c_i^{ref} > c_i^{(0)}$ to span the volume of V. In other words, constant volume works as a constraint which does not allow the inner springs to relax. For convenience, we introduce the (dimensionless) numbers

$$P_i = 1 - \frac{c_i^{(0)}}{c_i^{ref}} \,, \tag{9}$$

which values range from 0 (no prestress in the *i*-th direction) to 1 (limit value of prestress), to quantify the level of prestress.

The effect of prestress on the mechanical response of the corresponding 'balls and springs' material is determined via the strain-energy function [11]. To emphasize its dependence on prestress, the strain-energy function is denoted with W_{BS}^p. Analogous to (2), it can be divided into two contributions (both depending on prestress),

$$W_{BS}^p = W_{spring}^p + W_p' \,, \tag{10}$$

where W_{spring}^p is the "averaged" elasticity of springs and W_p' is the contribution due to the arrangement of microstructure. Analogous to the section 2, the "effective stretches" can be introduced as

$${}^p\beta_i^{eff} = \frac{1+\delta_i}{1+k_i}(\beta_i - 1) + 1 - \frac{k_i}{1+k_i} P_i \,. \tag{11}$$

Compared to (4) an additional term appears due to prestress, see [11] for details. It is worth stressing that for zero prestress ($P_i = 0, \forall i$), the prestress-dependent quantities coincide with those defined in section 2,

$$W_{BS}^p \to W_{BS}, \quad W_{spring}^p \to W_{spring}, \quad W_p' \to W', \quad {}^p\beta_i^{eff} \to \beta_i^{eff} \,. \tag{12}$$

Prestress-induced stiffening of the 'balls and springs' material can be shown by determining the Young's modulus. It is defined as

$$Y_{BS} = \left. \frac{\mathrm{d}^2 W_{BS}^p}{\mathrm{d}\varepsilon^2} \right|_{\varepsilon=1} \,, \tag{13}$$

where ε is a small deformation ($\beta_1 = 1 + \varepsilon \,, \quad \beta_2 = \beta_3 = (1+\varepsilon)^{-1/2}$ in the case of transverse isotropy). Unfortunately, it is not possible to determine the analytical solution of (13) in general. However, if

$${}^p\beta_1^{eff}\, {}^p\beta_2^{eff}\, {}^p\beta_3^{eff} \approx 1 \,, \tag{14}$$

it is possible to find an approximative formula,

$$Y_{BS}(P_1) \approx Y_0 \left[1 + f \cdot P_1\right] \,, \tag{15}$$

where Y_0 is the Young's modulus of the structure with no prestress, and f is a function of material parameters [10, 11]. Notice that the condition (14) is fulfilled for any stretch and any prestress if $k_i \ll 1$ and $\delta_i \approx k_i$. It means that the approximative formula is accurate for the case of smooth muscles (cells are considerably softer than the extracellular matrix and gaps between neighbouring cells are very small compared to their sizes).

The result shows the linear dependence of the stiffness on the level of prestress for the 'balls and springs' material. In fact, it corresponds to the mechanical behavior of a single microscopic element that is composed of an individual ball (representing the smooth muscle cell) surrounded by extracellular matrix [10]. It is in qualitative agreement with the so-called prestress-induced stiffening that is observed for living cells (see Fig. 6). By increasing a pre-tension (caused by shortening the rest lengths of cytoskeletal fibers) the stiffness of the cell grows. The prestress can thus be understood as a mechanism by which the living cells control their overall mechanical response.

As the numerical simulations illustrate, the similar effect of the prestress-induced stiffening is exhibited for the 'balls and springs' material also at the macroscale. In Fig. 7, the uniaxial traction test with the macroscopic sample (sizes in the order of at least millimeters) is depicted. The level of prestress is set at the microscale since it corresponds to the change of rest lengths of fibres within individual balls (sizes of the order of micrometers). From the figure it is clear that by increasing the prestress the overall mechanical response is stiffer and the material thus resists deformation.

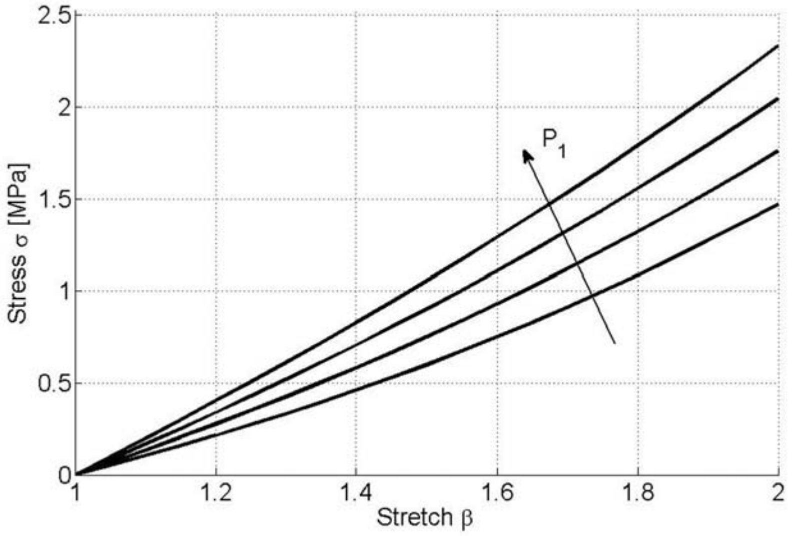

Fig. 7. Uniaxial traction test with the 'balls and springs' material. With an increasing prestress the material stiffens

4. Tissues' viscoelasticity

The 'balls and springs' model has been build on the hypothesis of elasticity. However a slight modification of the 'balls and springs' model allows us to catch a specific viscoelastic property that is also observed in biological tissues. Namely, when straining the tissue, the fluid filling the space between cells has to move. The reason is that the gap between neighbouring cells slightly varies and the fluid has to fill or drain the spaces. This movement is connected with some viscous forces that may be introduced in our model. The resulting continuum limit describes a material with internal variables.

In the continuum description, the thermodynamic state of elastic materials is fully determined by two variables distributed in space, $\mathbf{F}$ and Θ, the deformation gradient and the temperature distribution, respectively. Concerning dissipative materials, other state variables are necessary to characterize the inelastic behavior of the material. Such variables usually cannot be defined as some macroscopic quantities. Nevertheless, we suppose a set of some “hidden” ones that are not directly accessible by experimental measurements at the macroscopic level. We call them the *internal variables*.

Let us suppose an occurrence of n scalar internal variables, α_i, $i = 1, \ldots, n$. If all processes are isothermal, an explicit dependence on the temperature may be omitted. The fact that the internal variables are state parameters implies that the Helmholtz free-energy function may be written in the form $\Phi = \Phi(\mathbf{F}, \boldsymbol{\alpha})$ (where $\boldsymbol{\alpha}$ represents the set of all internal variables α_i). Following [3], we assume the dissipation in the material to be characterized by functions Ξ_i such that the internal dissipation, D_{int}, is described as

$$D_{int} = \sum_{i=1}^{n} \Xi_i(\mathbf{F}, \dot{\mathbf{F}}, \boldsymbol{\alpha}, \dot{\boldsymbol{\alpha}})\, \dot{\alpha}_i\,, \tag{16}$$

where the dot denotes the time derivative of the state variables. The second law of thermodynamics claims the validity of the Clausius-Planck inequality, namely

$$D_{int} = w_{int} - \dot{\Phi} \geq 0\,, \tag{17}$$

where $w_{int} \equiv \Pi : \dot{\mathbf{F}}$ is the rate of internal work and Π is the first Kirchhoff stress measure. Applying the chain rule for the time derivative of the free-energy function, we obtain from (17) that

$$D_{int} = \left(\Pi - \frac{\partial \Phi}{\partial \mathbf{F}}\right) : \dot{\mathbf{F}} - \sum_{i=1}^{n} \frac{\partial \Phi}{\partial \alpha_i} \dot{\alpha}_i \geq 0\,. \tag{18}$$

To fulfill the inequality (18) in all admissible processes the constitutive law,

$$\Pi = \frac{\partial \Phi(\mathbf{F}, \boldsymbol{\alpha})}{\partial \mathbf{F}}\,, \tag{19}$$

has to be valid. With (16), it implies that the time evolution of the n internal variables α_i is governed by n differential equations,

$$\Xi_i(\mathbf{F}, \dot{\mathbf{F}}, \boldsymbol{\alpha}, \dot{\boldsymbol{\alpha}}) + \frac{\partial \Phi(\mathbf{F}, \boldsymbol{\alpha})}{\partial \alpha_i} = 0\,. \tag{20}$$

In absence of dissipation (i.e. if $\Xi_i = 0$) the differential equations (20) reduce to a system of non-linear equations

$$\frac{\partial \Phi(\mathbf{F}, \boldsymbol{\alpha})}{\partial \alpha_i} = 0, \tag{21}$$

fixing the internal variables to their expressions $\boldsymbol{\alpha}^*(\mathbf{F})$ minimizing the energy at a given macro-deformation, and the free-energy function reduces to the strain-energy function, i.e.

$$\Phi(\mathbf{F}, \boldsymbol{\alpha}^*(\mathbf{F})) = W_{BS}(\mathbf{F}). \tag{22}$$

On the contrary, the expression of the internal variables (solution of 20) are time-dependent, i.e. $\boldsymbol{\alpha}(t)$, when the dissipation is taken into account.

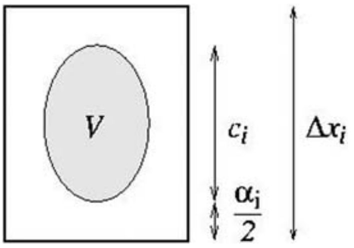

Fig. 8. Used notation in the RVE cross-section

With the 'balls and springs' structure, it is natural to associate the internal variables with some parameters characterizing the 'microstructure' as, for instance, the three principal dimensions of the gap separating neighboring balls. The balls dimensions c_i are functions of α_i and $\mathbf{F}$. Namely, they are given by the differences between the RVE dimensions, say Δx_i, and the distances α_i separating neighboring balls (see Fig. 8), i.e.

$$c_i(\mathbf{F}, \boldsymbol{\alpha}) = \Delta x_i(\mathbf{F}) - \alpha_i = \beta_i \Delta x_i^{ref} - \alpha_i \,, \tag{23}$$

where $\Delta x_i^{ref} = c_i^{ref} + \alpha_i^{ref}$ is the dimension of the RVE in the reference configuration. Since we consider the only dissipation involved by the extracellular fluid movement, the dissipation D_{int} is defined as the power of viscous forces necessary to fill or drain fluid in the gap between balls. Using the dimensional analysis we obtain [5] the expression

$$\Xi_i(\mathbf{F}, \dot{\mathbf{F}}, \boldsymbol{\alpha}, \dot{\boldsymbol{\alpha}}) = \frac{\dot{\alpha}_i}{\alpha_i^3} \frac{\tau G (c_j c_k)^2}{V^{rve}}, \;\; j \neq k, \;\; i \neq j, \;\; i \neq k \,, \tag{24}$$

where G is a characteristic energy per volume (it is a function of the material stiffness), V^{rve} is the characteristic material size (volume of the Representative Volume Element of tissue) and $\tau \equiv \eta\kappa/2G$ is a characteristic time of the problem (η is the viscosity of the extracellular fluid and κ is a dimensionless number).

The effect of the extra-cellular fluid viscosity on material behavior is clearly illustrated by the occurrence of hysteresis during cycles of loading/unlaoding tests, see Fig. 10. The loading and unloading branches differ, forming a loop which inner area measures the energy dissipation. A permanent loop (preconditioning) is reached after more or less cycles depending on material parameters.

5. Inspiration for engineering materials?

The 'balls and springs' material shows the way in which macroscopic mechanical properties of a specially designed material may be tuned by small changes within its structure. The main "trick" consists in occurrence of very soft cells within the material that are, however, firmly connected with surrounding (stiff) material. The incompressibility of cells then realizes a special linkage between deformations in different spatial directions. The tuning of the material stiffness consists in defining a measure of *how much* the stiff matrix has to deform in other directions to obtain the demanded deformation in a chosen direction. The existence of deep local minimum in the energy "landscape" means that small changes in material structure may influence the material stiffness essentially.

In engineering practice there are various materials (metals, wood, plastics, polymer composites and many others) whose mechanical properties depend on their structure and are governed

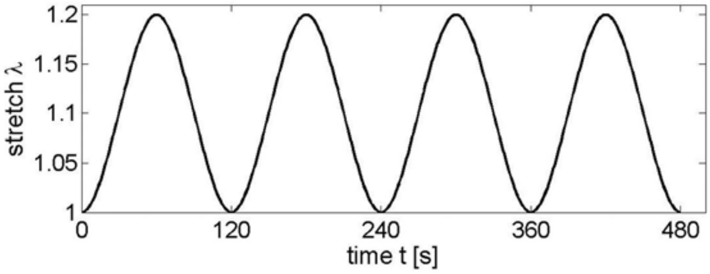

Fig. 9. Loading/unloading cycles: a strain oscillating between the values 0 and 20 % in a run of 60 s (as a polynomial of third degree) is applied to the material sample

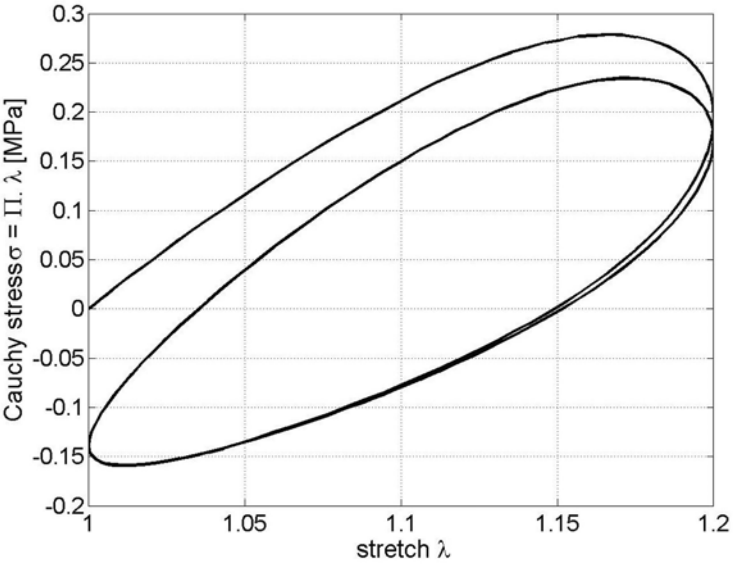

Fig. 10. The relationship between stress and stretch draws hysteresis loops when the loading process shown in Fig. 9 is applied. (The softening at the hysteresis angles results from the slowing of the stretch rate near the stretch peak $\lambda = 1$ or 1.2 due to the polynomial approximation.)

by different mechanisms in which the intermolecular forces play an important role. These forces also realize bindings among deformations in individual spatial directions so that they contribute to the material stiffness. The cellular structure of living tissues, however, allows tissues to use such bindings effectively so that the tissue may change its mechanical properties in a large extent. It may be an inspiration for designing some "smart" engineering materials – similar effects might arise, for example, in some special multi-phase polymer materials which materials sample manufacture is the object of our further efforts.

Another inspiration comes from a special "interaction" of fluids and the inclusions ("balls") within the structure. This special arrangement contributes in a special way to the structure viscous properties. This contribution has different properties than that coming only from the polymeric viscosity and depends on the structural parameters of the material. As a result, the viscous behavior of such a material might be also tuned by some microstructural changes.

Acknowledgements

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