Influence of nanoscale morphological characteristics on the mechanical properties of bone tissue

Artem S. Semenov Alexander S. Avrunin Alexey I. Grishchenko
Boris E. Melnikov Alexander A. Doktorov
semenov.artem@googlemail.com

Abstract

Bone is a hierarchically structured material with mechanical properties depending on its morphological parameters at all levels of hierarchy. At a nanolevel, bone is a composite with a quasiperiodic structure, consisting of hydroxyapatite crystals, which are embedded into the collagen fibrils. The purpose of the paper is to analyze the influence of the nanostructure of bone on its elastic and strength properties. Such studies are important for the creating artificial bone-substitute materials.

The new morphological nanoscale model of bone is proposed with taking into account the mineral bridges between the associations of the hydroxyapatite crystals. The analysis of variation influence in the morphological characteristics (hydroxyapatite crystals disorientation, sizes and orientation of mineral bridges, mineralization) on the local stress-strain state and mechanical properties of the representative volume element of bone is carried out by means of the direct finite element simulation and homogenization. The comparison of obtained results with experimental data is performed and discussed.

1 Introduction

The mechanical properties of a skeleton depend on the characteristics of the morphological organization of the elements forming the bone structures at each scale level [1, 2, 3, 4]. Often, methodological difficulties arise in the study of this relationship by objective methods, which can be overcome by means of numerical simulation. Classic examples of this are the studies of G. Galilei [5] and J. Wolff [6]. G. Galilei using this approach for the first time showed the connection of size and shape of bones with a body weight under a typical loading [5]. J. Wolff [6] formulated the law connecting the morphological parameters of the distribution of the trabeculae in the proximal femur with its mechanical properties. These and other studies clearly showed the influence of morphological parameters of the spatial organization of the skeleton from the macro- to nano-level on the mechanical properties [3, 7, 8].

At the present stage the computer simulation are used first of all in the study of the morphological characteristics of bone structures at the nanoscale. The results of computations of such problems are an important source of scientific information, opening the possibility to investigate the effect of microstructure on the mechanical properties of a bone tissue by means of multivariate numerical experiments. The proposed computational model of bone at nanolevel is developed on the basis of the morphological model by Yu.I. Denisov-Nikolsky and coauthors [9].

Most attention in the present investigation is paid to the analysis of influence of the following characteristics: the spatial order of minerals, their deviations from longitudinal axes, the presence of the mineral bridges between them, forming a rigidly connected mineral structure in each bone.

Structural ordering of mineral matrix depends on the ordering of organic matrix, as the latter initiates the nucleation of minerals and determines their directional growth, thus defining the spatial organization of the mineral matrix [4]. In other words, the mineral matrix in each locus of bone is programmed reflection of the organic ultrastructure. Therefore, the optimal spatial organization of the organic matrix relative to trend lines of everyday mechanical loads leads to the optimal mechanical properties of bone structures at each locus of the skeleton.

Mineral bridges between the minerals combine the mineral complex into a single matrix in each bone and forced to re-evaluate the role of organic and mineral components for mechanical properties of bone structures. Mineral bridges impugn the validity of the two dominant at present concepts ignoring their existence. According to the first a collagen prevents destruction of bone structures under tension, and mineral under compression [1]. According to the second a bone is a two-phase composite that combines linear-elastic behavior of the collagen and elastic-plastic mineral [1, 2].

Mineralization is an important morphological parameters of bone matrix. The mineralization is defined as the mass fraction of nanocrystals of calcium phosphate (hydroxyapatite) in the total weight of the bone matrix. Hydroxyapatite nanocrystals incorporated into the fibers of the mineral matrix increase their stiffness, but reduce the strength [10]. However bone marix has evolved a complex hierarchical structure and combines the high stiffness of the mineral phase and the high toughness of organic phase.

The purpose of this work is (i) to develop nanolevel morpho-structural model of the bone matrix with the union of its mineral elements into a single structure by means of mineral bridges and (ii) to determine the influence of mineralization and the axial disorder of its elements on the effective elastic moduli and on the strain and stress fields in the modeled structure.

2 Representative volume element

At the nanoscale (on the collagen fibrils level) bone is a heterogeneous material with a pronounced micro-inhomogeneity of the mechanical properties. A way of modeling a structure made of heterogeneous materials without treating all of the heterogeneities individually consists in trying to replace the heterogeneous medium by an equivalent homogeneous medium (EHM) endowed with so-called effective properties. The effective mechanical modules are used for the evaluation of the mechanical properties on the higher scale level indirectly sensitive to microstructure on the lower scale level. The effective properties are calculated on the base of the spatial average of stress-strain state within the representative volume element (RVE) of material.

The choice of RVE for the nanocomposite of bone is not a trivial task. At the nanoscale level, bone is not a perfect periodic structure, therefore it is possible to introduce various RVE with different degree of simplification of the real situation. The RVE can be introduced for a material with a statistically uniform distribution (ergodic hypothesis) taking into account the scale-separability of heterogeneities. In this case the least volume containing all the a priori statistical information on the distribution and morphology of the material heterogeneities can be correctly introduced.

The simplest two-dimensional variant of RVE (unit cell) (fig. 1a), which takes schematic into account the disordered orientation, is presented by the central conglomerate deviated from the vertical direction and 4 fragments of neighboring undeflected minerals ($\frac{1}{4}$ the area of each) with 4 bridges. Three-dimensional finite element (FE) model of RVE is shown in fig. 1b. The other types of RVE are considered in [11].

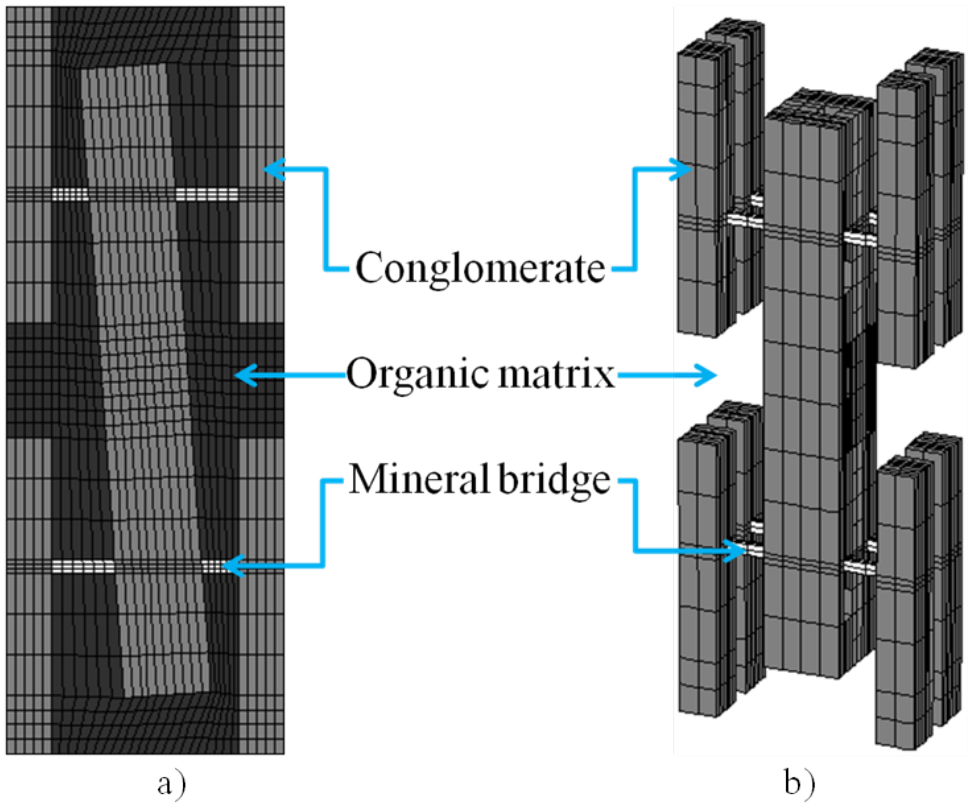


Figure 1: FE models of bone RVE at nanoscale: a) 2D model, b) 3D (organic matrix is not shown).

FE models of RVE are developed under an assumption that each pair of neighboring conglomerates (crystallites associations) has only one bridge, conglomerates are staggered and they are embedded in the organic matrix. RVE is considered as a heterogeneous three-component material. Boundary value problems for the stress-strain state determination in RVE are solved under an assumption of the linear elastic behavior. The mechanical properties of the individual components correspond to the isotropic material. The elastic moduli of bone components at the nanoscale are taken from the literature [12, 13] (see Table 3).

Table 3: The values of the elastic moduli of the individual components of RVE.

			Hydroxyapatite	Collagen	Bridge
Young's modulus	E	GPa	65	0.65	65
Poisson's ratio	ν	-	0.2	0.49	0.2

The computations have been performed using the FE program PANTOCRATOR [14], which allows to generate automatically the discrete models of RVE and to compute the effective elastic moduli of RVE by means of solutions of boundary value problems with subsequent analysis of the stress and strain fields distribution.

3 Results of FE modeling of deformation processes of bone RVE

FE simulations of the deformation processes of RVE are aimed to solve two main subtasks:

- to identify effective mechanical properties of the bone nanocomposite RVE (homogenization problem);
- to obtain the extreme values of stress fields within the heterogeneous RVE for the subsequent strength analysis (heterogenization problem).

3.1 Effective elastic properties

The effective elastic properties of the bone nanocomposite RVE are found by means of methods of the FE homogenization. It is supposed that the effective properties of homogenized bone material correspond to orthotropic elastic material, for which Hooke's law can be written as:

$$\bar{\boldsymbol{\varepsilon}} = {}^4\bar{\boldsymbol{C}} \cdot \cdot \bar{\boldsymbol{\sigma}}, \tag{1}$$

where $\bar{\boldsymbol{\varepsilon}} = \frac{1}{V_{RVE}} \int\limits_{V_{RVE}} \boldsymbol{\varepsilon} dV$ is the spatial averaged strain tensor, $\bar{\boldsymbol{\sigma}} = \frac{1}{V_{RVE}} \int\limits_{V_{RVE}} \boldsymbol{\sigma} dV$ is the spatial averaged stress tensor, ${}^4\bar{\boldsymbol{C}}$ is tensor of effective elastic compliances of 4th rank. The bar over tensors indicates a correspondence to the homogenized material. For orthotropic materials the tensor ${}^4\bar{\boldsymbol{C}}$ in its own axes of anisotropy corresponds to a symmetric matrix of elastic compliances $\left[\bar{\boldsymbol{C}}\right]$ with following structure:

$$\left[\bar{\mathbf{C}}\right] = \begin{bmatrix} \frac{1}{\bar{E}_1} & -\frac{\bar{\nu}_{21}}{\bar{E}_2} & -\frac{\bar{\nu}_{31}}{\bar{E}_3} & 0 & 0 & 0 \\ -\frac{\bar{\nu}_{12}}{\bar{E}_1} & \frac{1}{\bar{E}_2} & -\frac{\bar{\nu}_{32}}{\bar{E}_3} & 0 & 0 & 0 \\ -\frac{\bar{\nu}_{13}}{\bar{E}_1} & -\frac{\bar{\nu}_{23}}{\bar{E}_2} & \frac{1}{\bar{E}_3} & 0 & 0 & 0 \\ 0 & 0 & 0 & \frac{1}{\bar{G}_{12}} & 0 & 0 \\ 0 & 0 & 0 & 0 & \frac{1}{\bar{G}_{23}} & 0 \\ 0 & 0 & 0 & 0 & 0 & \frac{1}{\bar{G}_{31}} \end{bmatrix}, \quad \frac{\bar{v}_{ij}}{\bar{E}_i} = \frac{\bar{v}_{ji}}{\bar{E}_j}, \quad i, j = 1, 2, 3. \tag{2}$$

The elastic moduli are determined on the basis of relations $\bar{E}_i = \frac{\bar{\sigma}_{ii}}{\bar{\varepsilon}_{ii}}$, $\bar{v}_{ij} = -\frac{\bar{\varepsilon}_{jj}}{\bar{\varepsilon}_{ii}}$. 2D boundary value problems are solved assuming the plane strain condition. The determination of the two elastic moduli $\bar{E}_1$ and $\bar{E}_2$ requires to solve only two boundary value problems with boundary conditions for tension (or compression) in the directions of the anisotropy axes (in the vertical and in the horizontal directions for the RVE in Fig. 1)

The evaluation of the accuracy of numerical solutions is based on the analysis of practical convergence of the effective elastic moduli with increasing number of unit cells and number of finite elements. The upper and lower boundaries for moduli are obtained using boundary conditions for the displacements and for the tractions. Three types of boundary conditions are applied and compared:

kinematic uniform boundary condition

$$\boldsymbol{u}|_{S_u} = \bar{\boldsymbol{\varepsilon}}^* \cdot \boldsymbol{r}, \tag{3}$$

static uniform boundary condition

$$\boldsymbol{n} \cdot \boldsymbol{\sigma}|_{S_\sigma} = \boldsymbol{n} \cdot \bar{\boldsymbol{\sigma}}^*, \tag{4}$$

periodicity condition

$$\boldsymbol{u}|_{S_{u_1}} = \boldsymbol{u}|_{S_{u_2}} + \bar{\boldsymbol{\varepsilon}}^* \cdot (\boldsymbol{r}_1 - \boldsymbol{r}_2), \tag{5}$$

where $\bar{\boldsymbol{\varepsilon}}^*$ and $\bar{\boldsymbol{\sigma}}^*$ are prescribed constant symmetric tensors corresponding to the different possible states (axial tensions or shears), $\boldsymbol{r}$ is the radius-vector. The periodicity condition (5) can be rewritten in the form $\boldsymbol{u}|_{S_u} = \bar{\boldsymbol{\varepsilon}}^* \cdot \boldsymbol{r} + \tilde{\boldsymbol{w}}$, where a fluctuation $\tilde{\boldsymbol{w}}$ is periodic, i.e., $\tilde{\boldsymbol{w}}$ takes the same values on opposite sides of RVE. In this case also the traction $\boldsymbol{n} \cdot \boldsymbol{\sigma}$ takes opposite values on opposite sides. The boundary conditions (3)-(5) satisfy to Hill homogeneity condition and provide the existence and uniqueness of the solution of the corresponding boundary value problems.

As a result of multivariant computational experiments, it is found that by using periodicity conditions (5) satisfactory accuracy (close enough to the asymptotic value) is achieved (Fig. 2) even by using of RVE including one bridge ($\frac{1}{4}$ of the unit cell in Fig. 1). Whereas with the boundary conditions (3) and (4) the convergence is achieved (Fig. 2) only if RVE includes 64 bridges (4×4 unit cells) or more. The application of (5) allows to reduce significantly the dimension of FE model and the computation time.

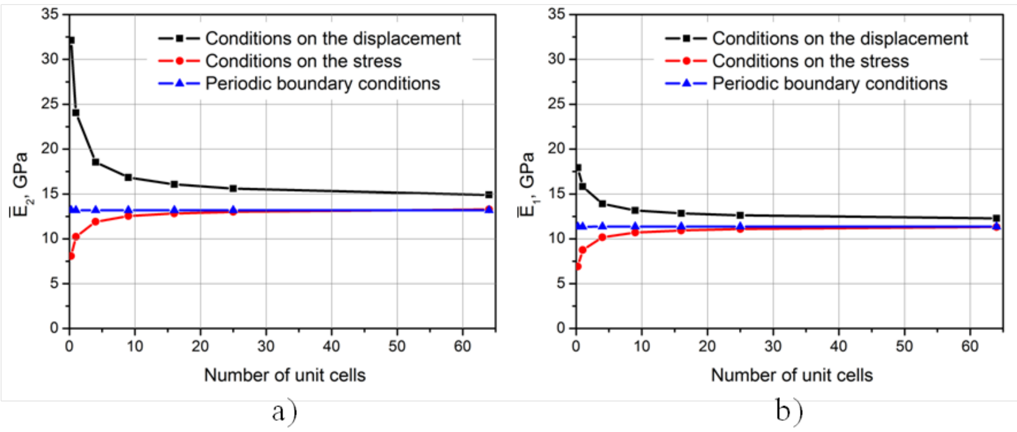


Figure 2: Dependence of the effective elastic moduli a) $\bar{E}_2$ (in the vertical direction) and b) $\bar{E}_1$ (in the horizontal direction) on the number of unit cells.

3.2 Analysis of the stress-strain state of RVE with bridges for different types of loading

Under tension of RVE in the vertical (Fig. 3a,b,e) and horizontal (Fig. 3c,d,f) directions there are two areas of maximum von Mises stresses σ_i: at the corners of conglomerates and in the joints of bridges with conglomerate. Stresses in the conglomerate are much higher than in the organic matrix (Fig. 3a,c) and the strains on the contrary are much lower (Fig. 3b,d). The stress components distribution is considered in more detail in [11].

The bridge is the most loaded element of RVE under both vertical (Fig. 3a,b) and horizontal (Fig. 3c,d) loading. The corners of conglomerates provides extremal stresses especially at the vertical loading (compare Fig. 3a and c). The bridge under the vertical loading works on a shear (Fig. 3e) and the stress state corresponds to the bending. Under the horizontal loading the bridge works on the tension/compression (Fig. 3f). Thus the largest risk of fracture under both horizontal and vertical loading occurs in zones adjacent bridge to conglomerate, but in the first case it is higher.

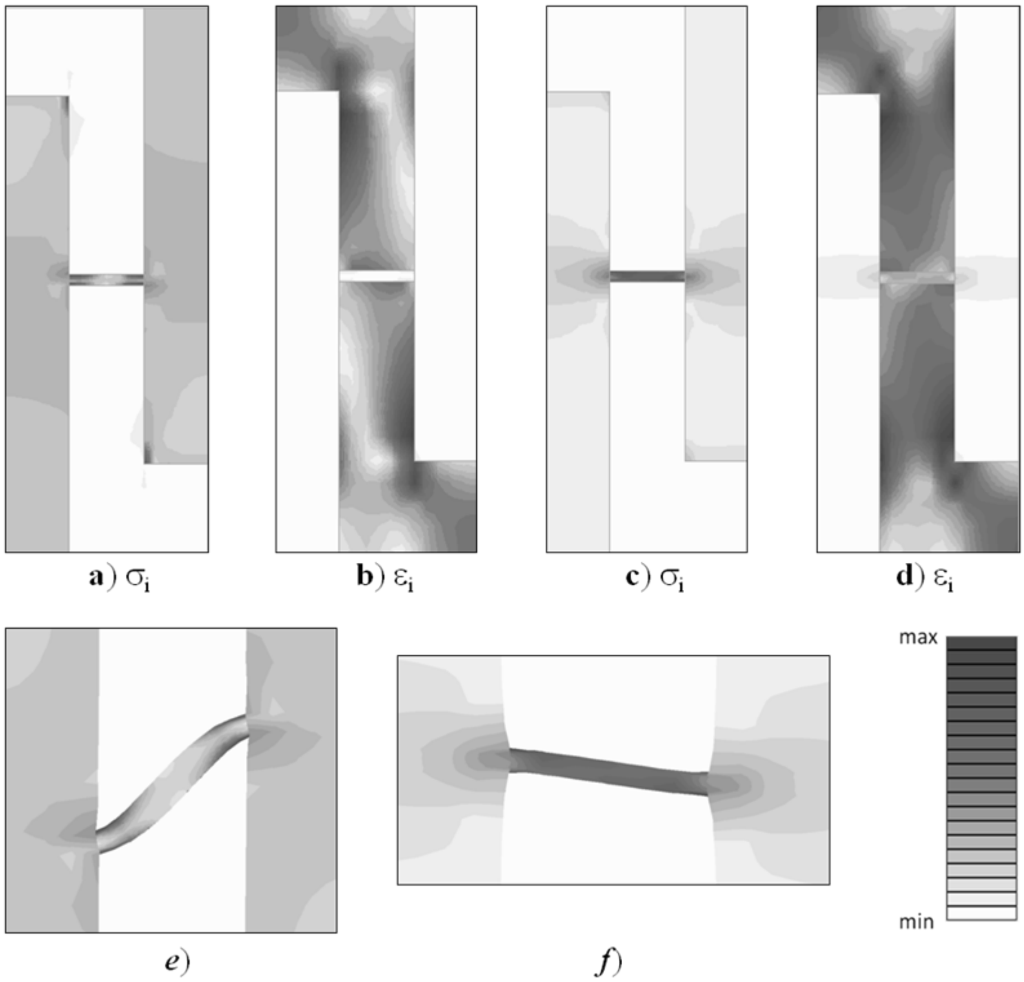


Figure 3: The field distribution in the central fragment of RVE ($\frac{1}{4}$ part) of: a) von Mises stress σ_i under vertical tension; b) von Mises strain ε_i under vertical tension; c) von Mises stress σ_i under horizontal tension; d) von Mises strain ε_i under horizontal tension. The deformed state of bridge (with fields of von Mises stress) under: e) vertical and f) horizontal tensions.

3.3 Comparison of the effective elastic moduli with experimental data

The results of computations of the effective elastic moduli $\bar{E}_1$ and $\bar{E}_2$ for the RVE with taking into account the bridges demonstrate sufficient accuracy in comparison with experimental data (Table 4).

3.4 Influence of the bridges on the effective elastic moduli and stress-strain state

A comparison of the FE results for bone RVE models with bridges and without has been performed. It was found that the presence of bridges increases the level of extreme stress (relative stress intensity $\sigma_i/\bar{\sigma}_i$ on 52%) under tension in the horizontal direction. In this case the level of stress intensity at the corner of the mineral is reduced by 19%. Under tension of RVE in the vertical direction the presence of bridges reduces extreme von Mises stress intensity in the corners of conglomerate by 4%.

Table 4: Comparison of the computed effective elastic moduli $\bar{E}_2$ and $\bar{E}_1$ with experimental data [15], [16], [17].

	The elastic modulus in the vertical direction $\bar{E}_2$, GPa	The elastic modulus in the horizontal direction $\bar{E}_1$, GPa
FE model of RVE	13.2	11.3
Katsamanis F. [15]	16.2	-
Bonfield W. [16]	18.5	9.5
Reilly D.T. [17]	17.0	11.5

Bridges increase the elastic modulus $\bar{E}_1$ (in the horizontal direction) by 17% (Table 3) and elastic modulus $\bar{E}_2$ (in the vertical direction) by 10%. It should be noted that the observed changes of the elastic moduli are caused by the change of internal structure RVE due to the appearance of bridges, but not due to slight increase of a mineralization, which is of two orders smaller.

Table 5: Comparison of the computed effective elastic moduli $\bar{E}_2$ and $\bar{E}_1$ for the RVEs with bridges and without.

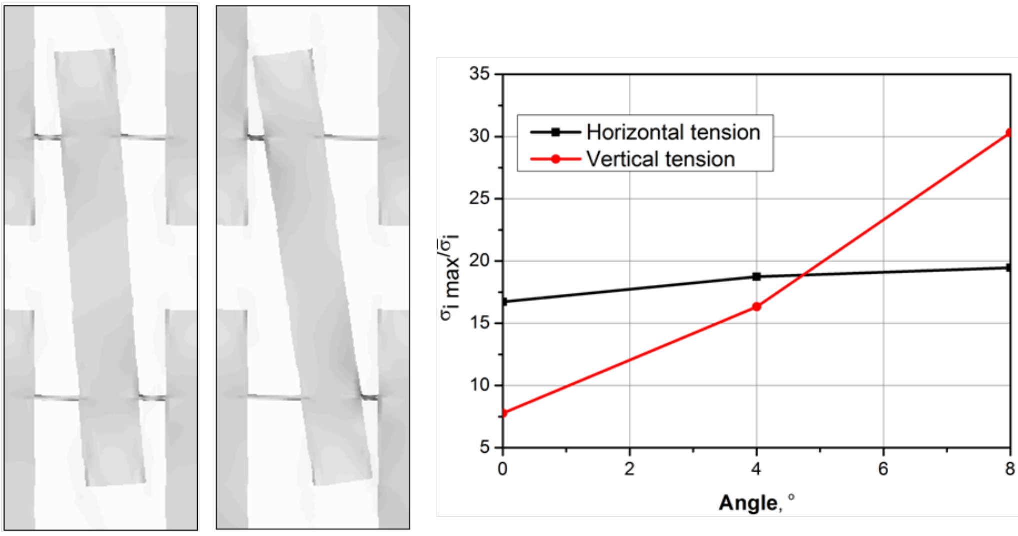
	The elastic modulus in the vertical direction $\bar{E}_2$, GPa			The elastic modulus in the horizontal direction $\bar{E}_1$, GPa		
Boundary conditions	Eq. (3)	Eq. (4)	Eq. (5)	Eq. (3)	Eq. (4)	Eq. (5)
RVE with bridges	16.0	12.7	13.2	12.7	10.8	11.3
RVE without bridges	14.9	11.7	11.9	11.0	9.3	9.6
Difference	7.4%	8.5%	10.1%	15.4%	16.1%	17.7%

Thus, the bridges affect on the deformation of collagen in the adjacent area, on the distribution of the stress intensity in the conglomerate in the vicinity of bridge joint and on the effective elastic moduli. The bridges increase the stiffness of bone tissue regardless of the direction of loading.

3.5 Influence of orientation of conglomerates

With aim to evaluate the effect of the angular orientation of the conglomerate on the stress-strain state and on the effective elastic moduli of RVE the FE computations are carried out for two different deviation angles of the central conglomerate from vertical direction: 4° and 8° (Fig. 4a,b). The results of computational experiments have shown that the rotation of a single conglomerate does not practically affect on the effective elastic moduli $\bar{E}_1, \bar{E}_2$ (difference is less than 1%), but it has considerable influence on the level of maximum stress intensity (Fig. 4c).

The obtained results allow us to establish that the disorder of conglomerates leads to an increase of the stresses in the bone RVE at nanoscale. Hence it reduces the strength properties of bone matrix on the nanoscale and therefore also the strength of bone on macrolevel.

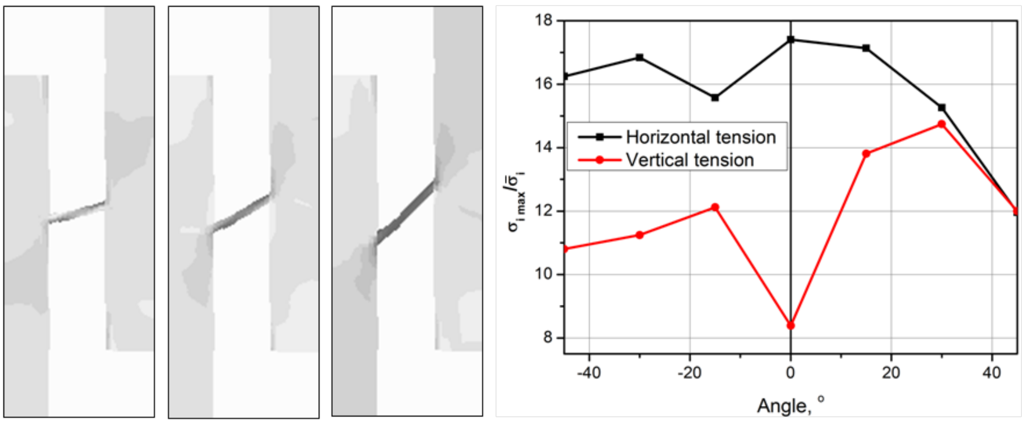


a) b) c)

Figure 4: Distribution of the von Mises stress fields in the RVE with rotated single conglomerate by a) 4° and b) 8° under the tension in vertical direction; c) dependence of the maximum von Mises stress on the central conglomerate rotation angle.

3.6 Influence of orientation of bridges

In order to evaluate the effect of the angular orientation of the bridge on the stress-strain state and on the effective elastic moduli of RVE the FE computations have been performed for three different deviation angles of bridges from the horizontal direction: 15°, 30° and 45° (Fig. 5a,b,c). The results of computational experiments have shown that the rotation of bridges has almost no effect on the effective elastic moduli $\bar{E}_1, \bar{E}_2$ (difference of less than 1%), but it has considerable influence on the level of maximum von Mises stress intensity (Fig. 5d).



a) b) c) d)

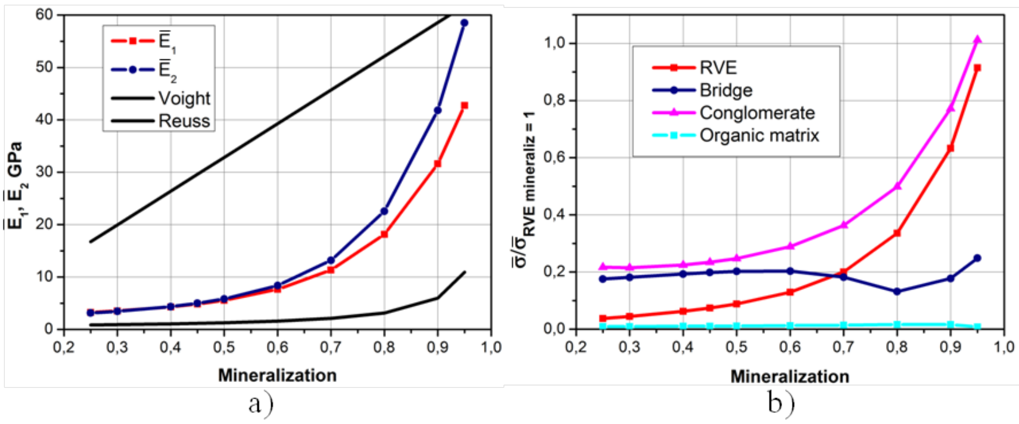
Figure 5: Distribution of von Mises stress intensity fields in the fragment of RVE with rotated bridge by a) 15°, b) 30°, c) 45° under the tension in the vertical direction; d) dependence of the maximum von Mises stress intensity on the bridge rotation angle.

The orientation deviation of bridge has multidirectional influence on the level of maximum von Mises stress in the RVE, leading in some cases to increase them, but also there are situations that lead to their reduction. Under tension in the horizontal direction the maximum von Mises stress at 15° deviation of bridge decreases by 2%, at 30° by 12%, at 45° by 47% in comparison with an idealized version without rotation (see Fig. 5d).

During the formation of bridges with the growth and development of bone unfavorably oriented variants of bridges can be destroyed by mechanical loads. The remaining bridges will have a directional orientation, changing with the age of the bone tissue and with the intensity of mechanical loadings. Thus, under the development of bone the optimal configuration of bridges occurs providing the stresses, which are lower than in the original configuration, and thus the creation of structures with improved strength characteristics compared to the original is takes place. The formation of similar texture affects on the absolute values and anisotropy of elastic and mechanical properties of bone tissue at the macro level.

3.7 Influence of bone mineralization

In order to evaluate the influence of the mineralization (mass fraction of hydroxyapatite (provided by conglomerates and bridges) in total mass of RVE) on the stress-strain state and on the effective elastic moduli of RVE the FE calculations are carried out with different mineralization levels: 25, 30, 45, 50, 60, 70, 80, 90 and 95%. The results of computational experiments have shown that the mineralization exerts considerable influence on the effective moduli (Fig. 6a) and on the averaged von Mises stress intensity (Fig. 6b). The increase of the mineralization from 60% up to 70% leads to increase of $\bar{E}_2$ on 55.7% and to increase of $\bar{E}_1$ on 47.7%. The same increase of the mineralization leads to increase von Mises stress intensity on 54%.



a) b)

Figure 6: Influence of the mineralization on: a) the effective elastic moduli (upper and lower curves correspond to the analytical estimates by Voigt (average of component stiffnesses) and by Reuss (average of component compliances)); b) the von Mises stress averaged over the whole volume of RVE and over its individual components.

The curves in Fig. 6 are obtained with proportional increase of conglomerate sizes under increasing mineralization. The deviation from these curves is observed under non-proportional change of the width and height of the conglomerate, which points out on the influence of a scenario of mineralization growth on the elastic moduli of bone.

Conclusion

1. The three-phase model of bone on submicroscopic level, taking into account the mineral bridges between the conglomerates of hydroxyapatite crystals, is proposed and investigated.

2. The effective elastic moduli of the RVE of the bone nanocomposite have been obtained by means of the finite element homogenization under assumption of orthotropic resulting properties. A comparison with experimental data shows a good agreement with the results of the proposed model.

3. The analysis of the stress-strain state of the RVE of bone tissue has been performed in order to determine the location of the most critical points and deformation mechanisms of bridges. The most loaded elements are the corners of conglomerates and corners of bridges.

4. The influence of bridges on the effective elastic moduli and on the stress-strain state has been analyzed. Taking into account of the bridges leads to decrease of the von Mises stresses in the corner of the conglomerate on 19% and to the increase of the effective elastic moduli in the vertical and horizontal direction on 10% and 17% respectively.

5. The effects of orientation of conglomerates and bridges on the stress state of the representative volume have been analyzed.

6. The influence of the bone mineralization on the effective elastic moduli and stress state has been investigated.

References

[1] Currey J.D. Three analogies to explain the mechanical properties of bone. Biorheology. 1964; 2:1-10.

[2] Burstein A.H., Zika J. M., Heiple K. G., Klein L. Contribution of Collagen and Mineral to the Elastic-Plastic Properties of Bone. J. Bone Joint Surg. Am. 1975; 57(7):956-961.

[3] Weiner S., Traub W. Bone structure: from angstroms to microns. FASEB J. 1992; 6(3):879-885.

[4] Avrunin A.S., Tikhilov R.M., Abolin A.B., Shcherbak I.G. Levels of organization of the mineral matrix of bone tissue and the mechanisms that define the parameters of their formation (analytical review). Morphology. 2005, 2:19-28.

[5] Galilei G. Dialogues concerning two new sciences. 1638. Translated from the Italian and Latin into English by Henry Crew and Alfonso de Salvio. With an Introduction by Antonio Favaro (New York: Macmillan, 1914).

[6] Wolff J. Das Gesetz der Transformation der inneren Architektur der Knochen bei pathologischen Veränderungen der Ausseren Knochenform. Sitzungsberichte der Königlich Preussischen Akadkmie der Wissenschaften zu Berlin. Sitzung der phys.-math. Classe v. 21. April. – Mittheilung v. 13. Man. 1884, 23 p.

[7] Weiner S., Wagner H.D. The material bone: structure-mechanical function relations. Annual Review Materials Science. 1998; 28(1):271-298.

[8] Currey J.D. Mechanical properties of vertebrate hard tissues. Proc. Inst. Mech. Eng. H. 1998; 212(6):399-411.

[9] Denisov-Nikolsky Yu.I., Mironov S.P., Omelyanenko N.P., Matveichuk I.V. Actual problems of theoretical and clinical osteoartrologe. Moscow: OAO "Typography" News", 2005. 336 p.

[10] Jager I., Fratzl P. Mineralized collagen fibrils: a mechanical model with a staggered arrangement of mineral particles. Biophys. J. 2000; 79(4):1737-1746.

[11] Avrunin A.S., Semenov A.S., Fedorov I.V., Melnikov B.E., Doctorov A.A., Parshin L.K. Effect of mineral bridges between crystallites on the mechanical properties of bone matrix. Traumatology and Orthopaedy. 2013. № 2

[12] Lawson A.C., Czernuszka J.T. Collagen-calcium-phosphate composites. Proc. Inst. Mech. Eng. H. 1998; 212(6):413-425.

[13] Fratzl P., Gupta H.S., Paschalis E.P., Roschger P. Structure and mechanical quality of the collagen–mineral nano-composite in bone. J. Mater. Chem. 2004; 14:2115-2123.

[14] Semenov A.S. PANTOCRATOR – the finite element program specialized on the non-linear problem solution. Proc. V Int. Conf. "Scientific and engineering problems of predicting the reliability and service life of structures and methods of their solution", 2003; 466-480.

[15] Katsamanis F., Raftopoulos D.D. Determination of mechanical properties of human femoral cortical bone by the Hopkinson bar stress technique. J. Biomech. 1990; 23(11):1173-1184.

[16] Bonfield W., Grynpas M.D. Anisotropy of the Young's modulus of bone. Nature. 1977; 270(5636):453-454.

[17] Reilly D.T., Burstein, A.H. The elastic and ultimate properties of compact bone tissue. J Biomech. 1975; 8(6):393-405.

Artem S. Semenov, SPbSPU, Politechnicheskaya 29; 195251, St. Petersburg, Russia

Alexander S. Avrunin, RITOS, Acad. Baykov 8; 195427, St. Petersburg, Russia

Alexey I. Grishchenko, SPbSPU, Politechnicheskaya 29; 195251, St. Petersburg, Russia

Boris E. Melnikov, SPbSPU, Politechnicheskaya 29; 195251, St. Petersburg, Russia

Alexander A. Doktorov, GNU VILAR, Grina 7; 117216, Moscow, Russia