collagen-PET vascular prostheses after amplification and seeding in high density [FIG.2A, 3B]. After exposure to short-term laminar shear stress similar to that in human vessels the cells showed good retention. Even if the cell detachement degree was statistically significant, the endothelium retained its confluent form on many of the investigated grafts [FIG.3B]. Immobilisation of the extracellular matrix protein laminin on the inner surface improved the cellular adhesion [FIG.2A] but decreased their shear stress resistance compared to the unmodified and fibrin-coated prostheses. Vice versa fibrin network coating [FIG.3A] resulted in worse adherence but better flow resistance compared to the graft with immobilized laminin.

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3D EVALUATION OF THE SURFACE ROUGHNESS USING STEREO IMAGES MADE IN SEM – INFLUENCE ON OSTEOBLAST CELL GROWTH

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The surface roughness creates one of the main conditions for contact of the living tissue with material. The cells adhesion, proliferation and differentiation are usually influenced by the surface morphology very strongly. Now, for biomaterial applications, it is necessary to better understand this influence and to define the roughness parameters, which control these interactions at the cell-material interface.
The surface roughness can be measured by number of methods; often, the contact profilometer can measure a number of parameters characterising the roughness. The disadvantage of this measurement is the line recording of the surface, which is principally two dimensional, and gives us information just from the path in one direction. The other possibility is to create the 3D reconstruction of a surface by suitable code from stereo-pair of images. For this method, using of Scanning Electron Microscope is very suitable and effective for its large depth of focus and simple change of imaging conditions.
We observed the osteoblast-like cells growing on carbon-carbon composites with various surface layers and also plain ones. Particular layers, which would by evaluated are pyrolytic graphite (PyG), titanium-carbon layer (Ti:C-H), diamond like carbon (DLC) and zircon nitride (ZrN). These materials were studied in the form of films prepared either by PACVD method or by pyrolysis on 2D C-C composites. The osteoblast-like cells were grown on the surface for defined time and generally adhesion, doubling time and spreading of cells were evaluated
The SEM images were obtained using JSM5410 scanning electron microscope (JEOL, Japan) and the *Scandium* software (Olympus Soft Imaging Solutions, Germany), which was used for 3-D surface reconstruction of images. Firstly, we compared the roughness parameters measured by line profilometer (Hommel Tester T 1000, Hommelwerke

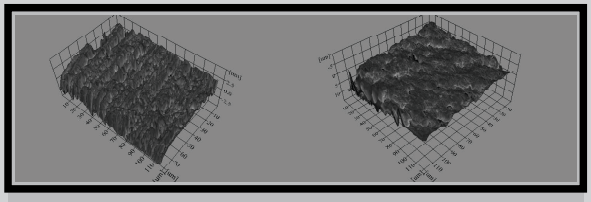

FIG.1. 3D reconstructed surface of ZrN layer deposited on C-C composite by the magnetron sputtering.

GmbH, VS-Schwenningen, Germany) and calculated by Scandium code. We obtained reasonable agreement, even though the length of measurement is substantially different. Than after we studied the dependence of cell spreading on single roughness parameters. For pyrolytic carbon, the optimal value of Ra was obtained, and also the particular correlation between spreading and roughness parameter Rsk was found.

The particular results achieved from the pyrolytic carbon layer shows TABLE 1.

The skewness (Rsk), describing the asymmetry of the

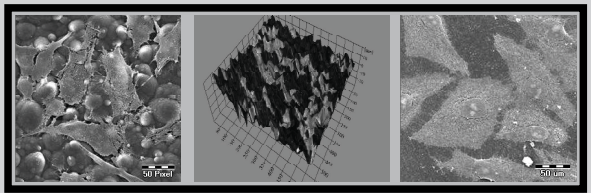

FIG.2. Osteoblast-like cells growth on the PyG layer, SEM; SEM – Scandium.

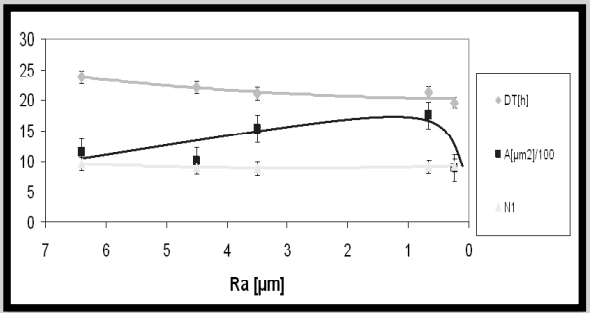

FIG.3. The dependence of biological parameters on the roughness parameter of the surface Ra for the pyrolytic carbon layer

Sample	#3	#4	#5	#6	#7
Cell density after 1 day [10^3/cm^2]	9,6 ±1,0	9,0 ±0,3	8,9 ±1,5	9,1 ±0,7	9,3 ±1,4
A [µm^2]	1155 ±222	1007 ±97	1527 ±425	1759 ±402	895 ±286
DT [h]	23,8 ±1,8	22,1 ±0,6	21,1 ±1,7	21,3 ±0,9	19,6 ±0,4

TABLE 1. Parameters of cell's features after one day cultivation: doubling time DT, number of cells N1 and cell areas A.

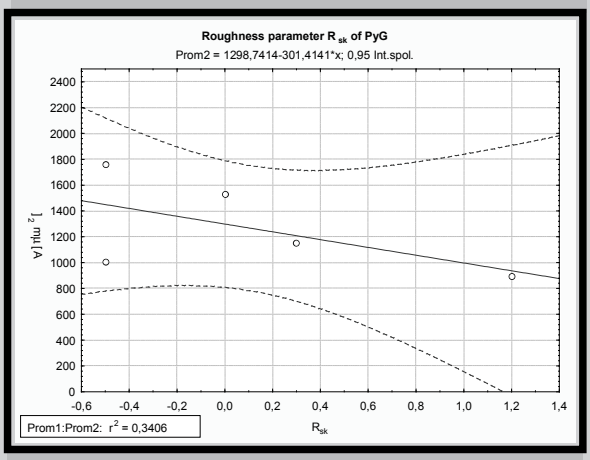

FIG. 4: Dependence of cell areas on Rsk.

profile, showed a tendency to be in negative values for best spread cells.

The next results for other types of surface films will be presented in the contribution.

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BIOLOGICAL EFFECTS OF POLYMERS MODIFIED WITH CARBON NANOTUBES ON HUMAN OSTEOBLAST-LIKE MG 63 CELLS

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Abstract

The tested materials were represented by a pure terpolymer of polytetrafluorethylene-polyvinyldifluoride-polypropylene (PTFE-PVDF-PP), pure polysulphone (PSU) and PSU modified with single- or multi-walled carbon nanotubes in concentrations of 0.5, 1 or 2 wt%. As control samples, a polystyrene cell culture dish and microscopic glass coverslips were used. The number and viability of human osteoblast-like MG 63 cells in cultures on these materials was detected with a Cell Viability Analyzer (Vi-CELL XR, Beckman Coulter) on 1, 3 and 7 days after seeding. On all tested samples, the cell number was similar or lower than that detected on the control polystyrene dishes. The cell viability on day 1 after seeding was relatively low on PTFE-PVDF-PP and some nanotube-containing samples, ranging from 10 to 100 % of living cells, but on day 7 after seeding, it reached at least 90% on all tested samples. The cell spreading area was detected in cells after immunocytochemical staining of β-actin on day 3 after seeding. In nanotube-containing samples, especially those with multi-walled nanotubes, this area was similar or even larger than that on the control materials. The beta-actin cytoskeleton was well developed in cells on all nanotube-containing materials and similar to that in cells on control surfaces. Thus, it can be concluded that nanotube-containing PSU supports the adhesion and growth of osteoblast-like cells and could be used for construction of bone implants in which the anchorage in the surrounding bone tissue is desirable.

Key Words**:** *carbon nanoparticles, carbon-polymer composites, bone cells, adhesion, proliferation, beta-actin*

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Introduction

In recent years, carbon materials, especially the carbon nanotubes, have been considered as very promising materials for biomedical applications including tissue engineering. Therefore, in our experiments, polysulphone was mixed with single- or multi-walled carbon nanotubes, because it is believed that the nanostructure of materials containing nanotubes can resemble the nanoarchitecture of the natural extracellular matrix (ECM). Nanostructured surfaces can