

Development and Characterization of Bio-Tribological, Nano-Multilayer Coatings for Medical Tools Application

L. Major, J. M. Lackner, M. Dyner, B. Major

Abstract—Development of new generation bio-tribological, multilayer coatings opens an avenue for fabrication of future high-tech functional surfaces. In the presented work, nano-composite, Cr/CrN+[Cr/ a-C:H implanted by metallic nanocrystals] multilayer coatings have been developed for surface protection of medical tools. Thin films were fabricated by a hybrid Pulsed Laser Deposition technique. Complex microstructure analysis of nanomultilayer coatings, subjected to mechanical and biological tests, were performed by means of transmission electron microscopy (TEM). Microstructure characterization revealed the layered arrangement of Cr₂₃C₆ nanoparticles in multilayer structure. Influence of deposition conditions on bio-tribological properties of the coatings was studied. The bio-tests were used as a screening tool for the analyzed nanomultilayer coatings before they could be deposited on medical tools. Bio-medical tests were done using fibroblasts. The mechanical properties of the coatings were investigated by means of a ball-on-disc mechanical test. The micro hardness was done using Berkovich indenter. The scratch adhesion test was done using Rockwell indenter. From the bio-tribological point of view, the optimal properties had the C106_1 material.

Keywords—Bio-tribological coatings, cell-material interaction, hybrid PLD, tribology.

I. INTRODUCTION

SURFACE engineering has become an indispensable technology for improving virtually all the properties of solid surface. Almost all types of materials, including metals, ceramics, polymers and composites can be coated with thin films or surface structures of similar or dissimilar materials [1]. In the recent time, much attention in the coatings development is paid to their multi functionality [2]. Single layer coatings are limited in how they can modify the wear resistant properties of surface. The mechanical properties of coatings can be enhanced over wide ranges by adding alternating, or periodic layers. The multilayer structure may result in both possibilities to control the properties of

particular layers (though their chemical composition and microstructure) and also the possibility to control the properties of the whole multilayer coating (through the quantity, thickness and the deposition sequence of component layers [3]-[7]. Introduction of numbers of interfaces parallel to the substrate surface can act to deflect cracks or provide barriers to dislocation motion, increasing the toughness and hardness of the coating. Despite much research on the development of multilayer coatings with superior mechanical and tribological properties, a problem still remains about the mechanisms, operating in the smallest length scale, underlying the mechanical response [8]. The presented paper deals with the protective, bio-tribological coatings elaboration for medical tools what may prolong their life time. Coatings should not only have high wear resistant properties but they should also not be cytotoxic. It is better to adequately protect the surface of metallic tools and properly carry out their sterilization. A number of health care institutions introduced programs for the proper protection and maintenance of instruments. Requirements for tribological, multilayer coatings for medical tools particularly are very high. Detailed biological and tribological study in connection with detailed microstructure analysis is still very rare in literature [9]. The goal of the presented paper was to make complex characterization of coatings for medical tools application by tribological and biological tests and by microstructure characterization.

II. MATERIALS AND METHODS

A hybrid PLD system (Pulsed Laser Deposition + magnetron sputtering) equipped with a high purity chromium (99.9% Cr) and carbon (graphite) targets were used for multilayer coatings deposition. Chromium (Cr) and chromium nitride coatings (CrN) were deposited using Cr target with different gas flow, non-reactive (Argon) and reactive (Nitrogen) respectively. Hydrogenated, amorphous carbon layers were deposited using graphite target in argon or in acetylene low flow. Coatings were composed of two parts: - the inner part (first from the substrate)-multilayer Cr/CrN (with 1:2 ratio); - the outer part (second from the substrate)-multilayer Cr/a-C:H (with different phases ratio). Several types of coatings have been deposited, they differed in the outer part in phases ratio a-C:H to Cr (1:1, 2:1 and 4:1) (Table I).

L. Major is with the Institute of Metallurgy and Materials Science; Polish Academy of Sciences, 25 Reymonta Street, 30-059 Cracow, Poland (corresponding author to provide phone: +48-12-2952800; fax: +48-12-2952804; e-mail: l.major@imim.pl).

J.M. Lackner is with Joanneum Research-Materials-Institute for Surface Technologies and Photonics; Leobner Strasse 94; 8712 Niklasdorf, Austria (e-mail: juergen.lackner@joanneum.at).

M. Dyner is with the CHIRMED-Manufacturer of Surgical and Medical Instruments; 8a Mstowska Street, 42-240 Rudniki, Poland (e-mail: info@chirmed.pl).

B. Major is with the Institute of Metallurgy and Materials Science; Polish Academy of Sciences, 25 Reymonta Street, 30-059 Cracow, Poland (e-mail: b.major@imim.pl).

TABLE I
TYPES OF COATINGS

Sign of the sample	A full description of the coating
C106_1	Multilayer (Cr: CrN= ratio 1:2)- inner part+ Cr interface and multilayer (Cr: a-C:H= ratio 1:1)- outer part
C106_2	Multilayer (Cr: CrN= ratio 1:2)- inner part+ Cr interface and multilayer (Cr: a-C:H= ratio 2:1)- outer part
C106_3	Multilayer (Cr: CrN= ratio 1:2)- inner part+ Cr interface and multilayer (Cr: a-C:H= ratio 4:1)- outer part

Coatings were deposited on the polished austenitic steel samples (DIN 1.4301). Details of the deposition process had been described elsewhere [10]. The cell-material interaction analysis using smooth muscle cells were used as a screening tool for the analyzed nanomultilayer coatings before they could be deposited on medical tools [11]. The biomedical analysis was done in the direct cell deposition. Cells were marked with Phalloidin for cytoskeleton and DAPI for nucleus. A cytotoxicity test determines whether a product or compound would have any toxic effect on living cells. In order to investigate the pathological death, necrosis was analysed. Necrotic cells were marked with PropidinIodit; alive cells were marked with Rhodamin (Mitotrucker). For the alive cells the active mitochondria were marked. The co localization method was applied for the channel separation and the detailed analysis of the percentage of the alive cells. The cells adhesion to the coatings surfaces were observed by confocal microscopy (Zeiss). The mechanical properties of the coatings were investigated by means of a ball-on-disc mechanical test using a 1N of the applied loads for 20000 cycles. The Al₂O₃ (alumina) ball with 6mm diameter was used for the test. The microhardness was done using Berkovich indenter. The scratch adhesion test was done using Rockwell indenter. The microstructure of the coatings was studied using transmission (TEM) (TECNAI G² F20 FEG (200 kV)) electron microscope, which allows microstructure observation in the smallest scale, especially, in the high resolution mode (HRTEM).

III. RESULTS AND DISCUSSION

Structure, microstructure, and nanostructure of surface treatments are critical aspects for surface engineering [12]. In the first part of the paper microstructure of bio-tribological protective coatings has been described. Paper deals with coatings for medical tools, this is why the second part is connected with biomedical analysis, while the last part with mechanical tests.

A. Microstructure Characterization Using Transmission Electron Microscopy (TEM) Technique

Microstructure of as-deposited coatings was characterized on cross-section (Fig. 1).

Coatings contained two parts. The inner part, first from metallic substrate, formed Cr/CrN (chromium/ chromium nitride) multilayer. Its roles were residual stress reduction as well as coating adhesion enhancement to substrate. Generally the inner part of the coating is a structural link with stress compensation ability to the substrate material [13]. The CrN

and Cr lattice parameters, allow a cube-on-cube close to epitaxial growth with a low mismatch (1.6%) (Fig. 2).

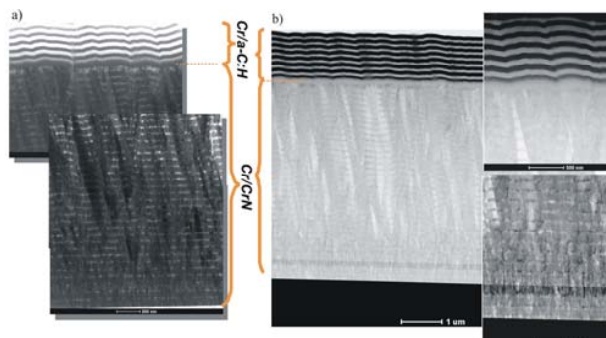


Fig. 1 Microstructure characterization of the Cr/CrN+Cr/a-C:H (the Cr to a-C:H ratio= 1:1) multilayer coating done by transmission electron microscopy technique; (a) image in TEM bright field mode (diffraction contrast); (b) image in STEM mode (contrast dependent on the atomic number Z)

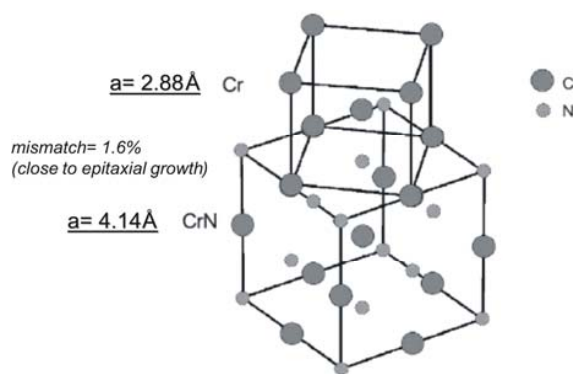


Fig. 2 The scheme of the Cr and CrN cell growth assembling [14]

The same diffraction contrast went through interfaces, what confirmed crystallographic dependence in between Cr and CrN phases. It was well seen on the TEM BF image (Fig. 1 (a)) [14]. The second, outer part of the coating formed multilayer structure built of Cr/ a-C:H multilayer (chromium/ amorphous carbon). They were characterized by low coefficient of friction and high wear resistance (Fig. 3).

Generally the outer part of the coating acts as a diffusion barrier with additional self-healing capability [15]. Quantitative chemical analysis has been performed along the line marked at Fig. 3 (a). The distribution of selected elements was presented at the diagram (Fig. 3 (b)). The chromium content changed subsequently with carbon. It has been noticed that the closer to the interface with the inner part of coating (Cr/CrN), the higher Cr content also in carbon layers. It is well known that hydrogenated amorphous carbon (a-C:H) coatings have low friction coefficient and low specific wear rates. Thus, the a-C:H coatings are very promising tribo-materials. However, poor adhesion strength to substrate, high residual stress and weak thermal stability would limit the application of a-C:H coatings [16], [17]. In the current work Cr nanograins have been utilized to modify structure and properties of

amorphous carbon. Cr nanograins were inserted into carbon structure in the form of nanolayers as it has been well presented on the image (Fig. 4).

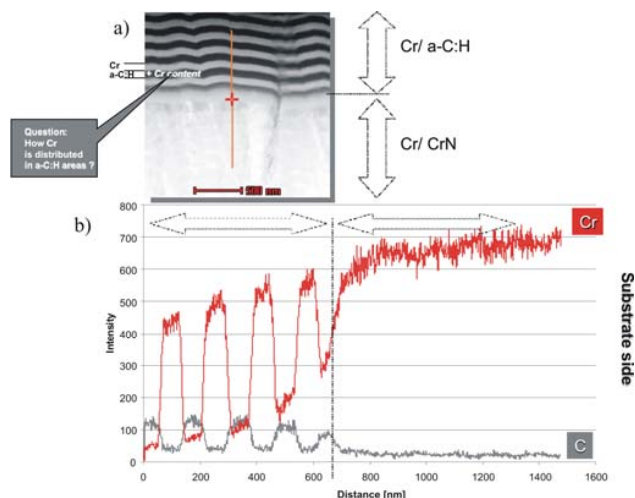


Fig. 3 Qualitative chemical analysis of the coating; (a) STEM image (in contrast dependent on the atomic number Z); (b) the diagram of selected elements distribution in the coating

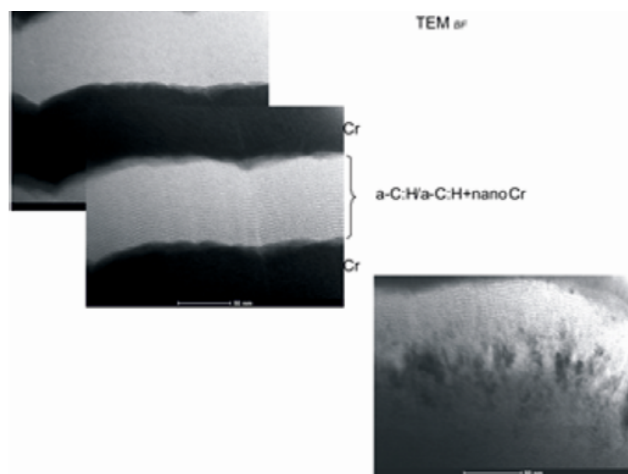


Fig. 4 Microstructure characterization of the outer part of the coating (Cr/a-C:H multilayer), revealing the nanomultilayer in a-C:H structure. Analysis performed by transmission electron microscopy technique in bright field mode

The multilayer structure of nanocrystallites in a-C:H structure was formed. Thus, the outer part of coating was built of multilayer structure of nanocrystals in a-C:H/Cr multilayer structure. High resolution analysis (HRTEM) not only confirmed the layered settings of Cr nanograins, but also allowed to perform their phase analysis. Experiments indicated that chromium which was inserted into amorphous carbon structure reacted with carbon forming chromium carbides Cr_{23}C_6 nanocrystals (Fig. 5).

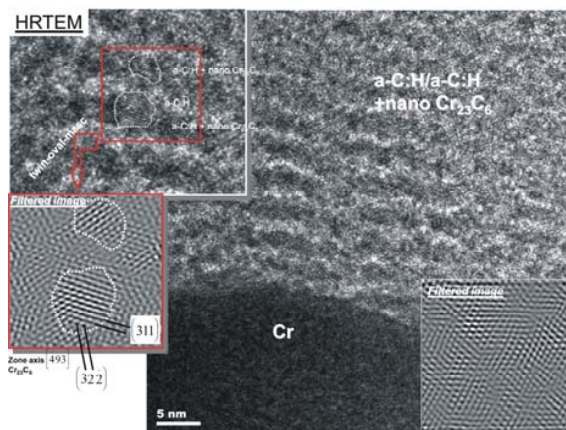


Fig. 5 The high resolution TEM characterization of the nanomultilayer structure in the Cr/a-C:H multilayer structure

This is a stable form of chromium carbide [18]. The HRTEM analysis allows performing phase analysis, and also indicated that thickness of individual Cr_{23}C_6 layers was equal to the diameter of nanocrystals ($\sim 3\text{nm}$).

B. Biological Test: The Cell-Material Interaction

The analysis of cell-material interactions was made using smooth muscle cells. For each material the adhered cells were counted (Fig. 6).

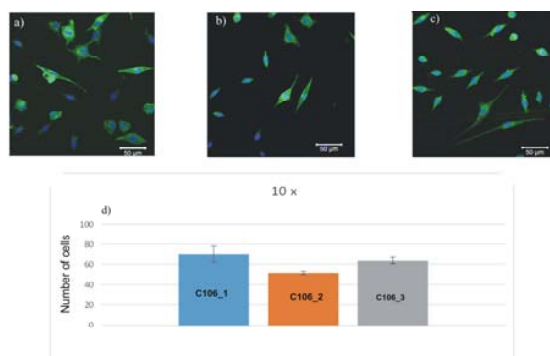


Fig. 6 Cell-material interaction done using fibroblasts; (a) topography image of the multilayer C106_1- [(Cr:CrN= 1:2)- inner part + Cr interface and multilayer (Cr: a-C:H= ratio 1:1)- outer part]; (b) topography image of the multilayer C106_2- [(Cr:CrN= 1:2) inner part + Cr interface and multilayer (Cr: a-C:H= ratio 2:1)- outer part]; (c) topography image of the multilayer C106_3- [(Cr:CrN= 1:2) inner part + Cr interface and multilayer (Cr: a-C:H= ratio 4:1)- outer part]; (d) diagram summing up results obtained in the cell- material interaction

The highest cells adhesion was found for the coating marked as C106_1- [(Cr:CrN= 1:2)- inner part + Cr interface and multilayer (Cr: a-C:H= ratio 1:1)- outer part]. The lowest biological properties were noticed for the coating indicated as a C106_2- [(Cr:CrN= 1:2)- inner part + Cr interface and multilayer (Cr: a-C:H= ratio 2:1)- outer part]. The goal of this part of the work was to judge cell viability on the surfaces which were dedicated to surgical tools. Thus the effectiveness

of the cell monolayer formation was considered as a potential probability of the investigated surface acceptance by cells.

Generally chromium is toxic, however in case of biotribological application for medical tools like surgical tweezers, where not only biological properties are taken under account but also mechanical one, it is accepted [19]. The suppressing issue was relatively good cells adhesion for coating with the higher Cr content in the outer part.

C. Biological Test: Cytotoxicity

The viability was analyzed using fibroblasts (Fig. 7).

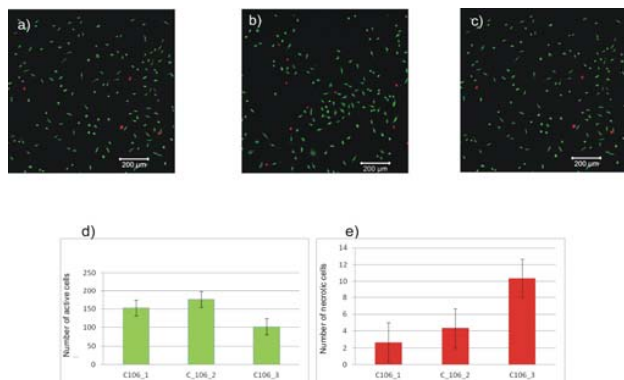


Fig. 7 Cytotoxicity results; (a) topography image of the multilayer C106_1- [(Cr:CrN= 1:2)- inner part + Cr interface and multilayer (Cr: a-C:H= ratio 1:1)- outer part]; (b) topography image of the multilayer C106_2- [(Cr:CrN= 1:2)- inner part + Cr interface and multilayer (Cr: a-C:H= ratio 2:1)- outer part]; (c) topography image of the multilayer C106_3- [(Cr:CrN= 1:2)- inner part + Cr interface and multilayer (Cr: a-C:H= ratio 4:1)- outer part]; (d) diagram showing number of active cells depending on the type of analyzed coating; (e) diagram showing number of necrotic cells depending on the type of analyzed coating

The highest number of necrotic cells was noticed in for the C106_3 coating, while the lowest for the C106_1. The best properties was found for the coating with the lowest chromium content in the outer part what was expected due to the relatively toxic properties of chromium.

D. Tribological Test: Indentation Test

Indentation tests were performed at a load of 2 and 5 mN. For the tests the diamond indenter with Berkovich geometry was used. Results of nanoindentation have been presented in Table II, and at Fig. 8.

TABLE II
RESULTS OF MICRO-HARDNESS ANALYSIS

Type of coating	2 mN		5 mN	
	Hardness [MPa]	Elasticity modulus [GPa]	Hardness [MPa]	Elasticity modulus [GPa]
C106_1	9.6± 0.9	142± 11	9.7± 0.5	148± 6
C106_2	10.6± 0.8	154± 12	10.2± 0.8	172± 9
C106_3	11.6± 1.1	175± 13	10.9± 0.9	184± 6

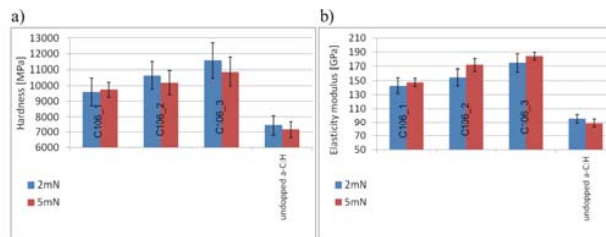


Fig. 8 Results of micro-hardness; (a) micro-hardness values; (b) elasticity modulus

The highest hardness and elastic modulus were found for the C106_3-[(Cr:CrN= 1:2)- inner part + Cr interface and multilayer (Cr: a-C:H= ratio 4:1)- outer part] coating.

E. Tribological Test: Scratch Test

The tests were conducted using a Rockwell C indenter. Surface images with characteristic loads under which cohesive (L_{C1}) and adhesive (L_{C2}) cracks appeared have been presented at Fig. 9.

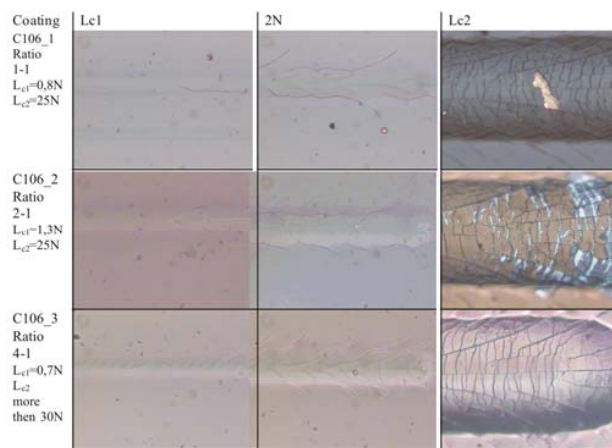


Fig. 9 Results of scratch adhesion test of the analyzed, multilayer coatings

For testing coatings the first cohesive cracks were formed under the of 0.7-0.8 N load excluding the C106_2 coating, which cracked at a 1.3 N load. The critical LC2 (presence of adhesive cracking) was in the range of 25N for C106_1 and C106_2 coating. Adhesive cracking for C106_3 coating were not observed even at the maximum load (30N), which was applied. Thus, the best coating adhesion was found for the C106_3-[(Cr:CrN= 1:2)- inner part + Cr interface and multilayer (Cr: a-C:H= ratio 4:1)- outer part] coating. The resultant Cr: a-C:H part of the coating, generally was marked by high crack resistance and high adhesion in comparison with pure a-C:H layer, what was also noticed by other researchers [20]. The presented results indicated that the higher Cr content in a-C:H/ Cr multilayer, the better mechanical properties.

F. Tribological Test: Wear Test

The tests were performed in the ball-on-disk contact. The number of cycles to coatings degradation has been presented in Fig. 10.

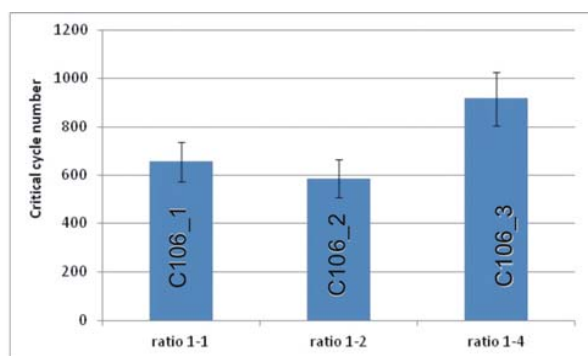


Fig. 10 Results of wear test of the analyzed multilayer coatings

The highest wear resistance was indicated for C106_3 - [(Cr:CrN= 1:2)- inner part + Cr interface and multilayer (Cr: a-C:H= ratio 4:1)- outer part] coating. It confirmed the earlier presented mechanical results. The higher Cr content in the a-C:H/Cr multilayer (the outer part) the better the properties.

G. APPLICATION

The novel type of coatings deposition technique linking pulsed laser deposition with magnetron sputtering (hybrid technique), supported by detailed and complex characterization of deposited coatings can lead to elaboration of novel type of films for very sophisticated application like metallic medical tools i.e. surgical tweezers. Manufacturer of surgical and medical instruments CHIRMED in Rudniki is a Polish producer of medical and surgical instruments. The main goal of the performed experiment was to elaborate and find optimum deposition parameters in coatings production for medical tools (Fig. 11).



Fig. 11 An example of tools produced by the Chirmed company where coatings may find an application

IV. CONCLUSION

- Microstructure characterization revealed the layered arrangement of Cr_{23}C_6 nanoparticles in multilayer structure
- The biological test, particularly cytotoxicity test which was done using eukaryotic cells showed that the material signed by the C106_3 number was the most toxic
- The tribological test, especially microhardness done using Berkovich indenter, showed the highest hardness for the C106_3 material
- The scratch adhesion test was done using Rockwell indenter. The test revealed that the best adhesion was

found for the C106_3 material. The C106_1 had slightly lower properties (acceptable). Poor adhesion was found for the C106_2 material.

- The highest wear resistance was found for C106_3 material. The lowest properties were found for C106_2
- From the bio-tribological point of view, the optimal properties had the C106_1 material
- Described coatings may find an application as protective films for surgical tools

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Research Project (National Science Centre): Bio-mechanical and microstructure analysis of multilayer nano-composite, protective coatings for metallic substrates for tissue interaction. Number: 2012/07/B/ST8/0339

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