

Correlative Microscopy in Life and Materials Science

MIM.4.P061

Interactions of bacteria and yeast in biofilms: transmission and scanning electron microscopic analysis

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Keyword: biofilm, bacteria, yeast, transmission and scanning electron microscopy

The processes of the shaping biofilms of bacteria and yeast as cultures on surfaces of metals, mucous surface of the cavities of the organisms and in external ambience are among actual problems of microbial ecology those having an important medico-biological importance; they are not more valuable for new biotechnology [1-3]. In the present work ultra-structural particularities of biofilms from some bacteria and yeast have been studied in order to install the intercellular contacts between cells *in vitro* and *in vivo*.

The main object of the study were bacteria referred to different taxonomic groups as *Escherichia coli* serogroup O124, *Shigella flexneri* 130, *Salmonella typhimurium* 546, *Staphylococcus aureus* 906, *Salmonella enterica* ATCC700931, *Lactobacillus Oenococcus oeni* as well as yeast *Candida guilliermondii* NP-4. Bacteria and yeast were cultivated on the ceramic plate porous of Zirconia from samples[4] of the mucous gastro-duodenal intestine tract sucked by *Helicobacter pylori* as well as of experimental models animal. For study of the bacterial surface structure bacteria used the electron microscopy with negative staining and ultrathin sections (TEM) and scanning (SEM) were used. The localization of mucopolysaccharides was determined by the method Luft. The computer analysis of the scenes was organized using the programs "Video-test; Struktura-5; Nanotechnology" and "Morphology".

Ultrastructure analysis of bacterial colonies *in vitro* showed a typical fine construction for gram-negative and gram-positive bacteria. The clarification of the structured particularities of the intercellular contacts zones is realized with more detailed presentation of the surface structures and cellular wall. The study of the surface structures of gram-negative bacteria has revealed the different forms of intercellular contacts: for enteropathogenic *E. coli* with adhesive characteristics, fimbriae formation (Fig.1) might be taken part in delivering of plasmid and in fastening to the other substrate. By means of computer program, reconstruction of stereo-metrical orientation of the saw was managed. The sizes of the saw varied within 100 to 200 μm , but diameter was 8 nm. The other varieties of intercellular contacts for bacteria revealed itself in the manner of thick adhesion to be taken to cellular wall beside of *Shigella* as well as thick contact peptidoglycan of microcapsule. TEM research the mutual relation salmonella with the lactobacillus -probiotics has revealed close intercellular contacts and changes surfaces of a cellular wall of salmonella (fig.2).

TEM and SEM of intact yeast cultures *C. guilliermondii* NP-4 have shown typical ultrastructure images for yeast cells and visualizations of intercellular contacts in biofilms (Fig. 3). Measurement of *Candida guilliermondii* by means of the program "Morphology" has shown: diameter = 1.15-2.71 μm , length = 3.22 μm , buds = 0.318 μm . Adhesion of yeast on the surface of plate porous of Zirconia and multiform division of cells and multitude of buds (Fig. 4).

Thereby, studied by electronic microscope as well as by computer picture analysis ultrastructure peculiarities of cellular interactions of different gram-positive and gram-negative bacteria and yeast in biofilms are being formed as fimbria, close contact, membranes adhesion and cellular plexus [1-3]. This revealed ultrastructure particularities of cellular contacts to consider that the process of the shaping biofilm from bacteria and yeast depends from ultrastructure architectonic of microbial surfaces and morphological properties of substrates.

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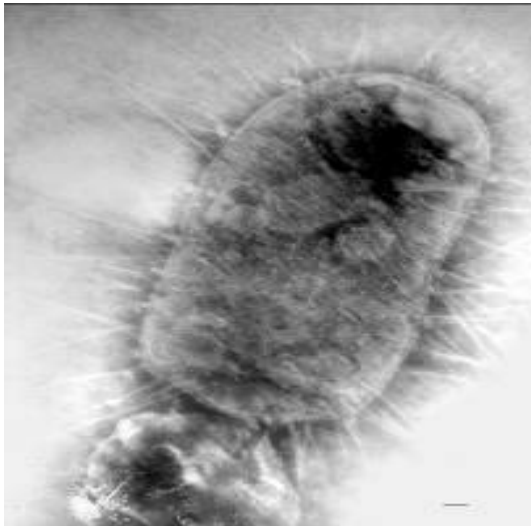


Figure 1. TEM of *E. coli* serogroup O124 (negative staining). Bar: 0,3 μm

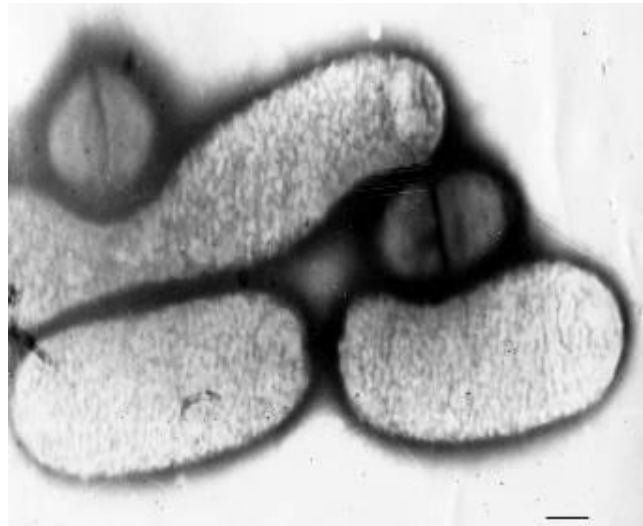


Figure 2. SEM of biofilm of *C. guilliermondii* NP-4. Bar: 0,3 μm

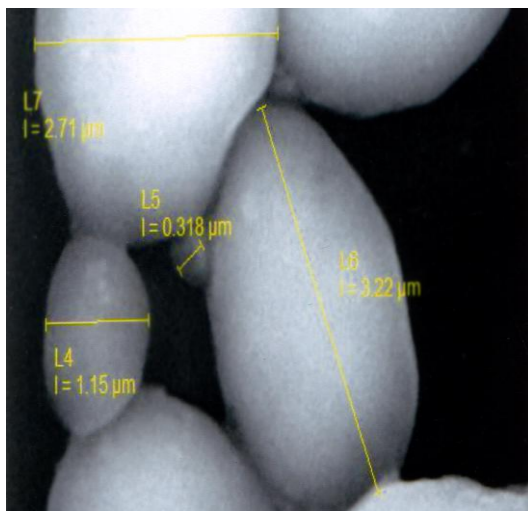


Figure 3. SEM of biofilm of *C. guilliermondii* NP-4.

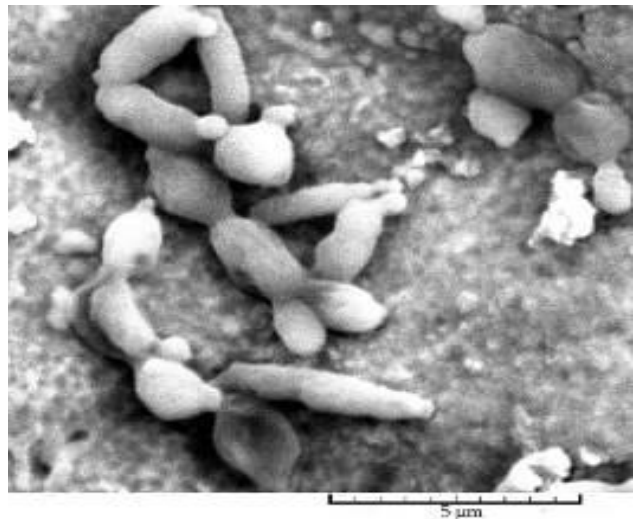


Figure 4. SEM of biofilm of *C. guilliermondii* NP-4. Formation of biofilm on ceramic porous plate of Zirconia.