

Changes of *Streptomyces violaceus* cell wall ultra structure during culture development

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Changes of *Streptomyces violaceus* cell wall ultra structure during culture development : Change in chemical content of cell wall of *Streptomyces violaceus* has been studied in the dynamics of culture growth. It has been established that according to growth phases a mass of cell wall of actinomycetes changes. It is maximal in logarithmic phase of growth and is minimal in stationary phase. An insignificant increase of cell wall mass was observed in the phase of dying. Qualitative changes of its constituents- Peptidoglycan, Teichoic acid and their monomers, amino acids and monosaccharide were not found. But quantitative ratios of separate components were changes.

Key words: Peptidoglycan, Teichoic acid, Amino acid, Monosaccharide.

INTRODUCTION

Among diagnostic features used in the systematization of actinomycetes, ratio of chemical composition of cell wall appears to be very important, in particular lipid, amino acidic and monosaccharide content [1]. Peptidoglycan and Teichoic acid appearing main components of prokaryotic cell wall are complex composition, containing amino acids and they may be used in the systematization of actinomycetes as one of the additional chemotaxonomic signs.

MATERIALS AND METHODS

Streptomyces violaceus obtained from the collection of microorganisms of Microbiology Department of Tbilisi Botanical Garden and Institute of Botany was used as the subject of the investigation.

The culture was grown on Krasilnikov synthetic medium. Morphology was studied using method of B. Krasilnikov and V. Kuznetsov [1, 2]. Cell wall was obtained according to L. N. Robson and Baddiley [3].

Peptidoglycan and Teichoic acid were obtained using the methods of Streshinskaia [3, 4]. Qualitative and quantitative analysis of amino acids and monosaccharide was carried out using thin-layer chromatography and densitometry method [5].

DISCUSSION

Streptomyces violaceus (synthetic medium of Krasilnikov) creates colonies with rough surfaces having 3-4 mm diameter, as well as with weakly developed grayish-violet colored aerial mycelium. Substrate mycelium is of violet color. Culture creates hard complex of pigments (pink, violet and red with the different specter of absorption I -610-585 mc, II -560-540-mc, III -510-499 mc). Dynamic of cultivation is determined; complete growth of culture is 150 h, logarithmic phase 0-48 h, exponential phase 48-96 h, stationary phase 96-114 h. Then 118 h later the phase of dying occurs. (Fig. 1).

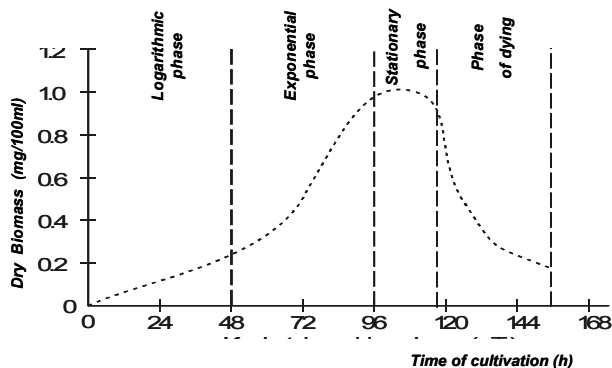


Fig. 1. Dynamics of *Streptomyces violaceus* growth.

As is shown from the table 1, cell wall of *Streptomyces violaceus* consists 9.04-13.05% of dry biomass. During growth and development of the culture, its mass changes. It is maximal in logarithmic phase and minimal –in stationary phase. In the phase of dying cell wall mass insignificantly increases. The amount of main component determining cell wall mass- Peptidoglycan varies within the limits of 63.15-67.5%, while the amount of Teichoic acid-approximately within the limits of 14.2-23.5% (in 1g of dry cell wall). Change in Peptidoglycan mass depends on the change of cell wall mass. It is maximal in logarithmic phase and minimal –in stationary phase. The mass of Teichoic acid reduces from logarithmic to the phase of dying.

Table 1.
Quantitative changes in *Streptomyces violaceus* cell wall and its main components in dynamics of culture growth.

Phases of growth	Mass of cell wall and its components, mg/g		
	Cell wall mg in 1 g of dry biomass	Peptidoglycan mg in 1 g of cell wall	Teichoic acid mg in 1 g of cell wall
logarithmic	130.57	675.0	235.0
Exponential	100.67	658.0	165.0
Stationary	90.4	631.5	150.0
Phase of dying	110.0	650.0	142.0

Quantitative and qualitative analysis of *Actinomyces violaceus* Peptidoglycane was performed. The results are given in Table 2.

As is shown from Table 2, main skeleton of *Streptomyces violaceus* cell wall Peptidoglycan to be a glycan fraction, consisting 59.1-65.6%, while peptide fraction consists 20.01-28.08%. The amount of monosaccharide is maximal in logarithmic phase, minimal –in stationary phase. In the phase of dying in peptide fraction of Peptidoglycan the amount of amino acids increases.

Table 2.

Phases of growth	Mass of chemical components in 1 g of Peptidoglycan (%)	
	Monosaccharide	Amino acids
logarithmic	65.6	28.08
Exponential	64.01	26.57
Stationary	60.8	24.08
Phase of dying	59.1	20.01

Results of qualitative analysis of Peptidoglycan are given in fig. 2 and fig. 3. It was shown that qualitative ratio of amino acids in Peptidoglycan of investigated culture changes according to growth phases.

Glycan fraction of Peptidoglycan is presented by two monosaccharide, quantitative ratio of which appears to be changeable in dynamics of culture growth (Fig. 3).

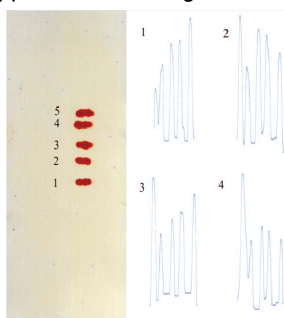


Fig. 2. Amino acid content of Peptidoglycan of *Streptomyces violaceus*

1. Diaminopimelic acid; 2. Muramic acid; 3. Alanine; 4. Lysine; 5. Glycine.

Densitograms: 1. Logarithmic phase; 2. Exponential phase; 3. Stationary phase; 4. Phase of dying.

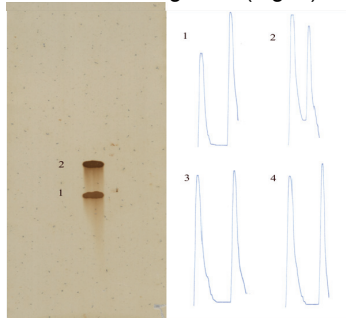


Fig. 3. Monosaccharide content of Peptidoglycan of *Streptomyces violaceus*

1. Glucosamine; 2. Galactose.

Densitograms: 1. Logarithmic phase; 2. Exponential phase; 3. Stationary phase; 4. Phase of dying.

Analysis of our investigation has shown that Ribite-Teichoic acid of cell wall of *Streptomyces violaceus* appears to be quantitatively changeable value and depends on conditions of development of the culture.



Fig. 4. Amino acid content of Teichoic acid of *Streptomyces violaceus* Alanine

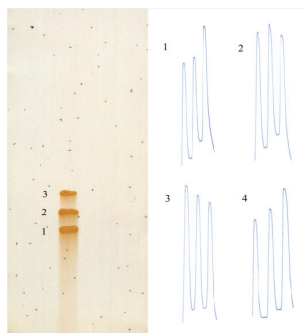


Fig. 5. Monosaccharide content of Teichoic acid of *Streptomyces violaceus*

1. Glucosamine; 2. Ribose; 3. Glucose.

Densitograms: 1. Logarithmic phase; 2. Exponential phase; 3. Stationary phase; 4. Phase of dying.

As is shown in Fig. 4., 5, Alanine is identified in the content of Teichoic acid of *Streptomyces violaceus*, while from monosaccharide- Glucose, Ribose and Glucosamine.

CONCLUSION

So, *Streptomyces violaceus* has a cell wall containing Peptidoglycane and Ribite-Teichoic acid that is characteristic for typical gram-positive prokaryotes. Quantitative ratio of monomers of Peptidoglycane and Teichoic acid-amino acids and monosaccharide- changes according to growth phases, but change in their qualitative content is not Proceeding from this, it may be used as one of the additional chemotaxonomic signs in the systematization of Actinomycetes.

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