

Specific anthrax bacteriophages as a factor for selection of subcultures with different phenotypic and genetic characteristics out of populations of *Bacillus anthracis* strains

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Motivation and Aim: The aim of the work was to study population composition of two virulent strains of the causative agent of anthrax by the factor of phage resistance to specific bacteriophages, to select phage resistant subcultures, and to study their properties comprehensively.

Methods and Algorithms: We used virulent strains *B. anthracis* 1 (CO) and 81/1 which were isolated from pathological material. Concentrations of phage corpuscles in the preparations of bacteriophages Gamma A-26, BA-9, K-VIEV were, correspondingly, 8×10^9 , 4×10^8 and 2×10^8 per 1 ml. Population composition of *B. anthracis* strains by their resistance to bacteriophages was studied by the method [1]. Subcultures of *B. anthracis* 1 (CO) strain were studied according to the basic identification tests and additional methods to study *B. anthracis* cultures. Six chromosomal loci were used to characterize strains by VNTR-loci [2].

Results: Not a single colony grew on plates of both strains treated with bacteriophage Gamma A-26. Plates treated with bacteriophage K-VIEV showed a 2.9 and a 4.8 % growth of colonies of the strains *B. anthracis* 1 (CO) and 81/1 respectively. Plates treated with phage “BA-9” showed a 10.9 % growth of colonies of the strain *B. anthracis* 1 (CO) and a 17.3 % growth of colonies of the strain *B. anthracis* 81/1, correspondingly. Distribution of the 22 variants of each strain into groups differing by their sensitivity to various bacteriophages was as follows. The retest showed that in both strains 16.7 % of variants separated on the basis of their resistance to bacteriophage BA-9 were sensitive to all the three bacteriophages. In variants of the strain *B. anthracis* 81/1 which were selected from the plates treated with bacteriophage BA-9 such variants made up 20 %, and in *B. anthracis* 1 (CO) – 10 %. Variants, resistant to the action of bacteriophage Gamma A-26, were found among cultures selected on the basis of their resistance to the other bacteriophages. Among 20 phage resistant variants of the strain *B. anthracis* 1 (CO) we found variants which were atypical in capsule formation, toxin production, nutritional requirements, protease, lecithinase, and hemolysin activities. Genetic studies revealed three variants of the plasmid structure, and four MLVA-genotypes.

Conclusion: A considerable variety of phenotypic and genetic properties among phage resistant subcultures of the strain *B. anthracis* 1 (CO) can testify to complex change of some of them which makes specific anthrax bacteriophages an effective factor for selection of atypical in many properties cultural variants out of populations of strains.

References

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