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Bacteriophage against *Streptococcus Equi* - A Step toward Overcoming Resistance in the Treatment of Strangles

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Abstract **-** *Streptococcus equi* subsp. *equi* (*S. equi*) is a host-specific pathogen that causes strangles, an infectious disease in horses. Rising antibiotic resistance among clinical isolates necessitates alternative therapeutic strategies. This study reports the isolation and characterization of lytic bacteriophage BV-0002, active against multidrug-resistant *S. equi*. The phage exhibited high specificity and strong lytic activity both in vitro and in vivo, suggesting its potential as a therapeutic agent in veterinary medicine.

Keywords**:** Bacteriophage, *Streptococcus equi*, equine strangles, antibiotic resistance, lytic activity, phylogenetic analysis, therapeutic efficacy

1. Introduction

It is expected that authors will submit carefully written and proofread material. Careful checking for spelling and grammatical errors should be performed. The number of pages of the paper should be from 4 to 8.

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Streptococcus equi subsp. *equi* is a major equine pathogen responsible for strangles, a disease marked by lymphadenopathy and systemic inflammation, leading to significant economic losses [1], [2]. Genomic analysis confirms its divergence from the zoonotic *Streptococcus zooepidemicus* via acquisition of prophages and integrative elements, and loss of metabolic functions [3]-[5]. In Kazakhstan, both sporadic and outbreak cases are frequent, especially in foals [6], [7]. Standard treatment involves β-lactam antibiotics, but increasing multidrug resistance (MDR) undermines effectiveness [8]-[11]. Phage therapy, due to its specificity and efficacy against resistant bacteria, is a promising alternative [12]-[14].

2.Materials and Methods

Environmental and clinical samples (soil, water, nasal discharge) were collected from horse farms in Almaty *Region*, Kazakhstan. Six bacteriophages were isolated using standard enrichment and purification; BV-0002 was selected based on its superior activity.

Phage lytic spectrum was tested on clinical *S. equi* isolates, including MDR strains, using the Appelman method for qualitative analysis and the Gratia method for quantitative titration. Specificity was assessed against other *Streptococcus* spp.

Capsid proteins were analyzed by SDS-PAGE following ultracentrifugation in cesium chloride and sucrose gradients, *as* well as ultrafiltration with lyophilization. Protein masses were matched with NCBI Protein data. Electron microscopy was used for morphological examination.

In *vivo*, 12 foals were intranasally infected with virulent *S. equi*. Six were treated with BV-0002 (intranasally and via abscess infiltration); six received placebo. Clinical parameters and bacterial clearance were recorded.

3.Results

BV-0002 lysed all tested *S. equi* isolates, including MDR variants. No lytic activity was detected against other *Streptococcus* species.

Sucrose gradient ultracentrifugation provided consistent capsid protein profiles (17, 19, 25, 26, 30, 34, and 40 kDa). A *consistent* 38–40 kDa hyaluronidase-like protein suggests a role in capsule penetration.

Electron microscopy confirmed Myoviridae morphology: ~70 nm head and ~190 nm contractile tail.

In *vivo*, BV-0002 reduced fever, decreased abscesses, and improved clinical signs within 48 hours. Recovery began by *day* 4; by day 10, all treated foals recovered without antibiotics. No side effects or reinfections occurred.

4.Conclusion

BV-0002 demonstrated potent and specific lytic activity against MDR *S. equi* and showed safety and therapeutic efficacy in a foal model. Its characteristics support its potential for veterinary phage therapy.

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