

# PIONEER BIOPROSPECTION OF ANTI-KLEBSIELLA PNEUMONIAE BACTERIOPHAGES OBTAINED FROM SEWAGE SAMPLES FROM VARIOUS REGIONS IN NORTHERN BRAZIL

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## RESUMO

The indiscriminate use of antibiotics has favored the emergence of strains of *Klebsiella pneumoniae* resistant to several antimicrobials. This pathogen is a frequent cause of sepsis and pneumonia. It is estimated that more than 10 million people will die by 2050 from multidrug-resistant bacteria, making them more deadly than cancer. In this sense, the use of bacteriophages (phages), viruses that infect bacteria, becomes an alternative to antibiotic therapy. Considering that the Amazon region comprises the richest biodiversity on the planet, the northern region becomes an extremely promising source of discoveries of new phages. Therefore, the objective of this study was to isolate, for the first time, anti-*K. pneumoniae* phages in sewage samples from several regions of northern Brazil with a view to using them in phage therapy. *K. pneumoniae* strains (KH1, NEWP 0083, ATCC 13883, ATCC BAA1705) were cultivated in 5 mL of Luria Bertani (LB) broth and incubated in an oven for 48 hours at 37 °C. Sewage collections were carried out in Santarém-PA, Macapá-AP, Rio Branco-AC, Belém-PA, Manaus-AM, Oriximiná-PA, Monte Alegre-PA and Porto Velho-RO. A total of 50 mL of each sewage sample was centrifuged at 4000×g for 30 minutes and the supernatant was filtered with a 0.45 µm membrane. The filtrates underwent phage enrichment, where they were mixed with equal amounts of LB medium and 4 mL of bacterial culture. After incubation for 18 hours at 37 °C, 10 mL was centrifuged and the supernatant was filtered again. Filtrates were tested for the presence of anti-*K. pneumoniae* lytic phages using the double-layer agar method. After observing the lysis plates, the phages were purified by three rounds of isolation using the double-layer agar method. Promising phages for phage therapy are those that are capable of lysing the largest number of strains within the same bacterial species, which is why the host range test was performed. To date, a total of 50 bacteriophages have been isolated and purified. Interestingly, we observed that of the 50 phage isolates, four were able to lyse all four strains of *K. pneumoniae*. Other analyzes are still being carried out: susceptibility of planktonic cells and biofilm to phages, genomic sequencing, phage cytotoxicity tests for human cells and infection and treatment of mice with *K. pneumoniae* and phages, respectively. Our initial results point to a new alternative for combating *Klebsiella pneumoniae* infection. **Funding source:** process 2022/1437972 FAPESPA/CNPq and UFOPA

**PALAVRAS-CHAVE:** phages, sewage, phage therapy Amazon region, kpneumoniae

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