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CkP1 is a novel S16-like broad-host-range myovirus that recognizes *Citrobacter koseri* lipopolysaccharide through its long tail fibres

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Citrobacter spp. is an emerging Gram-negative bacterial pathogen. The genus is currently divided in several species, being *Citrobacter koseri* (formerly named *Citrobacter diversus*) one of the most common isolated species in human clinical specimens. *C. koseri* can cause localized (urinary track, wounds infection, pneumonia, meningitis) and systemic life-threatening diseases (bacteremia, septicemia), being neonates, immunocompromised and elderly people, groups with increased risk of infection. The prevalence rate of *Citrobacter* infections in humans, range between 3–6 % among all *Enterobacteriaceae*, but mortality rate in cases of bacteremia reaches 56 %.

We have isolated a novel S16-like *C. koseri* myovirus vB_CkM_CkP1 (CKP1). CkP1 genome has a length of 168,463 bp and contains 261 coding sequences, encoding more than 100 proteins with unknown function. About 60% of the encoded proteins are shared with the *Salmonella* phage S16. CkP1 displays an extremely broad host range, infecting *C. koseri*, but not other species. The long tail fiber (gp267) was fused to a green fluorescence protein (GFP) and co-expressed with and without the gp268 chaperone as a bicistronic transcript. Functional analysis demonstrated that the tail fiber binds to, and is able to decorate, *C. koseri* cells, being equally broad and specific towards this species as observed with the phage. Surface plasmon resonance showed that the binding affinity of the tail fiber is in the nanomolar range, presenting similar values with and without the chaperone. Both phage and the tail fiber specifically bind to bacterial cells by the lipopolysaccharide polymer. This was demonstrated by creating deletion mutants, whole genome sequencing of resistant variants and complementation of insensitive CkP1 bacterial strains. We further demonstrate that CkP1 is highly stable towards different environmental conditions and able to control *C. koseri* cells in urine samples. In summary, CkP1 has high potential in the control and detection of *C. koseri*.