Extended Abstract Insights in Immunology 2018
Vol.1 No. 2

# Evalvuation of efficacy of saclac globule formulation of indigenous bacteriophages against multidrug resistant bacterial pathogens in water microcosm

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**Abstract**

Water is an important fundamental requirement of the mankind. Water covers nearly 75 % of the earth surface. Water resources are becoming limited due to the contamination problems. Unfortunately, life- threatening bacterial pathogens contaminate waters that we drink thus can be responsible for significant health risks. Waterborne bacterial pathogens are now a days developing resistance to chlorine that is used for the disinfection purpose. They are also developing resistance to antibiotics. To overcome such problems, associated with the use of chemical disinfectant like chlorine, an alternative strategy could be use of Lytic bacteriophages as biological disinfectant. Bacteriophages are the viruses of prokaryotes that can either instantly kill a bacterial cell or integrate their genome into the host chromosome. In this study, potent lytic phages were isolated from the natural resources. They were characterized and their infectivity abilities against specific pathogens were checked. Once the phages are produced in large scale, there is a need of specific carriers to maintain their stability. Therefore, there is a need of acceptable, eco-friendly formulations that will increase the longevity of phage particles. In order to achieve better efficacy of phage formulations in the bioremediation of water bodies', selection of the carrier to make good formulation is an important and mandatory step. Saclac globule (it's a globule made up of goat milk and sugar) formulations using efficient phages were prepared and their shelf-life was checked at 4ºC and 30 ºC. As theses globules contain goat milk sugar, this component supports the pathogen to remain in the metabolically active state. Bacteria in this state will be killed rapidly by phages, secondly, Saclac globules are water soluble that get dissolved rapidly as compared to other carriers used in the formulation. In this study, capability of the bacteriophages, viz., ϕSPB,BVPaP-3 and KPP was evaluated in the bioremediation of water microcosms prepared using the Pavana river water and swimming pool water spiked independently with the bacterial pathogens, viz., *Salmonella enterica* serovar Paratyphi B, *Pseudomonas aeruginosa* and *Klebsiella pneuminiae*. In this study, water microcosms were loaded with an individual pathogen, and then treated with the monovalent saclac globule phage formulation of., ϕSPB,BVPaP-3 and KPP independently and also in a cocktail form (ϕSPB,BVPaP-3 and KPP) at different conditions. Efficacy of saclac globule monovalent formulations of indigenous ϕ SPB (Salmonella phage) and BVPaP-3 (Pseudomonas phage) were evaluated against *Salmonella enterica* serovar Paratyphi B and *Pseudomonas aeruginosa* respectively in the water microcosms prepared using river water and swimming pool water spiked independently with the mentioned respective pathogens at different environmental parameters,viz.,growth phase of the cell (log and stationary), incubation conditions (shaking and static) and varied MOI (multiplicity of infection) values (MOI<1, MOI=1, MOI=50 and MOI=100). In case of application of monovalent phage formulation, at different MOI values, there was a great difference in the inactivation rate of *Salmonella enterica* serovar Paratyphi B and *Pseudomonas aeruginosa*, with the maximum inactivation rate at MOI = 100. There was a significant difference ($P < 0.05$) in pathogen inactivation rate at shaking and static condition.The rate of inactivation of the log phase cells was higher than stationary phase cells both at shaking and static conditions at MOI 50 and 100.In case of *Klebsiella pneuminiae* no inactivation of the cells by KPP was observed. There was no reduction in the number of pathogens in the filter-sterilized water micrtocosms (containing natural phages) spiked with the individual pathogens (*Salmonella enterica* serovar Paratyphi B, *Pseudomonas aeruginosa* and *Klebsiella pneuminiae*) treated with respective phage ( ϕSPB,BVPaP-3 and KPP). Results also indicated that the application of phage cocktail did not affect pathogen population. Saclac globules contain goat milk sugar that keeps the bacteria in a log phase of growth for long period of time. At different MOI values, there was a great difference in the inactivation rate of *Salmonella enterica* serovar paratyphI B and *Pseudomonas aeruginosa* with maximum inactivation rate at MOI=100.There was a significant difference ($P< 0.05$) in pathogen inactivation rate at shaking and static condition. The rate of inactivation of log phase cells were higher than the stationary phase cells at shaking as well as static conditions at MOI=50 and MOI=100. Result presented in this study strongly support that saclac globule formulation of bacteriophages in the monovalent form can be used as an effective biological disinfectant for killing many kinds of water borne bacterial pathogens.

Bacteriophages might have a lytic cycle or a lysogenic cycle. With lytic viruss like the T4 phage, microorganism cells ar broken open (lysed) and destroyed when immediate replication of the particle. As before long because the cell is destroyed, the virus issue will realize new hosts to infect. Lytic viruss ar a lot of appropriate for phage medical care. Some lytic phages bear a development called lysis inhibition, wherever completed virus issue won't at once lyse out of the cell if animate thing virus concentrations ar high. This mechanism isn't just like that of temperate virus going dormant and typically, is temporary. In distinction, the lysogenic cycle doesn't end in immediate lysing of the host cell. Those phages able to bear condition ar called temperate phages. Their infectious agent order can integrate with host DNA and replicate along side it, comparatively harmlessly, or might even become established as a cellular inclusion. The virus remains dormant till host conditions deteriorate, maybe because of depletion of

nutrients, then, the endogenous phages (known as prophages) become active. At now they initiate the procreative cycle, leading to lysis of the host cell. because the lysogenic cycle permits the host cell to still survive and reproduce, the virus is replicated all told offspring of the cell. associate degree example of a phage known to follow the lysogenic cycle and also the lytic cycle is that the virus lambda of E. coli. typically prophages might offer advantages to the host microorganism whereas they're dormant by adding new functions to the microorganism order, in a very development referred to as lysogenic conversion. Examples ar the conversion of harmless strains of C. diphtheriae or vibrion cholerae by bacteriophages, to extremely virulent ones that cause contagious disease or infectious disease, severally. ways to combat bound microorganism infections by targeting these toxin-encoding prophages are projected A virus to boot noted informally as an outbreak is also an outbreak that infects and replicates among being and archaea. The term was derived from "bacteria" and thus the and thus the (phagein), which suggests "to devour". Bacteriophages square measure composed of proteins that encapsulate a DNA or RNA ordering, and may have structures that square measure either simple or elaborate. Their genomes may inscribe as few as four genes and as many as several genes. Phages replicate among the bacterium following the injection of their ordering into its protoplasm. Bacteriophages square measure among the foremost common and numerous entities inside the half. Bacteriophages square measure ubiquitous viruses, found wherever being exist. it's enumerable there square measure over 1031 bacteriophages on the world, over every completely different organism on Earth, in addition as being, combined. one of the densest natural sources for phages and completely different viruses is water, where up to 9x108 virions per metric unit of measurement ar found in microorganism mats at the surface, and up to seventieth of marine being is additionally infected by phages. Phages are used since the late twentieth century as an alternate to antibiotics within the former Russia and Central Europe, in addition as in France. they're seen as a potential medical care against multi-drug-resistant strains of the many bacterium (see virus therapy). On the opposite hand, phages of Inoviridae are shown to complicate biofilms concerned in illness|respiratory illness|respiratory disorder} and monogenic disorder and to shelter the bacterium from medication meant to eradicate disease, so promoting persistent infection.