Isolation and characterization of Salmonella and E.coli bacteriophages

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Background Salmonella and E.coli both live in the intestinal tracts of humans and other animals. Salmonella causes a wide spectrum of disease, including gastroenteritis, enteric fever (caused by typhoid and paratyphoid serotypes), bacteremia, fecal infections, to a convalescent lifetime carrier state. There are many types of E. coli, and most of them are harmless. But some can cause very serious infections such as bloody diarrhea, severe anemia or kidney failure, urinary tract infections and other infections, which can lead to death.

The majority of cases of *Salmonella* and E.coli infections are food-borne; Contaminated foods are often of animal origin, such as beef, poultry, milk, or eggs, but any food, including vegetables, may become contaminated. Some *Salmonella* and *E.coli* bacteria have become resistant to antibiotics, largely as a result of the use of antibiotics to promote the growth of food animals. Its increasing antimicrobial resistance, prevalence, virulence, and adaptability are a challenge worldwide. The phages nowadays are seen as a possible therapy against multi-drug-resistant strains of many bacteria.

Goal: The present study aims isolation and selection of the phages active against *Salmonella* sp.and *E.coli* which would be then used for construction of theraputic or prophylactic bacteriophage mixtures applicable for humans and/or animals.

Materials: Altogether 35 bacteriophage clones active against *Salmonella* sp.(30) and *E.coli* (5) have been isolated from 10 sewage samples.

Methods: Traditional methodology described by Adams (1958) has been used for isolation and characterization of bacteriophages, such as identification of host ranges, serological relatedness, etc. Genetic characterization was performed using fRFLP (fluoroscent Restriction Fragment Length Polymorphism) analysis (Merabishvili et al., 2007).

Results: All newly isolated phage clones have been studied for the morphology of their negative plaques and virions and host ranges. The phages have been screened against 81 strains of different geographical origin related to *S, typhimurium, S. enteritdis, S. agona, S.pullorum, S. gallinarum* and *S. cholera-suis.* The selected clones of bacteriophages showed higher effectiveness to *S. typhimurium* and *S. enteritidis* which varied between 72,1% to 86, 5%.

In total 7 phage clones related to *Salmonella* sp. and 4 phage clones related to *E.coli* have been tested using phage neutralization reaction and fRFLP tests. Among *Salmonella* spp. phage clones several groups with similar genotypes were identified. Group I includes bacteriophages N4 (BTR S.g. 1), N6 (BTR 2) and N7 (Mtkvari # 2 S.p.), group II – N8 (S.e. MG) and N9 (S.t. MG), group III – N1 (Mtkvari 2 S.p.) and N10 (S. p. MG). Phage clone N 2 (S. t. Gotua str) showed absolutely distinguished genome fingerprint in all kind of analysis and thus is genetically distanced from all other phages. All four *E. coli* phage clones proved to have very similar (almost identical) genotypes. These results were completely proved also by serology study performed using phage neutralization reaction with the corresponding anti-phage sera.

Conclusion: The phage clones characterized in the scope of the present study were approved as virulent phages with broad overlapping host ranges and may be recommended for construction of phage mixtures for therapeutic and/or prophylactic use.