Instructions for use

E. COLI BACTERIOPHAGES

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For *in vitro* diagnostic use

Intended use
E. coli bacteriophages are intended for determination of capsule polysaccharides (K antigens) with the cross-brush method[1].

Description
E. coli bacteriophages from SSI Diagnostica are supplied in vials with 1 mL. *E. coli* bacteriophages consist of phages that recognize K polysaccharides on the surface of *E. coli* bacteria. Phage serotyping of K antigens is crucial for serotyping of K antigens, for which it is not possible to produce polyclonal antibodies in mamalians. The maximum number of tests per agar plate is limited to six bacterial cultures and one positive control (see figure 1).

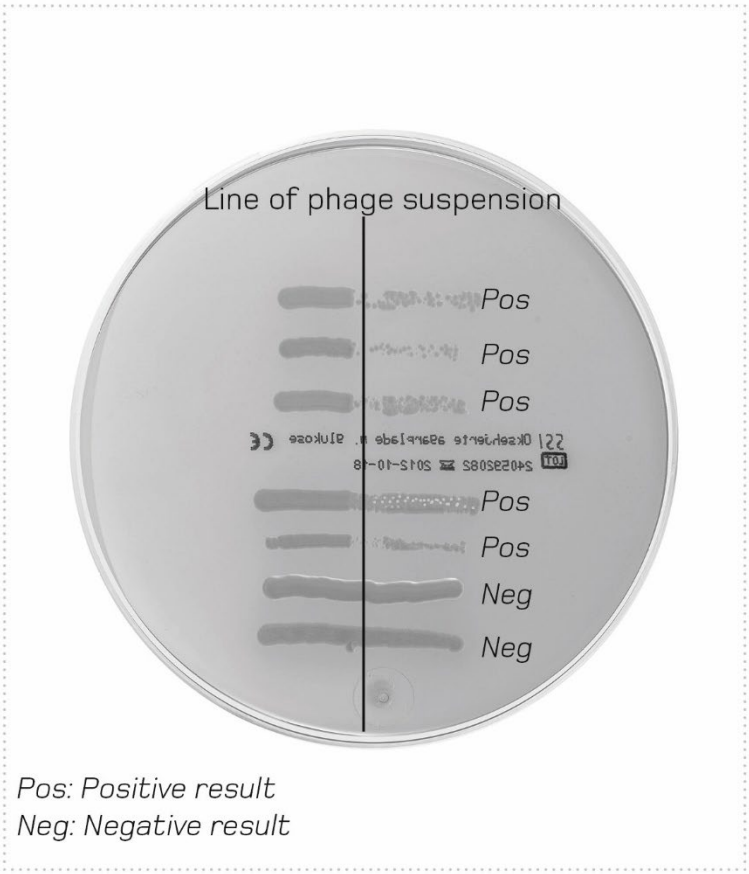

Figure 1. Result from phage typing.

Principle
E. coli phage suspensions are intended for a cross-brush method. Phages are epitope-specific in relation to specific antigens, and these can therefore, be used for serotyping. If a bacterial culture is mixed with a specific phage suspension on a fresh culture medium, the phages will attach to the surface components and infect the bacteria resulting in lysis of the bacteria.

After six hours, bacteria expressing a phage receptor will show no or very limited growth in the phage suspension area of the culture medium, and significant growth on the clean culture media. Bacteria not expressing a phage receptor will not be infected and growth will be observed in the full length of the bacterial streak across the phage

line (see figure 1).

Materials required but not provided
- Beef broth culture
- Beef broth agar plate
- 10 µL and 1 µL inoculation loop
- Inoculation needle
- Incubator (35-37 °C)

Procedure
1. A beef broth culture is inoculated with the selected strain and incubated at 35-37 °C overnight.
2. With a 10 µL inoculation loop, a vertical line of phage suspension is applied onto a beef broth agar plate. The phage suspension is applied equally in the whole length of the line by drawing the loop up and down twice.
3. The agar plate is left for 10 min., or until the applied phage suspension has dried.
4. Streak a horizontal line of live broth culture from left to right, crossing the phage suspension, using a 1 µL inoculation needle (see figure 1).
5. The agar plate is incubated for 6 hours at 37 °C after which the result is read. In case the result is difficult to interpret at this stage, the plate may be placed in the incubator again for 2 hours and read again. Alternatively place the plate at 4 °C overnight and incubate for a couple of hours at 37°C the next day before reading the result.
6. A positive reaction is seen by no or limited growth after crossing the phage line. The negative reaction is seen by growth in full length of the bacterial streak. A positive reaction can also be identified by the plaque method.

Positive and negative controls
Positive control strains for all phage suspensions are available at SSI Diagnostica, listed under reference strains for K antigens.

Quality certificate
SSI Diagnostica's development, production and sales of *in vitro* diagnostics are quality assured and certified in accordance with ISO 13485. Certificate of analysis can be downloaded from our website: ssidiagnostica.com.

References
1. Ørskov F., Ørskov I., Serotyping of *Escherichia coli*, Methods in Microbiol., 14:44-112, 1984.

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