SSI DIAGNOSTICA

# SUMMARY
# STABILITY STUDY
# *E. coli* antisera

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## PRODUCT GROUP(S):

The SSI Diagnostica product range of *E. coli* antisera, are divided into the following categories:

- O single and pool serum represented by **O26, O55, O145, O157 and O177** antiserum
- OK O single and pool serum represented by **OK O26, OK O121, OK O145, OK O157 and OK O pool 1** antiserum
- H single and pool serum represented by **H2, H11 and H40** antiserum

By selecting the above-mentioned antisera, we represent the majority of the *E. coli* product range. The results are therefore applicable for the above-mentioned range of *E. coli* products.

## DESCRIPTION OF STUDY:

The stability study has been performed from 2002 and reports the stability of the above-mentioned antiserum. The report is conducted according to ISO 23640:2015.

The chosen antisera were stored at 2-8°C and were tested after 1, 3, 6, 9, 12, and 18 months, then at year 2, 3, and 4 or until reactions are no longer positive. Two positive strains were tested at the different time points. All the antisera were ready-to-use products and there was not a single vial for each time point meaning that the same vial have been opened serval times. All tests were made with non-sealed vials and no sterility testing were performed during the duration of the study. All results are stored locally and a summery are reported here.

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1. **NUMBER OF LOTS:** The ready-to-use antisera used in this study were from bulk antisera produced at different timepoints, but all were bottled in 2002. The study includes one lot antisera for each product. Each lot were bottled in tree lots.

2. **LOT NO.:** O26 A1-1 (O26), O55 A1-1 (O55), O145 A1-1 (O145), O157 A1-1 (O157), O177 A1-1 (O177), OKO 26 A3-1 (OK O26), OKO 121 A1-1 (OK O121), OKO 145 B1-1 (OK O145), OKO 157 A1-1 (OK O157), OK O pool 1 A2-3 (OK O Pool 1), H2 D1 (H2), H11 D2 (H11), and H40 D1 (H40).

3. **TEMPERATURE:** The temperature was 2-8°C throughout the entire study.

4. **TEST:** Tests have been performed every three months over the first year, every six months over the second year, and annually thereafter in 4 years. The test methods have been performed according to the IFU.
   O antisera are used for agglutination of boiled cultures in round bottom microtitre plates. Equal quantities (80 µL) of antiserum and culture are mixed, and the result is read after overnight incubation at 50-52 °C. A positive reaction is seen as a "grey carpet", covering the bottom of the well, often in a clear fluid. When the reaction is negative the bacterial suspension is seen as a small white spot in a clear or a milky turbidity centered in the well.
   H antisera are used for agglutination of formalin-killed cultures (final formalin concentration of 0.48%) in test tubes. Equal quantities (180 µL) of antiserum and culture are mixed, and the result is read after 1½-2 hours' incubation at 50-52 °C. A positive reaction is seen as a loose and fluffy cloud in a clear fluid. A negative reaction is seen as a homogenous milky turbidity.
   All tests are performed with 2 positive strains for each specific antiserum. All strains used are documented in the raw data of the study.
   OK O antisera are intended for slide agglutination. 20µL antiserum and 3 representative colonies of the strain to be tested are mixed on a slide. The slide is tilted for 5-10 sec. A positive reaction is seen as a visible agglutination. A negative reaction is persistence of the homogeneous milky turbidity.
   Physiological saline pH 7.4 is used as a negative control and must be negative

5. **RETEST:** If strains during the study do not react with homologue antisera or auto agglutinates, the strain is substituted with another strain with similar antigen definitions.

6. **HUMIDITY:** N/A

7. **TEST OF INTEGRITY DURING TRANSPORT:** The antisera product line might obtain bruises to their containers when handled roughly but it will not affect the performance of the products. For raw date see report "Drop-test for ImmuView, antisera and Culture Media".

8. **IN-USE STABILITY:** Equal to storage stability

**ACCEPTANCE CRITERIA:**

The acceptance criteria for a positive agglutination is as described in the IFU. For O antisera a gray carpet in the microtiter well, for OK O antisera grains in a clear fluid on a glass slide and for H antisera a fluffy cloud in a clear fluid.

- **Titer:** The titer for *E. coli* antiserum are controlled during the stability study.
- **Long-term stability:** At the termination of the stability study, the antisera must show a positive reaction in ready-to-use solution for the reference strains.
- **Other:** Strains used as QC panel can be substituted with strains with similar antigen combination during the stability study.

**RESULTS:**

All results are recorded throughout the period of 4 years.

In table 1 the results are visualized, stating performance at the beginning and the end of the study (4 years), all raw data are available on request.

*Table 1: Summarizing of results from stability study over 4 years*

| | Antisera | | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | O26 | O55 | O145 | O157 | O177 | Pool 1 | OK O26 | OK O121 | OK O145 | OK O157 | H2 | H11 | H40 |
| **Start of study** | POS | POS | POS | POS | POS | POS | POS | POS | POS | POS | POS | POS | POS |
| **End of study** | POS | POS | POS | POS | POS | POS | POS | POS | POS | POS | POS | POS | POS |

*(POS = positive)*

**CONCLUSION:**

*E. coli* antiserum stored at 2-8° C have a duration of minimum 4 years of storage. The shelf life is determined as 4 years from date of manufacturing at a temperature range of 2-8° C.

**Transport:** The antisera product line might obtain bruises to their containers when handled roughly but it will not affect the performance of the products.

**In-use:** The in-use stability is not affected by working with the antiserum on the bench. During testing if it is stored at 2-8° C for no longer than 4 years form production date. In conclusion the shelf life and performance of the antiserum are set to four (4) years from the date of manufacturing.

**APPROVED:**

**DATE: 2020-09-09**

**SIGNATURE:**

IVD TEAMLEAD

IVD MANAGER