



# Mutagenic Potential of PHYTOCEE® in *Salmonella typhimurium* mutant strain Ta98

## OBJECTIVE

To evaluate the mutagenic potential of PHYTOCEE® and / or its metabolites to induce reverse mutations at the histidine locus in the genome strains of *Salmonella typhimurium*.

## MATERIALS AND METHODS

Histidine auxotrophic strains of *Salmonella typhimurium* bacteria viz. TA98 were used in this experiment. 2-Nitrofluorene (2NF) + 4-Nitroquinolene-n-oxide (4NQNO) at the final concentration of 2 + 0.5 µg/ml were used as positive control for experiments without metabolic activation. For experiments with metabolic activation 2-Aminoanthracene at the final concentration of 5 µg/ml was used as positive control. PHYTOCEE® was used as test substance at concentrations of 5000 µg/ml, 1510 µg/ml, 450 µg/ml, 140 µg/ml, 40 µg/ml and 10 µg/ml, using DMSO as the control. *Salmonella typhimurium* reverse mutation assay was carried out using standardized in-house assay protocol.

## RESULTS

Table 1. Effect of PHYTOCEE® on colony counts of histidine revertants in the *Salmonella typhimurium* strain TA 98

| Treatment                    | Concentration (µg/ml) | Number of revertants colonies |                          |    |    |                 |                             |
|------------------------------|-----------------------|-------------------------------|--------------------------|----|----|-----------------|-----------------------------|
|                              |                       | S-9                           | Individual colony counts |    |    | Mean ± S.D      | Fold increase over baseline |
|                              |                       |                               | 1                        | 2  | 3  |                 |                             |
| Phytocee                     | 10                    | -                             | 2                        | 0  | 1  | 1.00 ± 1.00     | 0.55                        |
|                              |                       | +                             | 1                        | 1  | 1  | 1.00 ± 0.00     | 0.55                        |
|                              | 40                    | -                             | 0                        | 0  | 1  | 0.33 ± 0.58     | 0.18                        |
|                              |                       | +                             | 2                        | 0  | 0  | 0.67 ± 1.15     | 0.37                        |
|                              | 140                   | -                             | 2                        | 1  | 0  | 1.00 ± 1.00     | 0.55                        |
|                              |                       | +                             | 0                        | 1  | 4  | 1.67 ± 2.08     | 0.92                        |
|                              | 450                   | -                             | 1                        | 1  | 1  | 1.00 ± 0.00     | 0.55                        |
|                              |                       | +                             | 2                        | 1  | 0  | 1.00 ± 1.00     | 0.55                        |
|                              | 1510                  | -                             | 2                        | 1  | 2  | 1.67 ± 0.58     | 0.92                        |
|                              |                       | +                             | 1                        | 1  | 0  | 0.67 ± 0.58     | 0.37                        |
|                              | 5000                  | -                             | 1                        | 2  | 1  | 1.33 ± 0.58     | 0.73                        |
|                              |                       | +                             | 1                        | 1  | 0  | 0.67 ± 0.58     | 0.37                        |
| Negative control             | DMSO (4%)             | -                             | 2                        | 0  | 0  | 0.67 ± 1.15     | ---                         |
|                              |                       | +                             | 0                        | 2  | 0  | 0.67 ± 1.15     | ---                         |
| Positive control (2NF+4NQNO) | 2NF (2) + 4NQNO (0.5) | -                             | 48                       | 48 | 48 | 48.00 ± 0.00*** | 26.37                       |
| Positive control (2AA)       | 2AA (5)               | +                             | 48                       | 48 | 48 | 48.00 ± 0.00*** | 26.37                       |

2NF, 2-Nitrofluorene, 4NQNO, 4-Nitroquinoline-N-oxide, 2AA - 2-Aminoanthracene; \*\*\*p<0.01

CONCLUSIONS

The results demonstrated that PHYTOCEE® administration at all the tested concentrations did not induce mutations both in the presence and absence of metabolic activation in *Salmonella typhimurium* mutant strains TA98 and TAMix.

OUTCOME

Hence, PHYTOCEE® was found to be non-mutagenic in *Salmonella typhimurium* mutant strain TA98 up to a concentration of 5000 µg/ml.