

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

### 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 TAMix 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 TAMix

Treatment	Concentration (µg/ml)	Number of revertants colonies						
		S-9	Individual colony counts			Mean ± S.D	Fold increase over baseline	
			1	2	3			
Phytocece	10	-	2	2	0	1.33 ± 1.15	0.70	
		+	1	2	1	1.33 ± 0.58	0.54	
	40	-	3	1	2	2.00 ± 1.00	1.05	
		+	1	1	0	0.67 ± 0.58	0.27	
	140	-	0	0	1	0.33 ± 0.58	0.17	
		+	0	1	0	0.33 ± 0.58	0.13	
	450	-	0	1	2	1.00 ± 1.00	0.52	
		+	0	2	1	1.00 ± 1.00	0.40	
	1510	-	0	1	0	0.33 ± 0.58	0.17	
		+	0	2	0	0.67 ± 1.15	0.27	
	5000	-	1	1	2	1.33 ± 0.58	0.70	
		+	1	1	1	1.00 ± 0.00	0.40	
	Negative control	DMSO (4%)	-	1	1	2	1.33 ± 0.58	---
			+	2	0	2	1.33 ± 1.15	---
Positive control (2NF+4NQNO)	2NF (2) + 4NQNO (0.5)	-	48	48	48	48.00 ± 0.00***	25.13	
Positive control (2AA)	2AA (5)	+	45	47	47	46.33 ± 1.15***	18.61	

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 TAMix up to a concentration of 5000 µg/ml.