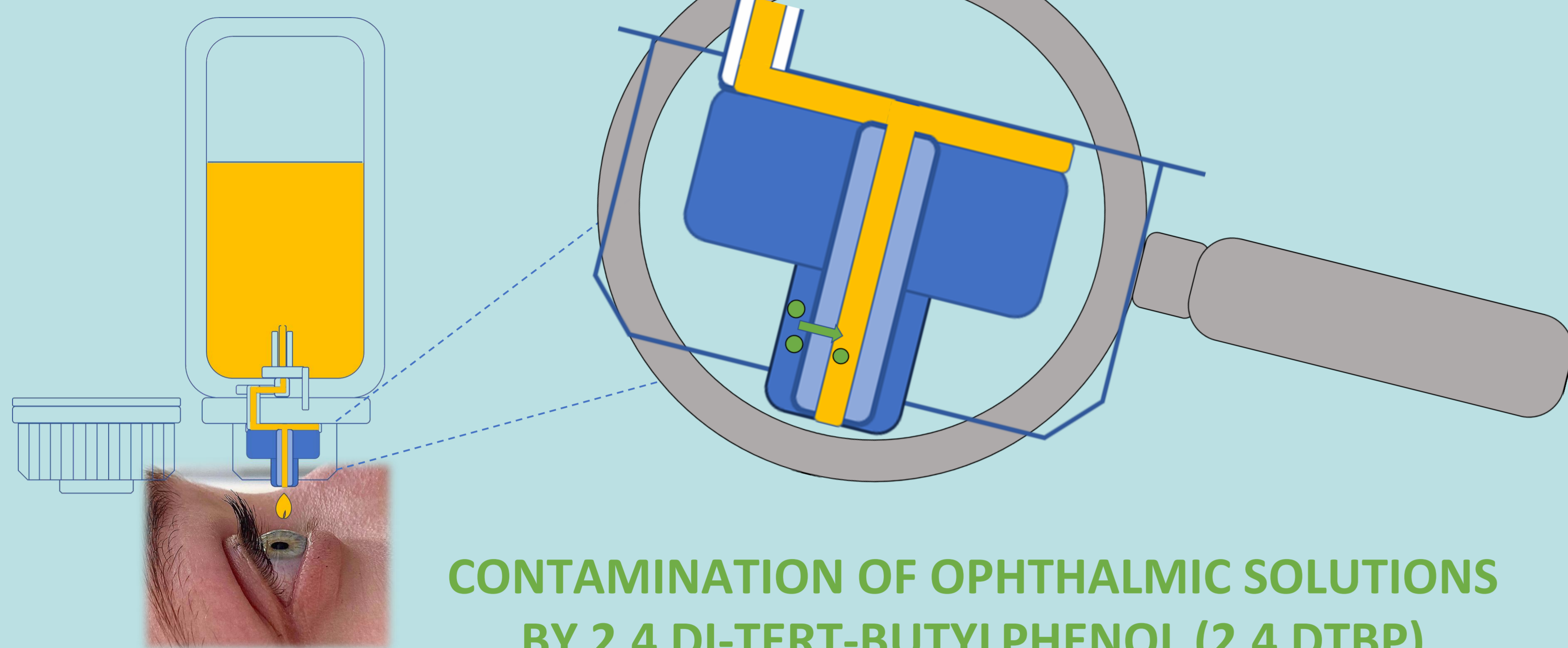


3PC – 017: MIGRATION OF LEACHABLE 2,4-DI-TERT-BUTYLPHENOL FROM PRESERVATIVE-FREE MULTI-DOSE DROPPER SYSTEMS INTO OPHTHALMIC PREPARATIONS AND ASSESSMENT OF IN VITRO CYTOTOXICITY

M. Barrieu¹, O. Allard², L. Bernard¹, V. Sautou¹, P. Chennell¹

¹ University Clermont Auvergne, CHU Clermont Ferrand, Clermont Auvergne INP, CNRS, ICCF, France - ² CHU Clermont Ferrand, France.

Introduction



CONTAMINATION OF OPHTHALMIC SOLUTIONS BY 2,4 DI-TERT-BUTYLPHENOL (2,4 DTBP)

Eye drops packaged in preservative-free multi-dose containers (MDC) eliminate the need to add antimicrobial preservatives, which is a significant advantage due to their local cytotoxicity. Under certain conditions, contact between MDC and the eye drops may lead to the release of compounds such as 2,4 DTBP, thereby contaminating the patients. The local toxicity of 2,4 DTBP and the clinical consequences should be assessed.

Objectives : To determine if and how much 2,4 DTBP migrates into various eye drops from the MDC and to assess its cytotoxicity threshold.

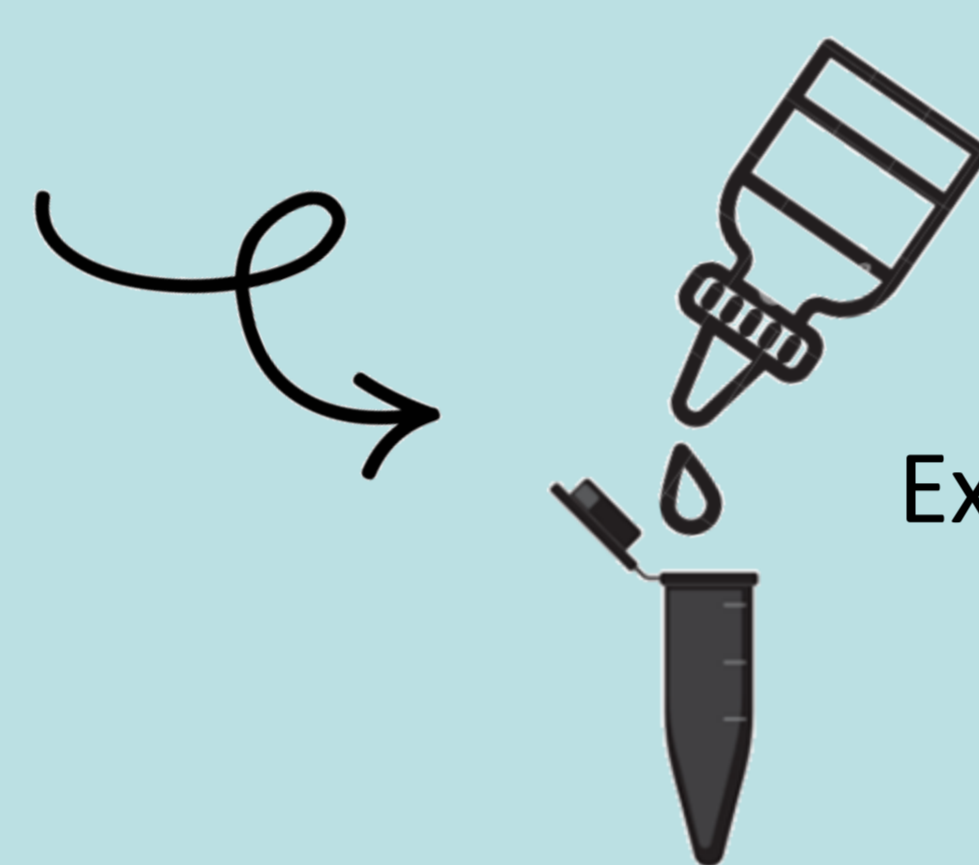
Material and methods

*MTT: 3-(4,5-diméthylthiazol-2-yl)-2,5-bromure de diphenyltetrazolium

STEP 1: IN USE ASSAY AND QUANTIFICATION OF 2,4 DTBP

SELECTION OF OPHTHALMIC SOLUTIONS, CONDITIONNED IN NOVELIA® MDC (NEMERA)

Tacrolimus (TAC) 0.2 mg/mL
Ciclosporine (CSA) 1, 10 and 20 mg/mL
Atropine 0.1 and 0.5 mg/mL
Ceftazidime and Vancomycine 25 mg/mL
Voriconazole (VCZ) 10 mg/mL
Costec® = Dorzolamide 20 mg/mL + Timolol 5mg/mL



COLLECTION OF DROPS

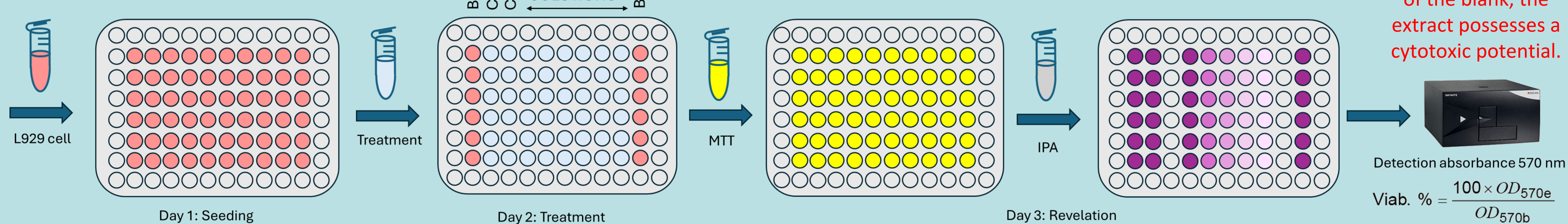
Twice a day for one month
Excluding weekends and public holidays



HPLC-UV ANALYSIS OF DROP CONTENTS

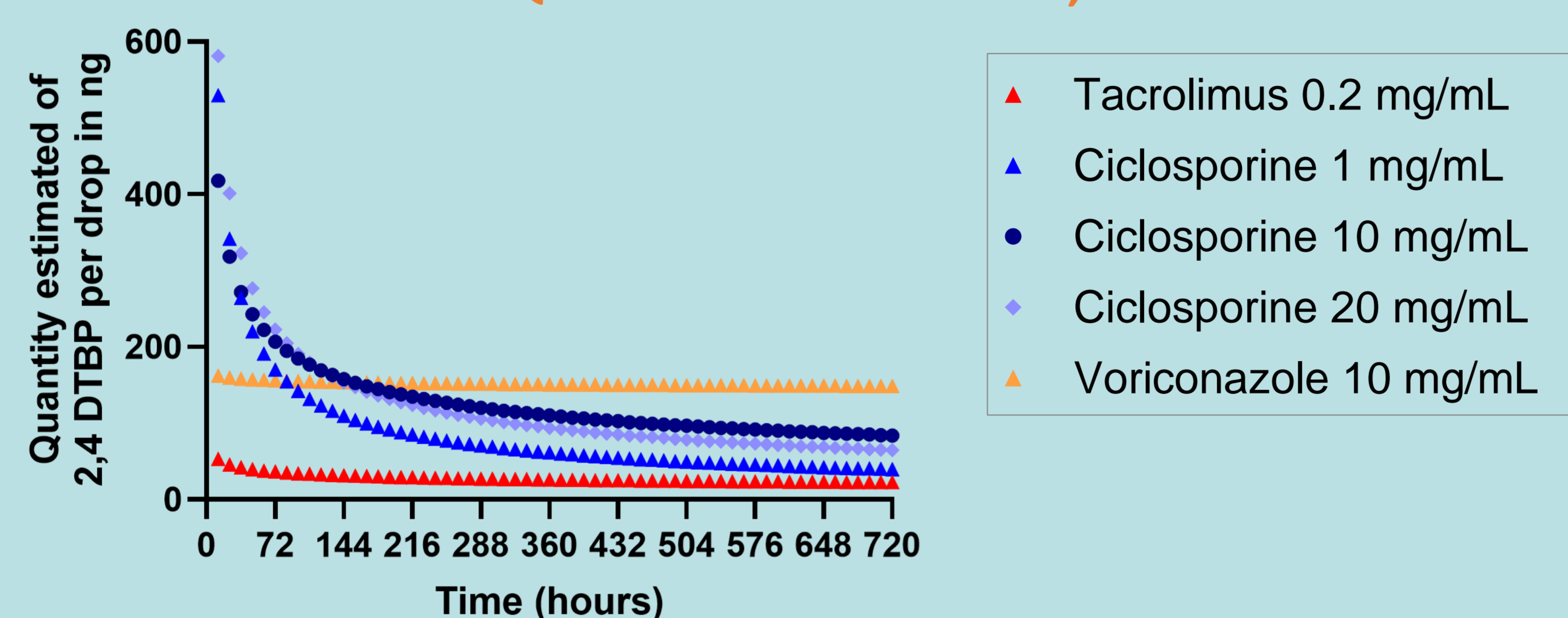
STEP 2: CYTOTOXICITY ASSAY

MTT* TEST ACCORDING ISO 10993-5



Results and discussion

IN USE ASSAY AND QUANTIFICATION OF 2,4 DTBP

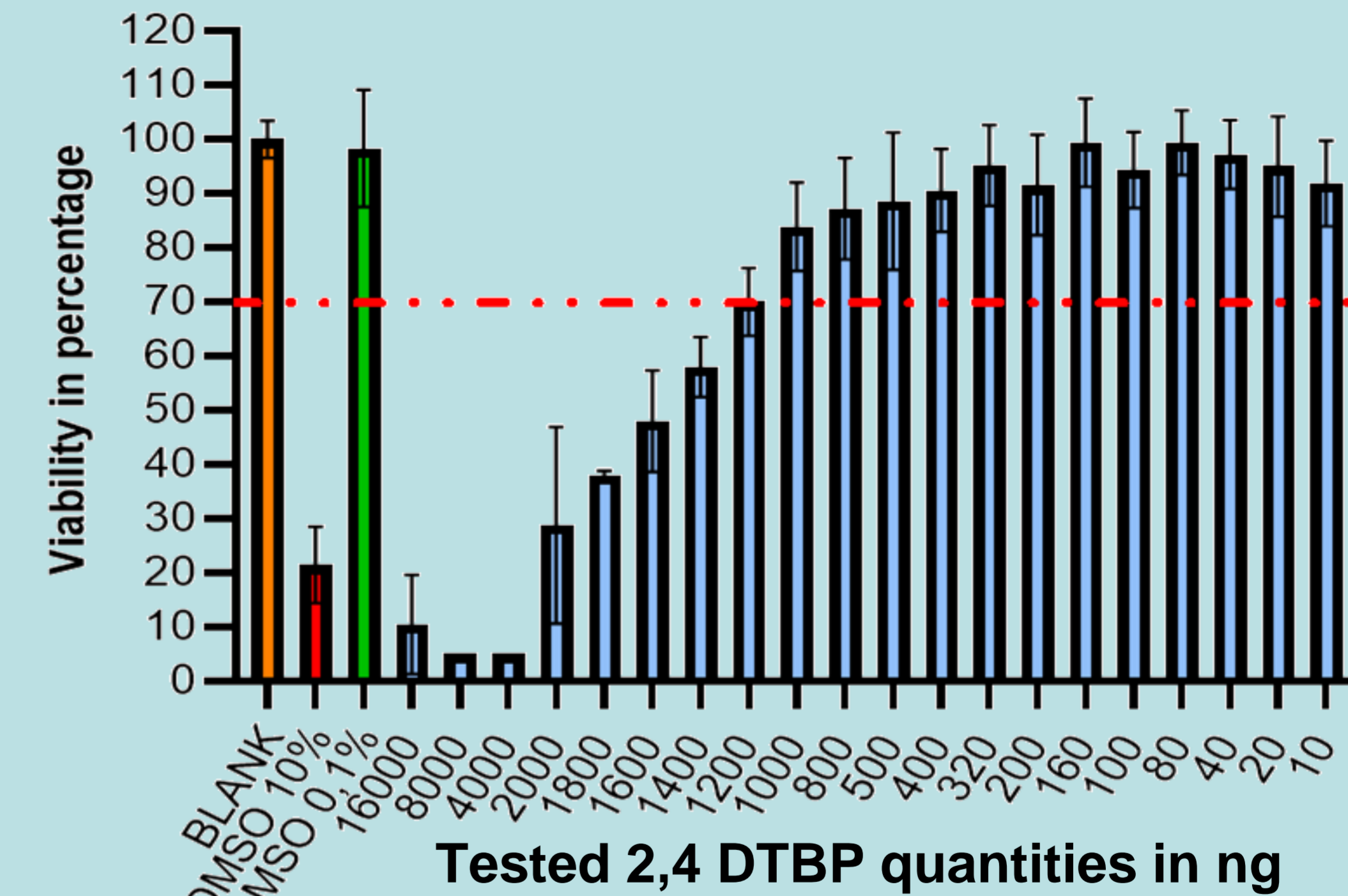


Estimated quantity of 2,4 DTBP per eye drop (ng)	TAC 0.2 mg/mL	CSA 1 mg/mL	CSA 10 mg/mL	CSA 20 mg/mL	VCZ 10 mg/mL
Max	53	530	418	573	162
Min	23	40	84	66	149
Mean	28	89	131	123	152
Cumulated over one month	1687	5353	7858	7531	9108

Impact of the formulation on the migration of 2,4 DTBP

- Hydrophobic active substance, containing a surfactant for TAC and CSA and cyclodextrin for VCZ : increased leaching
- Hydrophilic active substance and no lipophilic excipient : no leaching

CYTOTOXICITY ASSAY



Daily exposure to 2,4 DTBP quantities found in TAC, CSA and VCZ eye drops was **below the cytotoxic limit of 1.2 µg** and **lower** than existing toxicological thresholds:

- ISO/TS 21726: For medical devices with limited and prolonged contact, the value is **120 µg/day** and for a period exceeding 10 years is **1.5 µg/day**
- Product Quality Research Institute: For ophthalmic irritants and sensitizers compounds, the value is **5 µg/day**

Conclusion

The nature of the formulation seems to influence the migration of 2,4 DTBP. Cytotoxicity tests performed on the L929 cell line did not reveal any toxicity of 2,4 DTBP after 24 hours at the quantities found in the drops for the tested medicines, which is reassuring. Further studies are needed to determine its cumulative toxicity during chronic administration and adapting the MTT test to corneal cells would enable a more accurate evaluation of its ocular cytotoxicity.

