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Genotoxicity assessment of a polycarbonate sol-gel coating and its precursors intended to be used for food contact.

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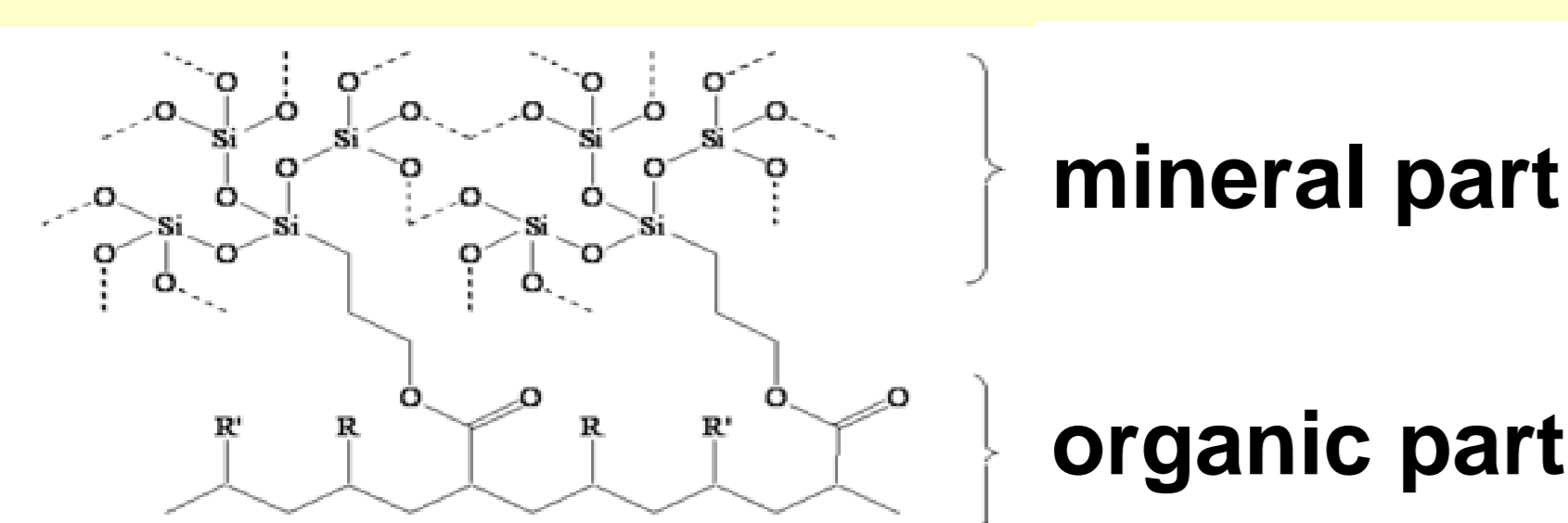
INTRODUCTION

Polycarbonate (PC) is a widely used polymer in food packaging all around the world. However, due to the release of Bisphenol A, an endocrine disruptor during repeated washing cycles and certain physico-chemical conditions, its use becomes compromised. To solve this issue, sol-gel coatings based on organoalkoxysiloxane were developed for PC to act as a physical barrier. Common precursors, namely tetraethylorthosilicate (TEOS) and 3-glycidyloxypropyltriethoxysilane (GPTES), were used to prepare a sol-gel system (A8). These two chemicals, as well as the films obtained from the sol-gel systems were studied with regards to their potential toxicity *in vitro*. Our motivation for studying the toxicity of these precursors is to assess whether they are still present as traces in the final material, in case the polymerization step is incomplete and/or if the precursors are released during the packaging life time. Migration of extracts was measured and their genotoxicity was assessed using *in vitro* bioassays (Ames test and micronucleus assay).

SOL-GEL TECHNIQUE

Hydrolysis: $\equiv\text{Si-OR} + \text{HOH} \rightarrow \equiv\text{Si-OH} + \text{R-OH}$

Condensation: $\equiv\text{Si-OH} + \text{HO-Si}\equiv \rightarrow \equiv\text{Si-O-Si}\equiv + \text{H}_2\text{O}$
 $\equiv\text{Si-OH} + \text{RO-Si}\equiv \rightarrow \equiv\text{Si-O-Si}\equiv + \text{R-OH}$



- « Soft » chemistry
- Easy to use
- Low process temperature

COMPOSITION OF A8 FORMULATION

- Silanes (TEOS, GPTES)
- Colloidal silica
- Acetic acid
- Solvent (alcohol)
- Water

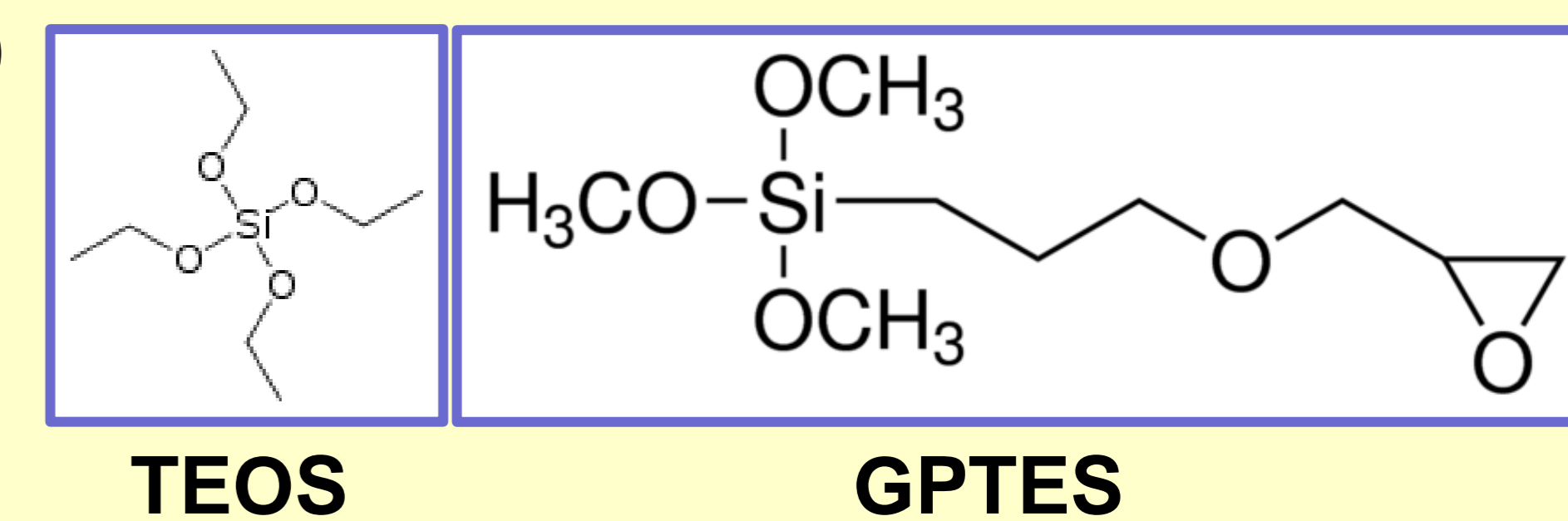
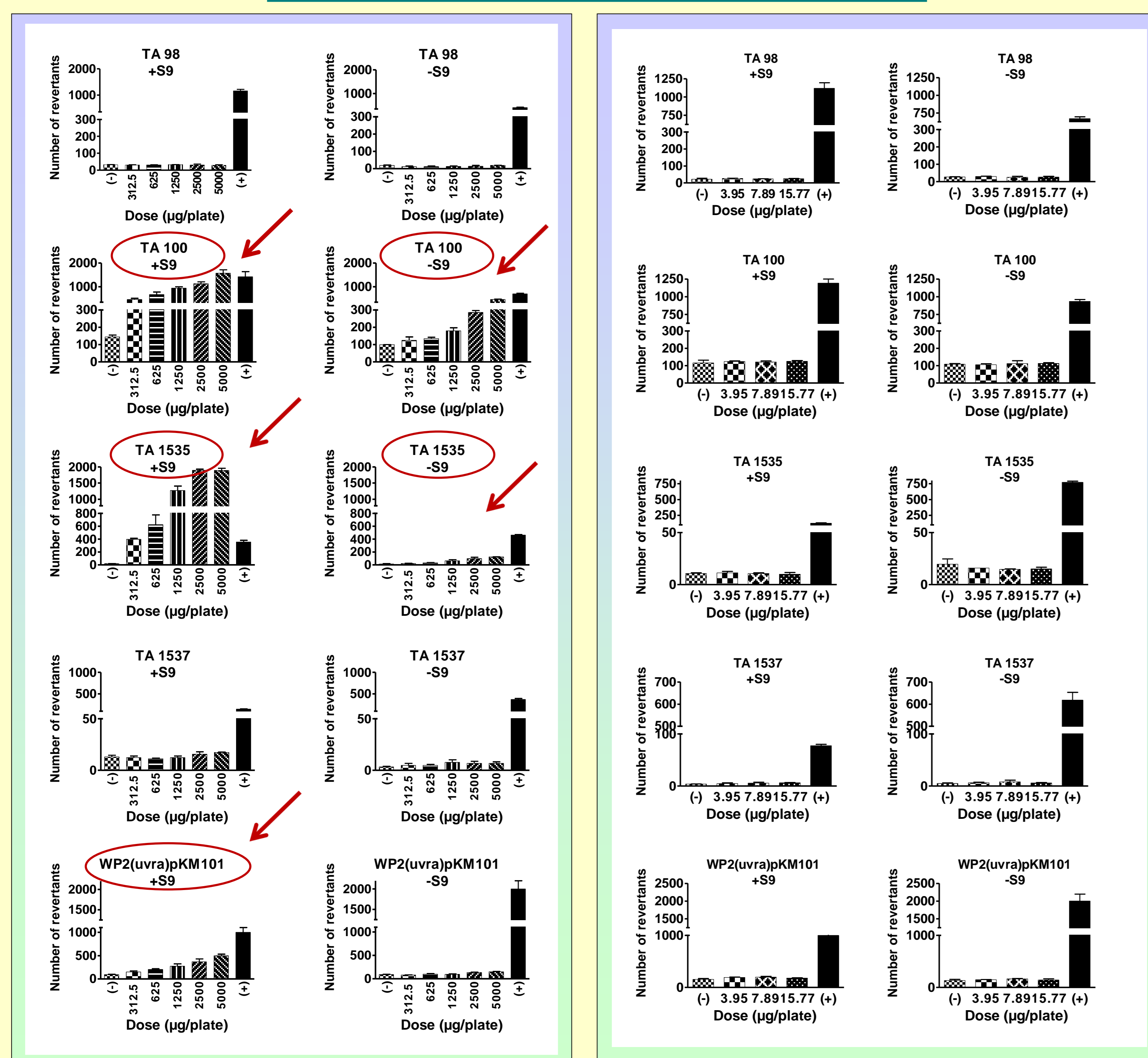


Table 1: Results of experimental migration for A8 formulation, expressed in mg/dm².

A8 formulation	Food simulants	Individual values	Mean (mg/dm ²)
1h, 100°C	3% acetic acid	2.6; 6.8; 1.1	3.5
1h, 100°C	10% ethanol	6.1; 16.2; 2.7	8.3
3h, 60°C	95% ethanol	4.1; 1.1; <0.1	1.8
1h, 60°C	Isooctane	0.3; 0.1; <0.1	0.2
1h, 100°C	Olive oil	0.4; 0.8; 11.7	4

RESULTS/DISCUSSION



Precursors:

- ❖ TEOS was not mutagenic in the Ames test (data not shown).
- ❖ In contrast, a significant positive response was observed with GPTES in the TA100, TA1535 and WP2(uvra)pKM101 strains. The mutagenic effect was more pronounced in the presence of the exogenous metabolic activation system with an increase of the induction factor (ten-fold increase for the TA1535 strain) (Figure 1). This effect is certainly due to the chemical structure of GPTES with the presence of an epoxy group, which is a structural alert in genotoxicity (Ashby and Tennant, 1991).
- ❖ In the micronucleus assay, GPTES gave negative results even in the presence of an exogenous activation system (Figure 3).

Coating:

- ❖ To ascertain the possibility of using this precursor in food contact material, migration tests were performed on coating formulation using food stimulants.
- ❖ The highest migration was obtained in 10% ethanol for A8 formulation (Table 1).
- ❖ Ames test (Figure 2) and MN assay (Figure 4) were also performed with A8 extract. No genotoxic effect was observed. This indicates that probably no or few unreacted GPTES molecules were present in the extract.
- ❖ This could be due to epoxy reactivity: it is well-known that in presence of polar bonds such as C-NH₂, (formed on the PC surface during N₂/H₂ plasma treatment), epoxy rings can open and react with the latter, creating covalent bonds at the interface.
- ❖ Therefore, in addition to improve the coating for substrate adhesion, the N₂/H₂ plasma treatment allows the epoxy group transformation which reduces the probability of observing mutagenic effect.

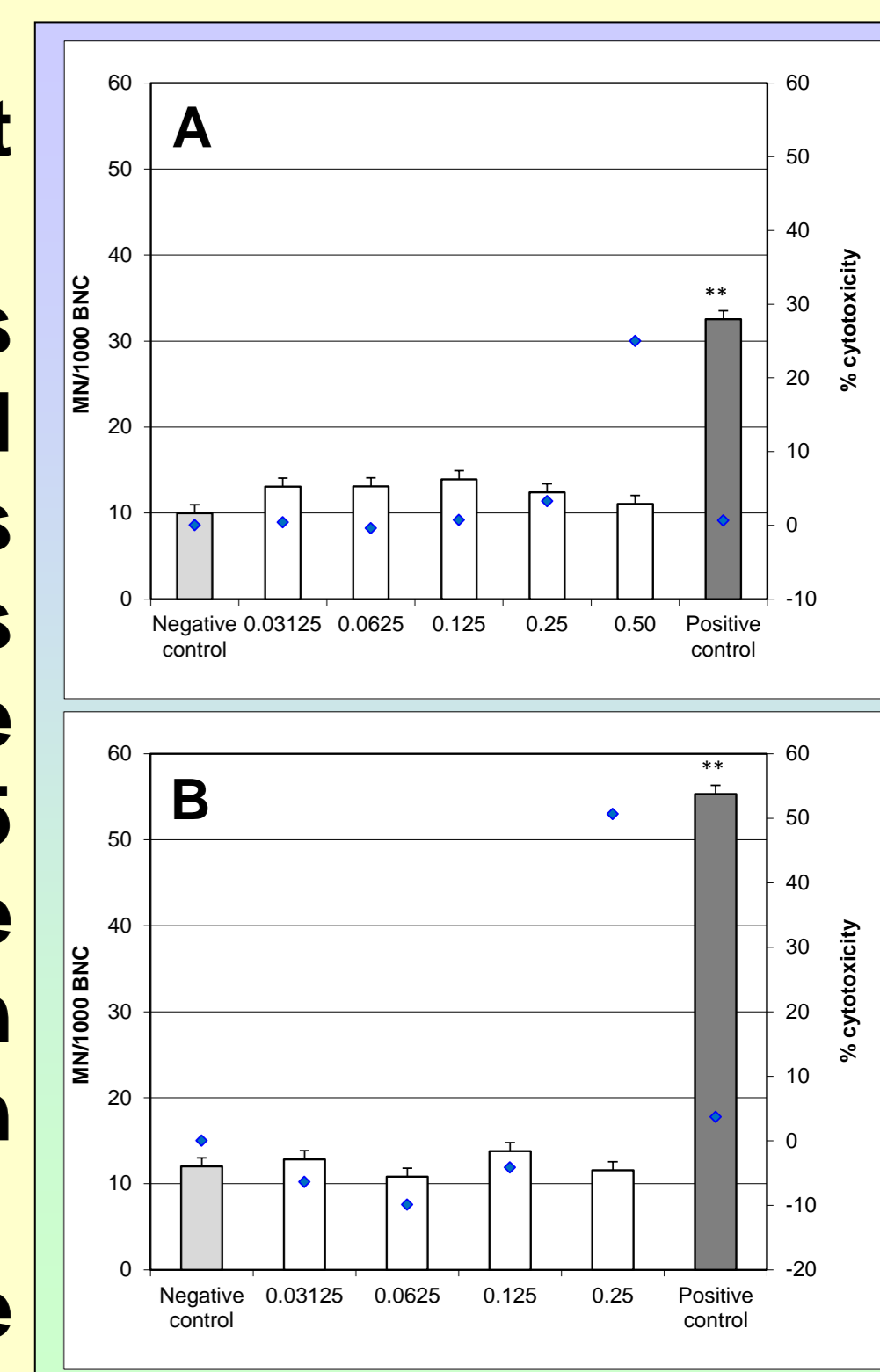


Figure 3: MN and % of cytotoxicity data when the HepG2 cells were exposed to GPTES (mg/ml) with the S9 mix (4 h) (A) or without the S9 mix (24 h) (B). ** P < 0.01, significantly different from the negative control. Positive controls: cyclophosphamide (A) at 10 µg/ml and vinblastine sulfate (B) at 0.625 ng/ml.

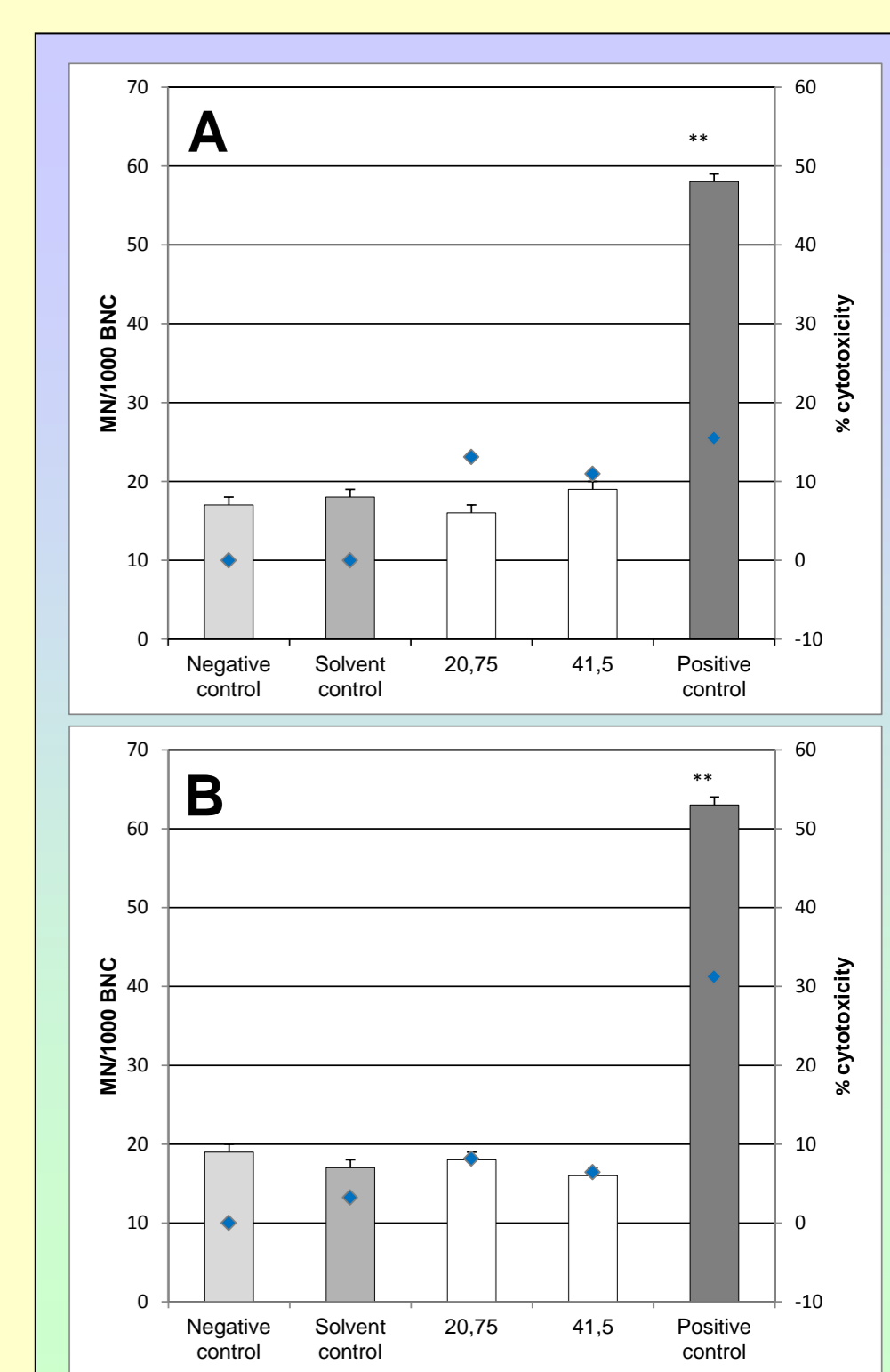


Figure 4: The MN and % of cytotoxicity data when the HepG2 cells were exposed to A8 formulation extract (µg/ml) with the S9 mix (3 h) (A) or without the S9 mix (24 h) (B). ** P < 0.01, significantly different from the negative control. Positive controls: cyclophosphamide (A) at 10 µg/ml and vinblastine sulfate (B) at 0.625 ng/ml.

CONCLUSION

Using regulatory battery of genotoxicity assays, only GPTES was positive in the Ames test. Migration of A8 formulation was the highest in the 10% ethanol simulant but under the regulatory overall migration limit (EU 10/2011). The sol-gel coating formulation is promising for food contact, but additional data, such as chemical analyses, or release study, are on the way to obtain the complete characterization of the extract and to claim A8 derived coatings as food grade materials.

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