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Emergence of biocide resistance in *Salmonella* Typhimurium via directed evolution experiments

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1 INTRODUCTION

Salmonella spp. is a biotic agent responsible for foodborne outbreaks. It is the second most reported toxi-infection, causing more than half of all hospitalizations related to foodborne. The use of antibiotic treatments for clinical cases of salmonellosis may lose effectiveness due to the emergence of antimicrobial resistance.

The phenomenon of antibiotic resistance is considered a global problem due to the threat it poses to public health, so it is being widely studied. However, it is uncertain whether enhanced resistance might occur against other antimicrobial compounds, such as biocides.

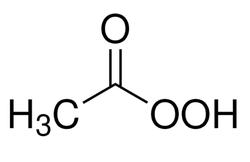
Among the available biocides, peracetic acid (PAA) is one of the main biocides used as surface disinfectants in the food industry. The mechanism of its biocidal activity consists of the oxidation of components of its cell membrane components, mainly membrane proteins, which leads to instability and subsequent cell lysis.

2 OBJECTIVE

This study aims to determine whether peracetic acid (PAA) exposure could lead to the emergence of resistant variants in *Salmonella* Typhimurium populations.

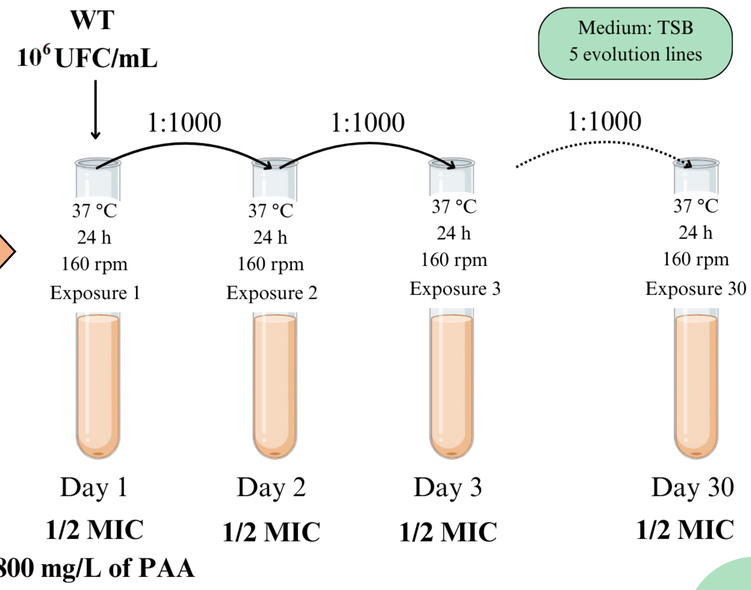
3 MATERIAL & METHODS

Peracetic acid
(Supelco, Alemania)

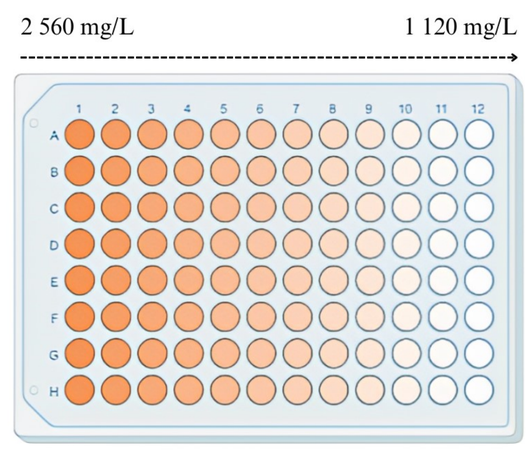


Salmonella enterica subsp. *enterica*
serovar Typhimurium str. LT2
(CECT 722)

Adaptative Laboratory Experiment **ALE**



Minimum Inhibitory Concentration **MIC**



Turbidity is measured at **620 nm** to determine the MIC

MIC determination of evolved populations and isolated strains

4 RESULTS

Table 1. MIC comparation between parental strain of *S.* Typhimurium (WT) and evolved populations.

Bacterial Populations	MIC (mg/L)
S. Typhimurium	1600
1P	1760
2P	1760
3P	1760
4P	1760
5P	1760

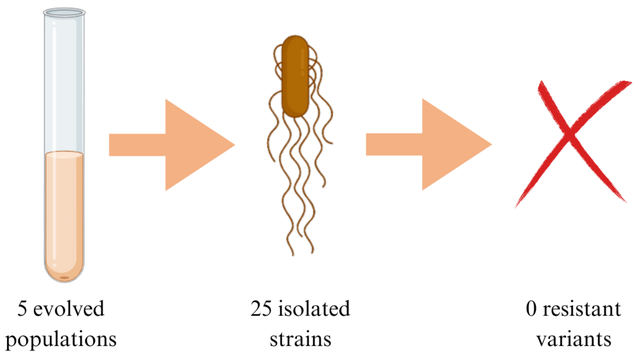


Figure 1. Selection of resistant variants from PAA -exposed populations.

The 5 **evolved populations** showed a **10% increase in direct resistance** against PAA after a 30-day exposure to subinhibitory concentratrions.

Despite the increased resistance observed in the 5 evolved populations, **no resistant variants to PPA were detected among 25 isolated strains selected**
This phenomenon is consistent with previous findings described by Lopatkin *et al.* (2021).

5 CONCLUSIONS

Prolonged exposure to subinhibitory doses (1/2 MIC) of PAA leads to increased resistance in populations but this is not translated into the isolation of resistant variants. However, more studies are needed to understand in depth the relationship between the use of PAA and the emergence of resistance to biocides.

In addition, it would be interesting to evaluate the occurrence of cross-resistance to antibiotics or food preservation methods, as well as other methodologies of adaptive laboratory experiments, such as lethal treatments.

6 KEY REFERENCES

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