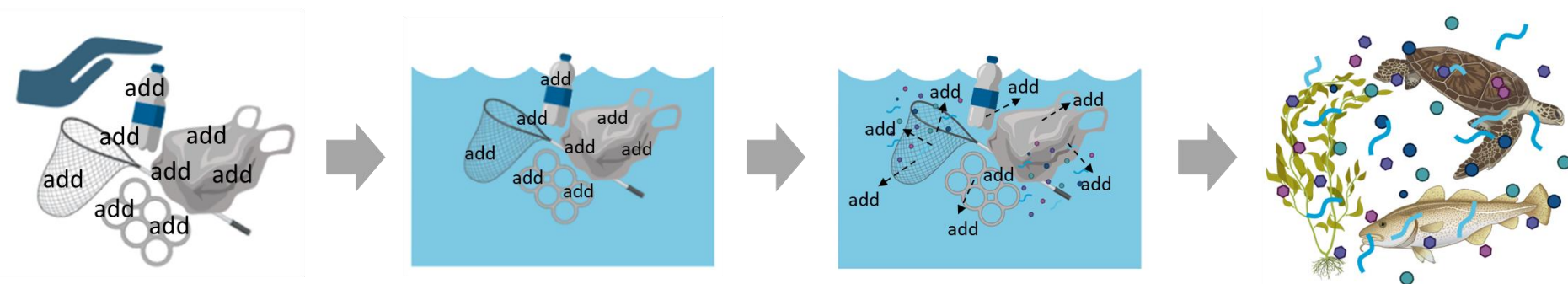


## ABSTRACTS

Plastic waste entering the ocean releases harmful substances and poses ecotoxicological risks. Despite this, research on the toxicity of plastic leachates is limited. This study prepared leachates from various virgin plastics and assessed their toxicity on the marine microalgae *Skeletonema costatum*. The plastics included petroleum-based (PS, PP, PVC) and biodegradable types (scPHA, PBEAS). 500 mg of this plastic powder was placed into 50 mL bottles, filled with filtered natural seawater. Leaching conducted in a dark room at 20°C with a rotator mixer for 24 hours, 72 hours, and one week. Leachates were filtered with glass microfiber filters (Whatman, GF/F, 0.7 μm) and exposed to *S. costatum* for 72 hours. The results showed that PS and PVC leachates leached for one week reduced the growth rate of *S. costatum*. Unlike PS and PVC, PP had non-toxic effects observed in leachates. Between biodegradable plastics, only PBEAS leachates (24 hours, 72 hours and one week) caused growth inhibition, while scPHA showed no toxic effects. This study is significant as it establishes a protocol for preparing plastic leachates and conducting toxicity assessments, demonstrating that certain leachates can inhibit the growth of marine microalgae.

## INTRODUCTION

With the annual increase in plastic production, a significant amount of plastic waste enters the ocean. In marine environments, plastics can release various harmful substances (additives such as plasticizers and flame retardants), raising ecotoxicological concerns. However, studies on the toxicity of plastic leachates are relatively scarce.



## PURPOSE

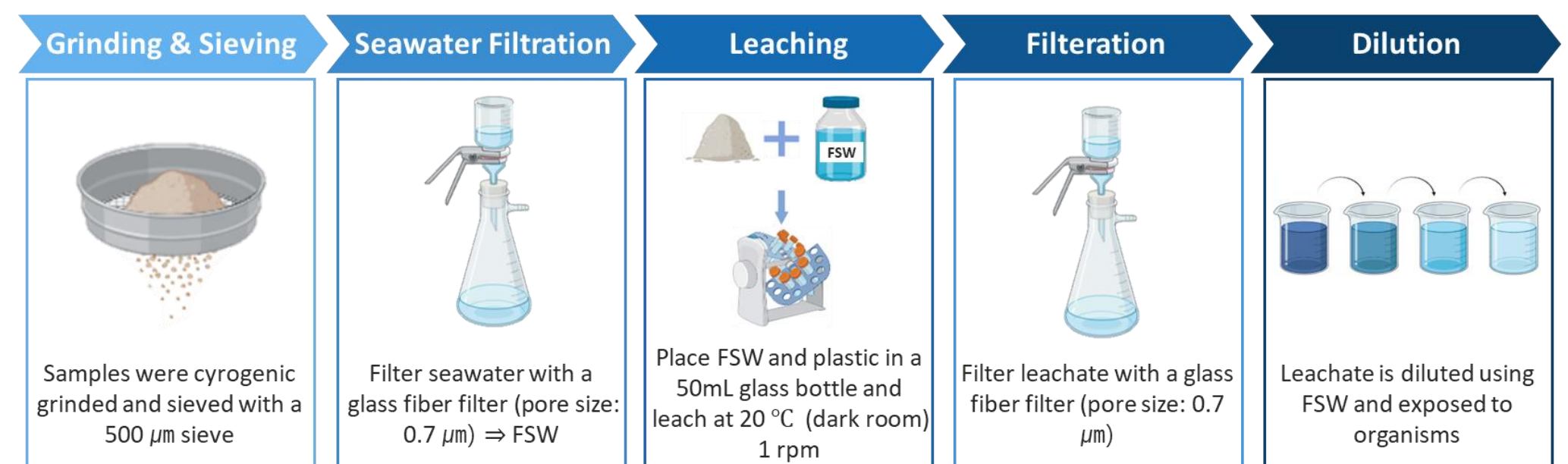
This study aims to investigate the adverse effects of leachates from various virgin plastics (plastic before manufacturing) on the marine microalgae *Skeletonema costatum*.

## MATERIALS AND METHODS

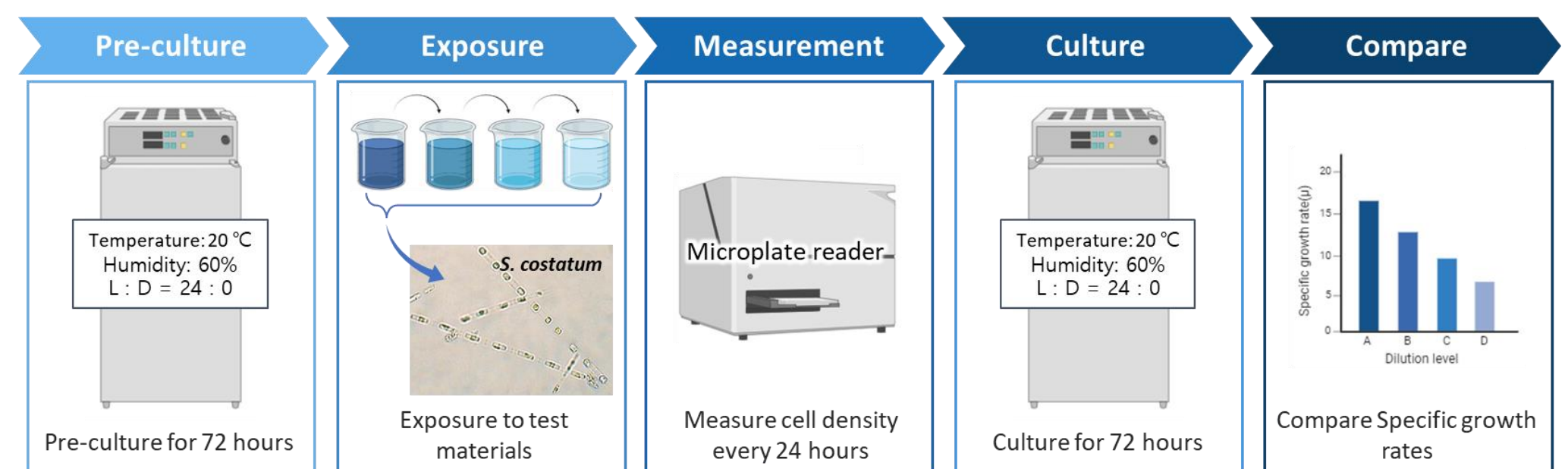
### Preparation of leachates from virgin plastic materials

Test materials		
Petroleum-based plastic	PS	Polystyrene
	PP	Polypropylene
	PVC	Polyvinyl chloride
Biodegradable plastic	scPHA	semi crystalline Polyhydroxyalkanoate
	PBEAS	Polybutylene succinate co-butylene adipate co-ethylene adipate

- Leaching medium: natural seawater
- Solid-liquid ratio: 10 g/L
- Leaching time: 24 hours, 72 hours, 1 week
- Mixing conditions: 20°C, dark room, 1 rpm

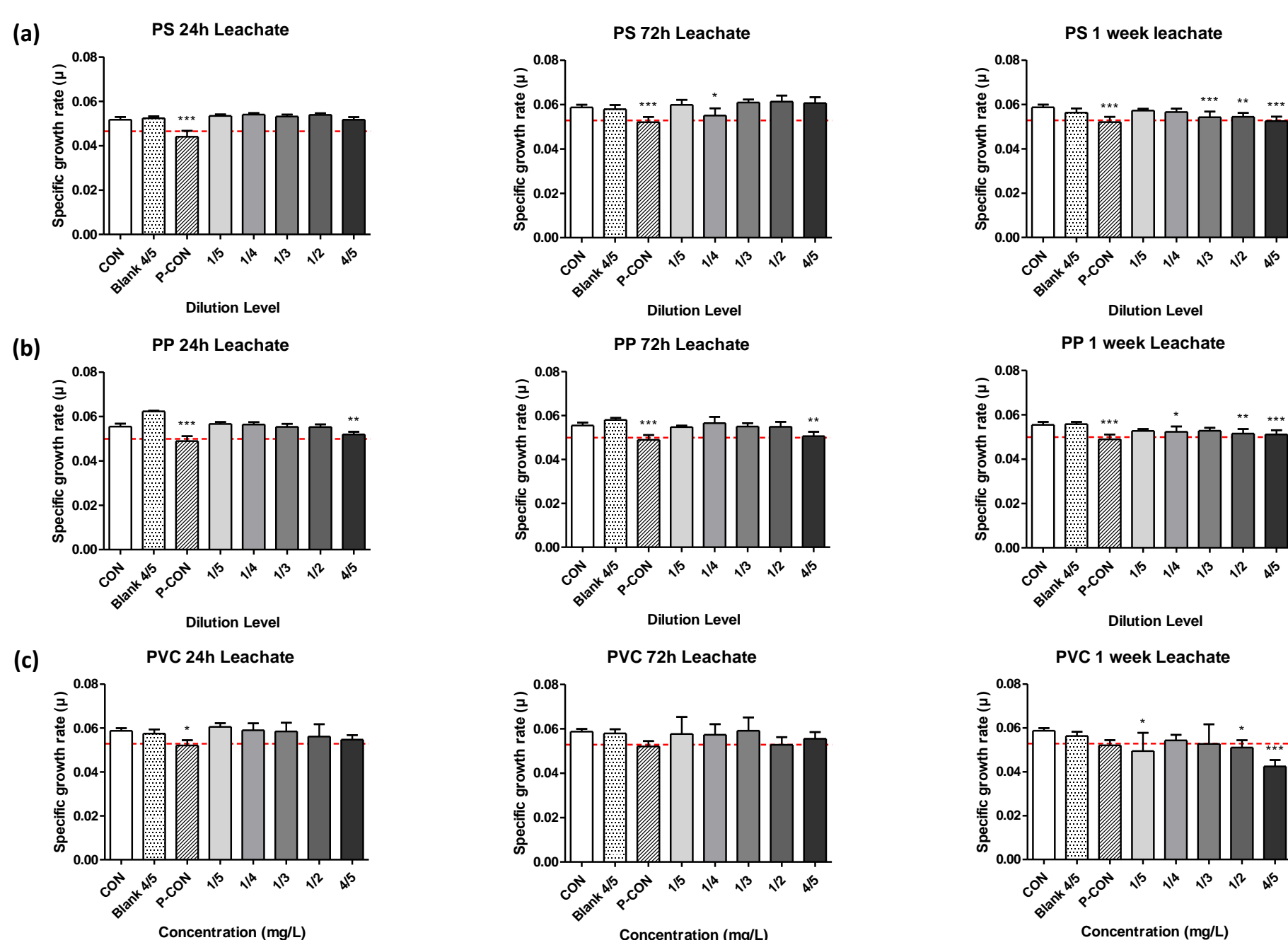


### Ecotoxicity testing with *S. costatum*



## RESULTS

### Ecotoxicity test of petroleum-based plastic leachates



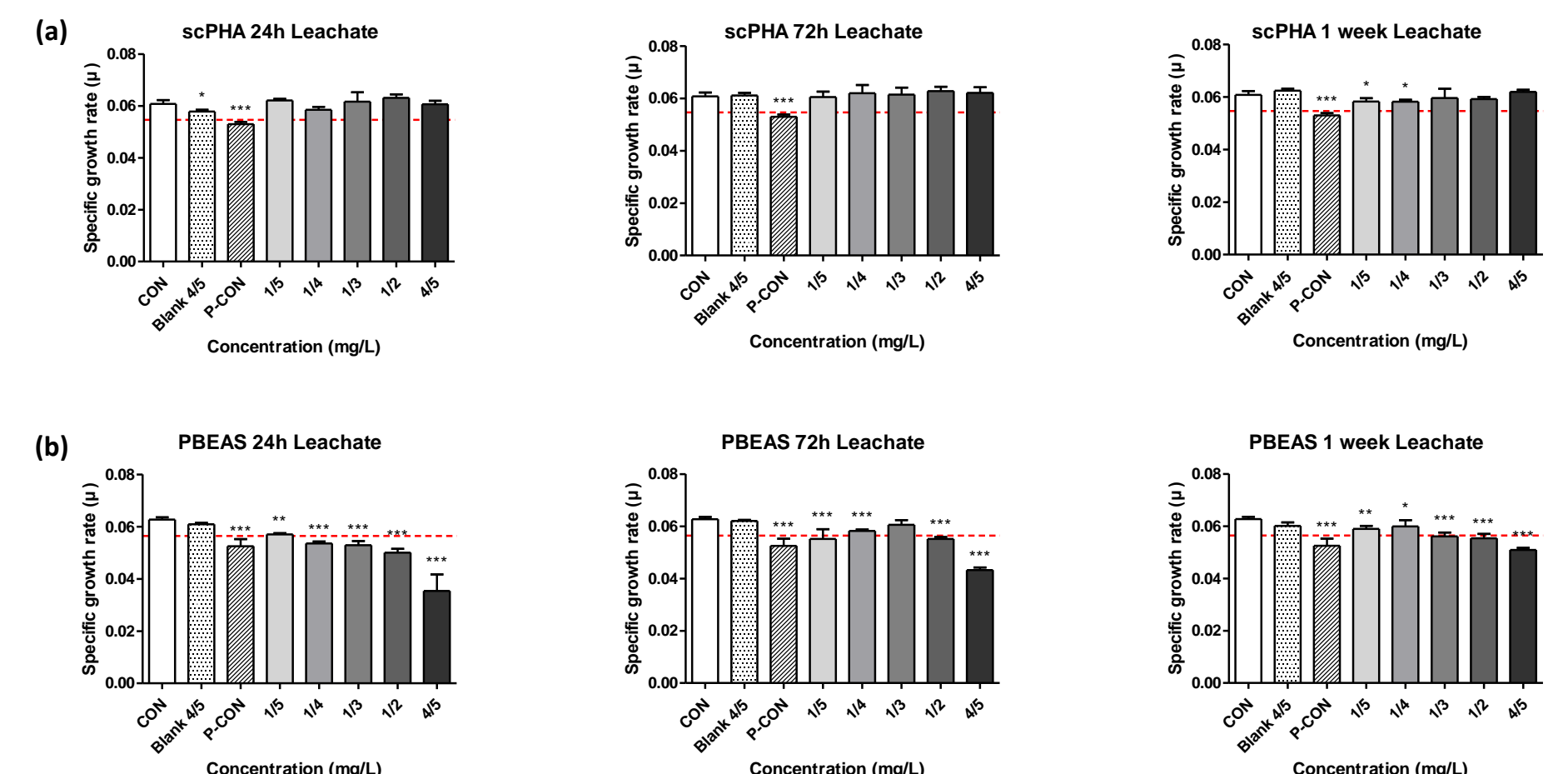
**Figure 1. Specific growth rates of *S. costatum* after exposure to different dilution level of PS, PP and PVC leachates for 72 hours.** The data are mean ±SD values of triplicate cultures (n=3, exception control: n=6). P-CON is potassium dichromate 1.25 mg/L (IC<sub>50</sub>). Blank control was performed at the minimum dilution level of 4/5. Asterisks represent significant differences between concentrations (One-way ANOVA Dunnett multiple comparison) (\*<0.05, \*\*<0.01, \*\*\*<0.001). (a) Specific growth rates of *S. costatum* exposed to different dilution levels of PS leachates. (b) Specific growth rates of exposed to PP leachates. (c) Specific growth rates exposed to PVC leachates.

The growth rates of *S. costatum* showed no reduction when exposed to the PP leachates. But the specific growth rate of the *S. costatum* was reduced when exposed to one week leachates of PS and PVC in the conducted tests.

## DISCUSSION

- The inhibitory effect of *S. costatum* was measured after exposure to assess the impact of virgin plastic leachates. The exposure results indicated that all three petroleum-based plastics decrease in the growth rate of *S. costatum*, whereas among the biodegradable plastics, only PBEAS exhibited growth inhibition, with scPHA observing no adverse effects.
- This study revealed that the tested polymers may have different effects on *S. costatum*, depending on their type. Virgin plastics are not a pure polymer because some chemicals and metals are added during the polymerization and molding. Furthermore the differences in toxicity among the samples could be attributed to variations in metal content within the plastics. Eriksen et al. (2018) reported various metal and metalloid contents in virgin plastics (PET, PP, PS, PE) that do not contain organic additives. Therefore, further studies should include chemical analyses of the leachates. Techniques such as ICP-MS and GC/MS should be employed to determine the presence of metals and organic additives in the leachates.
- And a glass fiber filter is used to remove microplastics after leaching, which cannot remove nanoplastics. Therefore, nanoplastics may be included in leachates, and a process of quantifying and separating the nanoplastics in leachates is required.

### Ecotoxicity test of biodegradable plastic leachates



**Figure 2. Specific growth rates of *S. costatum* after exposure to different dilution level of scPHA and PBEAS leachates for 72 hours.** The data are mean ±SD values of triplicate cultures (n=3, exception control: n=6). P-CON is potassium dichromate 1.25 mg/L (IC<sub>50</sub>). Blank control was performed at the minimum dilution level of 4/5. Asterisks represent significant differences between concentrations (One-way ANOVA Dunnett multiple comparison) (\*<0.05, \*\*<0.01, \*\*\*<0.001). (a) Specific growth rates of *S. costatum* exposed to different dilution levels of scPHA leachates. (b) Specific growth rates of exposed to PBEAS leachates.

When exposed to scPHA leachates the specific growth rate was observed no adverse effect on *S. costatum*. In the case of all PBEAS leachates, it was observed that the growth inhibition.

## CONCLUSION

- This study introduces a method for assessing the toxicological effects of virgin plastic leachates. A methodology for leaching medium, solid-liquid ratio, leaching time and mixing conditions was established. Using this method, experiments were conducted with petroleum-based (PS, PP, PVC) and biodegradable (scPHA, PBEAS) plastics.
- *S. costatum* was exposed to leachate, and the toxic effect of each plastic leachate was measured. The results were different for each type of plastic, which means that the type of polymer may have different effects on *S. costatum*.
- Further research needs to include a nanoplastic separation process and a chemical analysis of leachate to minimize the effects of nanoplastic contained in leachate.

## REFERENCE

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