

# Clean Biocide Project: Natural Corrosion Inhibitors

## Halophilic Plant Extracts for Biofilm Mitigation

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### Background

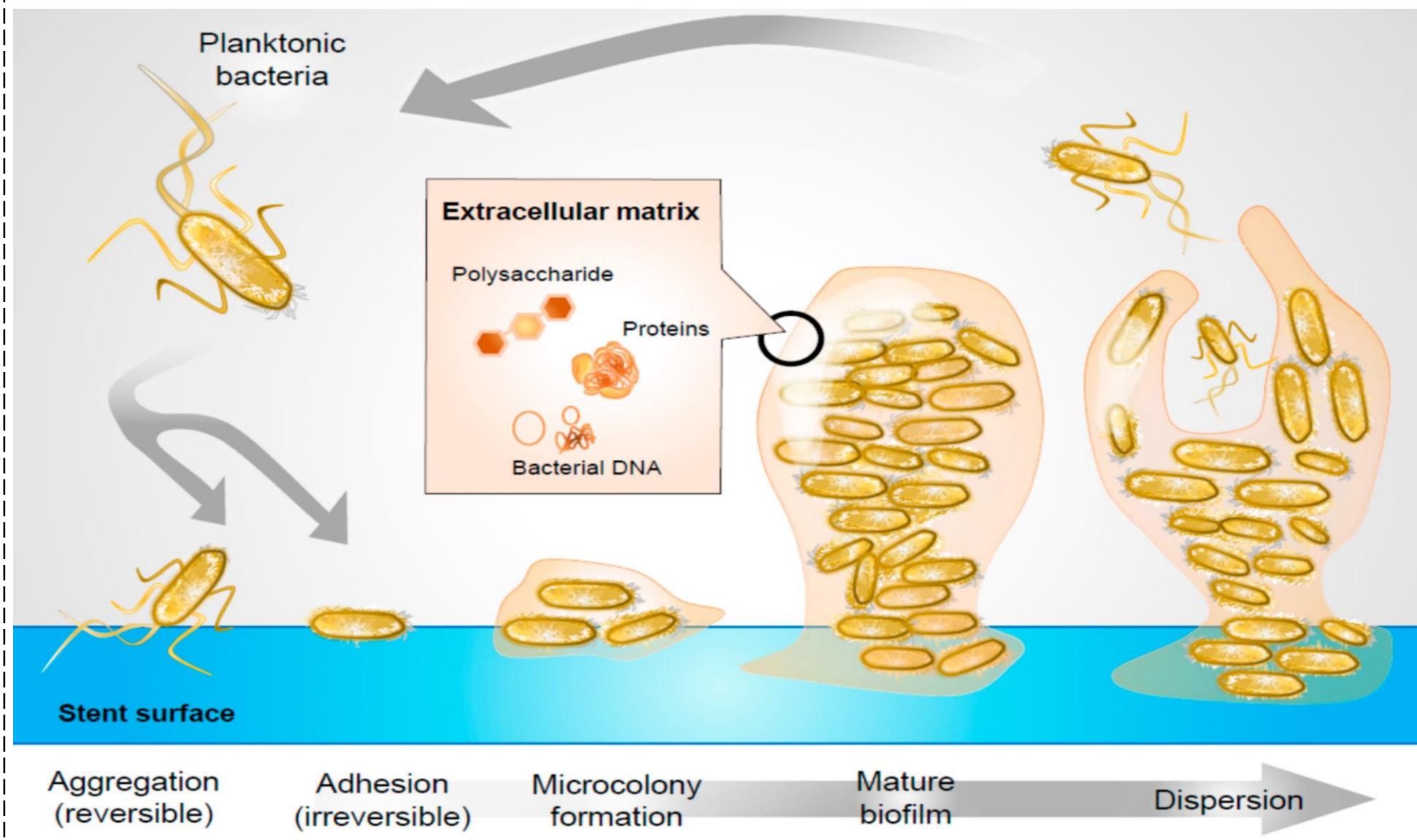
#### Biocorrosion

Pumping seawater into reservoirs has become a common way to enhance oil recovery by artificially maintaining high reservoir pressure. Seawater, however, contains high concentrations of sulfate, setting the grounds for Microbiologically influenced corrosion (MIC).

MIC is caused by microorganisms using metals as part of their metabolism, corroding the metal over time. Sulfate-reducing bacteria's (SRB) metabolism corrodes iron while reducing sulfate to H<sub>2</sub>S, which is corrosive and highly toxic to humans. [1]

Corrosion is aided by the production of "biofilm". The biofilm functions as a selective barrier, providing protection while allowing nutrient transfer. [1]

**Biofilms are the main reasons why mitigation of MIC is difficult.**



Currently, MIC is mitigated in two ways:

- **Chemical removal:** Tetrakis-hydroxymethyl phosphonium Sulfate (THPS) or Glutaraldehyde, none of which are especially good at destroying biofilms.
- **Physical removal**, which often requires partly shutting down production.

### 16S rRNA Amplicon Sequencing

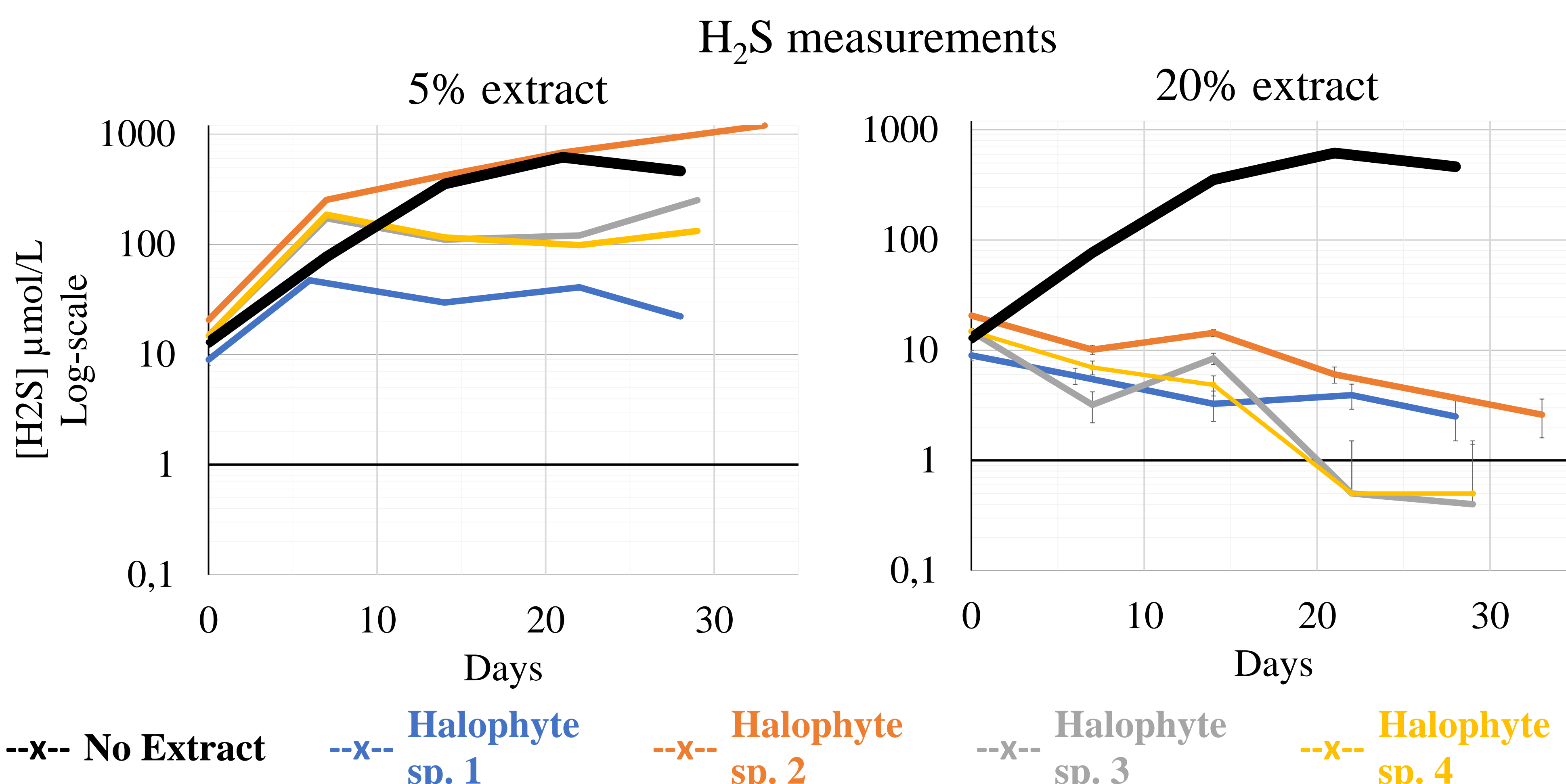
Sequencing the microbial species of a control sample and a sample with added extracts showed a decrease in abundance of **SRB species** and **methanogens**, which are thought to be important to biofilm formation.

BACTERIA GENUS	CONTROL	20% EXTRACT
Clostridium sensu stricto 13	18%	5% ✓
Methanosarcina	8%	0% ✓
Terrisporobacter	7%	1% ✓
Vibrio	6%	23% ✓
Desulfosporosinus	6%	0% ✓
Clostridium sensu stricto 1	4%	6% ✓
Paraclostridium	3%	4% ✓
Shewanella	3%	2% ✓
Clostridium sensu stricto 7	1%	38% ✓
OTHER	28%	14% ✓

### Screening Results

Each colored line represents a sample to which an extract has been added. The black line represents the control sample to which no extracts were added. The reduction in H<sub>2</sub>S is given in logarithmic scale.

The significant drop in H<sub>2</sub>S concentration suggests that the extracts are very effective at reducing the presence of SRBs.



### Halophyte Extracts

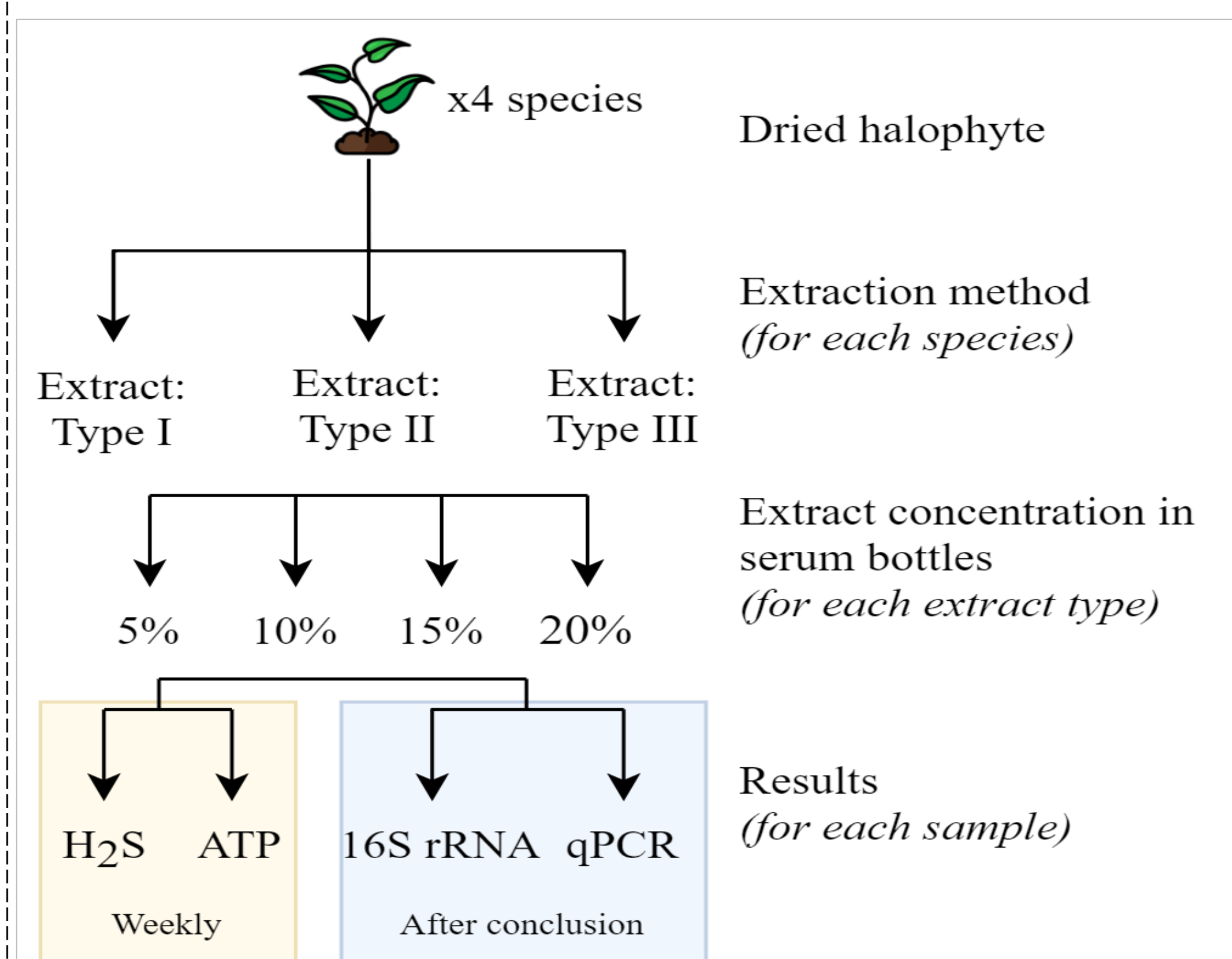
Halophytes (salt-tolerant plants), produce a variety of bioactive compounds and some of these compounds have antimicrobial activity. [2]

By varying the extraction method and -conditions, it is possible to maximize the amount antimicrobial properties of the extract, while reducing growth-promoting factors such as naturally present sugars.



### Screening of Halophyte Extracts

A series of fermentations were made to which various amounts of extracts were added. Extracts were produced from four different plants.



Screening of best-suited extract was made using 100 mL serum bottles with a rubber cap for sampling and injecting. Weekly samples were taken, and H<sub>2</sub>S and ATP concentrations were measured. The photo shows the ferment samples with extracts from one of the four halophyte species.

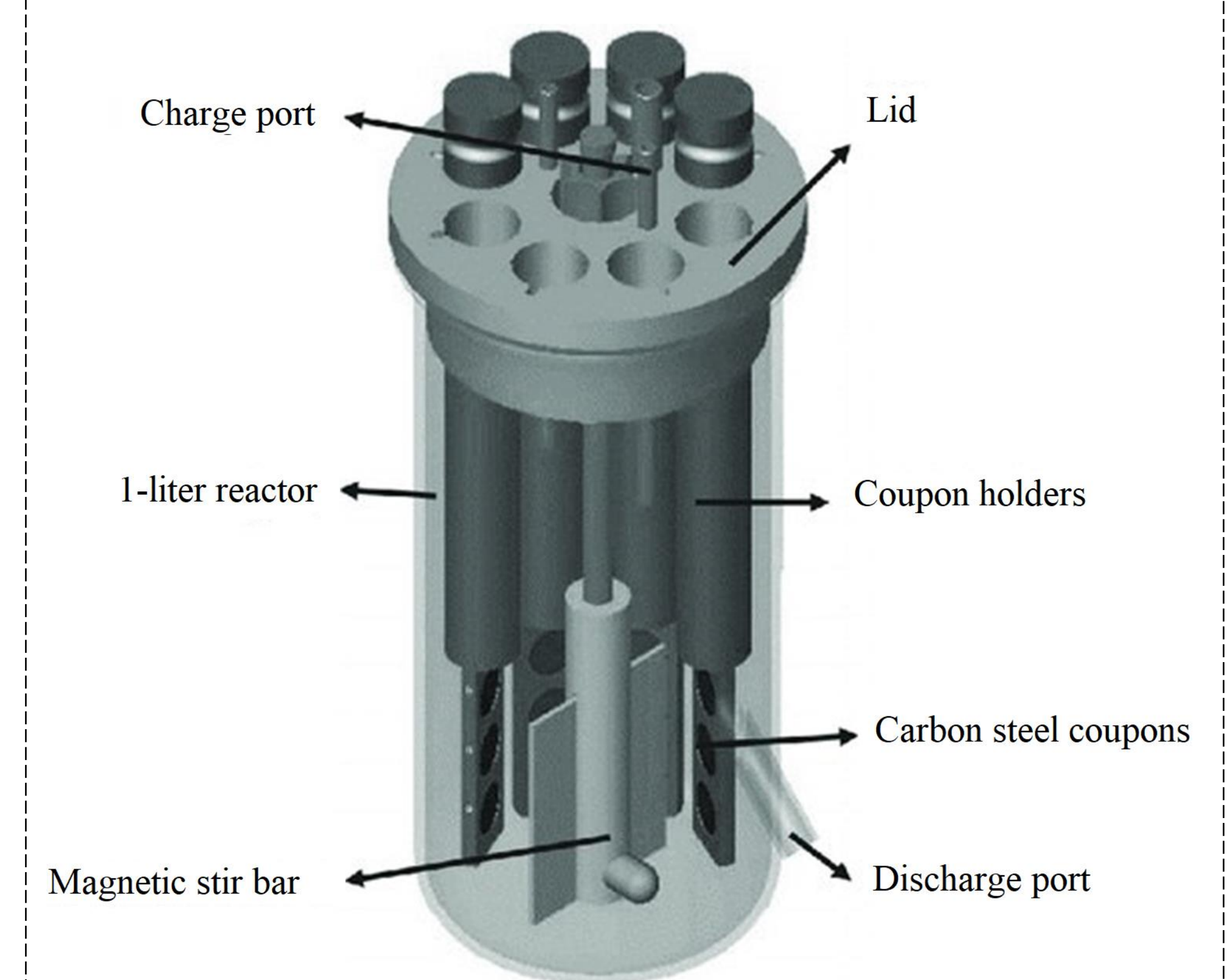


### Biofilm does not readily form in monocultures [3]

To mitigate this, the experiments were inoculated using anaerobic sediment dug from the Wadden Sea on the Danish west coast. Postgate medium was used as growth-medium, mimicking the chemical composition of produced water.

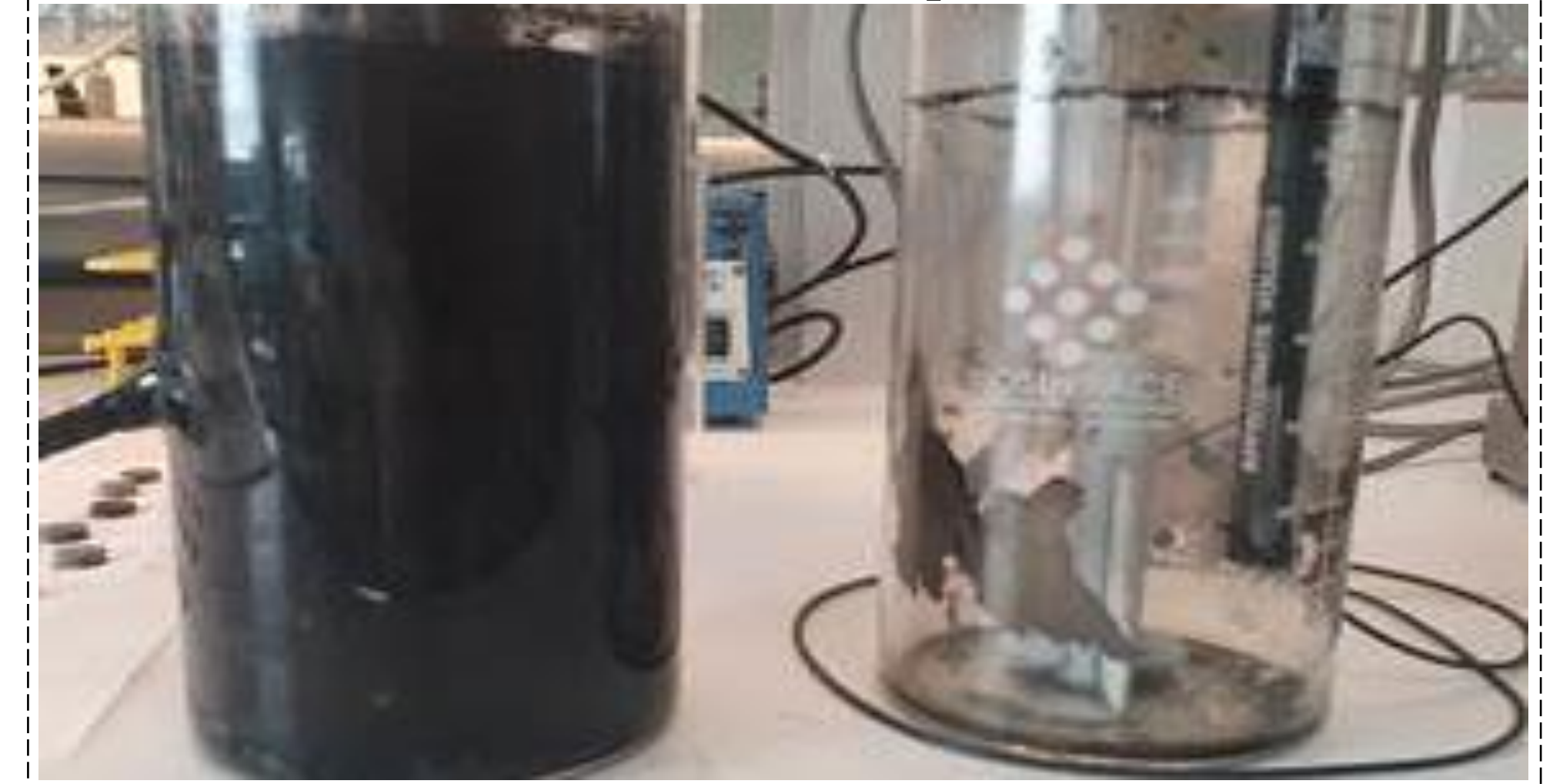


### Biofilm Reactor



The best performing extract from the screenings was scaled up to run in an anaerobe biofilm reactor. The reactor has rods designed for fixating corrosion coupons, which will get covered in biofilm. The medium inside the bioreactor was changed twice a week.

The bioreactor ran 14-days of building up biofilm followed by 14 days of adding biocides with the medium before the setup was taken down and corrosion was inspected in controls.



After the experiment had ended,

- the bioreactor without the extracts was covered entirely in biofilm. Steel coupons had a visible layer of corrosion.
- Then bioreactor where extracts were added only had remainders of biofilm. Steel coupons had no visible signs of corrosion.

### Acknowledgements and References

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- [1] Skovhus, T. L. et. al. (2017) "Microbiologically Influenced Corrosion in the Upstream Oil and Gas Industry". CRC Press.
- [2] Cybulska, I. et. al. (2014) "Phytochemical composition of some common coastal halophytes of the United Arab Emirates", Emirates Journal of Food and Agriculture, 26(12), pp. 1046–1056
- [3] Brileya, K. A. et. al. (2014) "Biofilm growth mode promotes maximum carrying capacity and community stability during product inhibition syntrophy" Front. Microbiol., vol. 5, no. DEC, p. 693