GAIN
Green Aquaculture Intensification

SUMMER SCHOOL

FROM AUGUST 30TH TO SEPTEMBER 3RD

This project receives funding from the European Union's Horizon 2020 research and innovation programme under grant agreement No 773330 (GAIN)

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Valorising shells

Leticia Regueiro ANFACO-CECOPESCA

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Historic examples of shellfi by-product valorisation

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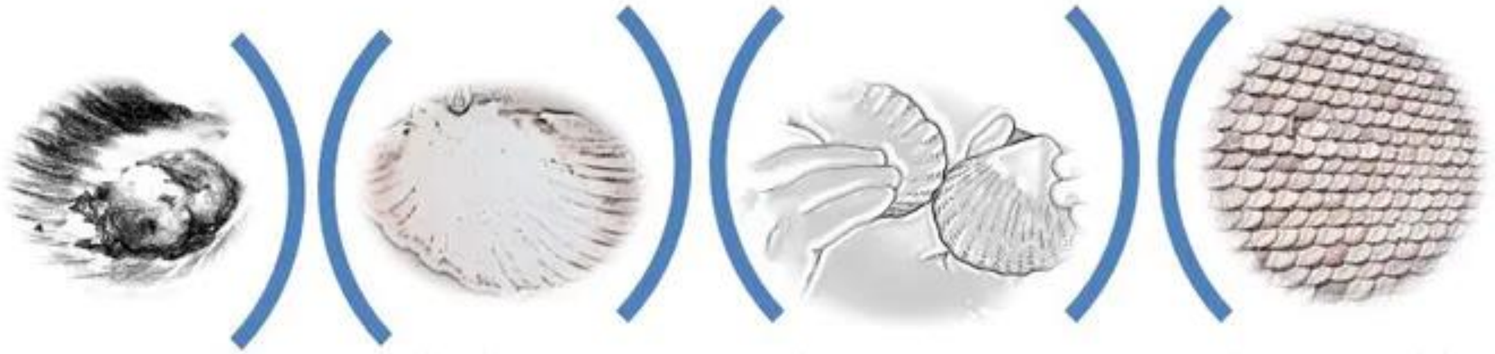
Scallop a circular economy example from 1000 years ago

Circular economy concept in scallops come from XII century.

Scallops have been used as: ashtrays, musical instrument, for cooking, amendment to the soil, drinking utensil and as construction material.



Construction

Shells from molluscs have been used in the construction in many coastal settlements around the globe.
For instance, shells have been used in construction in the Saloum Delta, Senegal as far back as 5000 years ago

Specifically “Tabby” is a form of concrete made by heat-treating shells to form lime, and then creating a mixture of lime, sand, water, ash and broken shells. Oyster shells have been traditionally used in this process. Tabby structures are still standing in Florida, Georgia and the Carolina’s, as well as parts of Spain and Southern England.

Construction

Tabby consists of: lime+shells+sand+water+ash

PAST USES: medicinal uses.

In Southern Africa, in the 19th Century, wearing a shell amulet was thought to help maintain health, fertility and luck. Oyster shell amulets coated in gold, silver, or electrum found in Egypt were known to have been worn during the Middle Kingdom as a form of functional jewellery that promoted good health.

British Museum

PAST USES: Jewellery

Mollusc shells and jewellery have been intricately linked through history. In fact, the oldest identified piece of jewellery in the world was discovered in Israel and is made from the shells of the sea snail *Nassarius gibbosulus*. This sea snail shell jewellery dates back to between 100,000 and 135,000 years ago

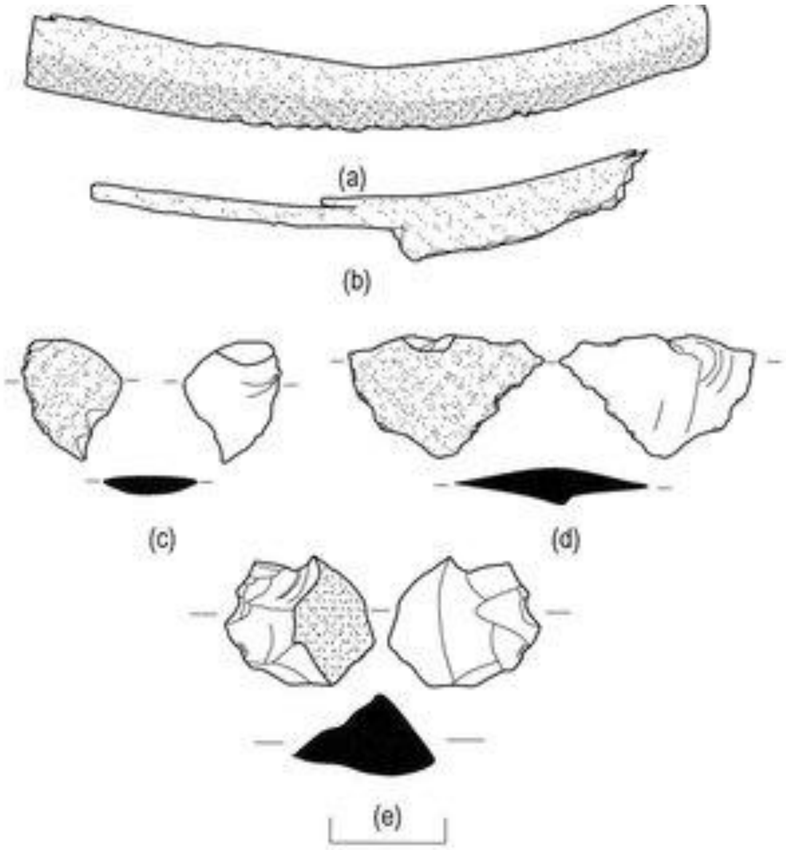
By 50,000 years ago, during the Upper Palaeolithic, shell adornments were common across Europe and Asia (Fernandez and Joris, 2007). Further, records of shell jewellery in Middle Palaeolithic archaeological sites have been used as evidence for the paradigm shift that early modern humans in the Levant and Africa were more behaviourally, socially, and culturally advanced than had been previously assumed.

However the shells as part of jewellery continues today.

PAST USES: Tools

There are countless records of the use of mollusc shells as tools. Many of these applications are centred on their weight bearing potential and toughness: this is testament to mollusc shell strength and hardness which must have been clearly understood throughout modern human history.

A study of a single archaeological site in Texas recovered over 3000 shell artefacts, many of which would have been used as tools for a variety of purposes including hammering, bevelling, chipping, chopping, and cutting. Other examples of mollusc shells as tools include the sea snail *Melo melo* whose common name is the “Bailer shell”, due to its use as a water bailing device in the canoes of native Australians. The shell is thought to have been used for many other tools, including as cooking pots by Australian Aboriginals.



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Current examples of shellfi
by-product valorisation

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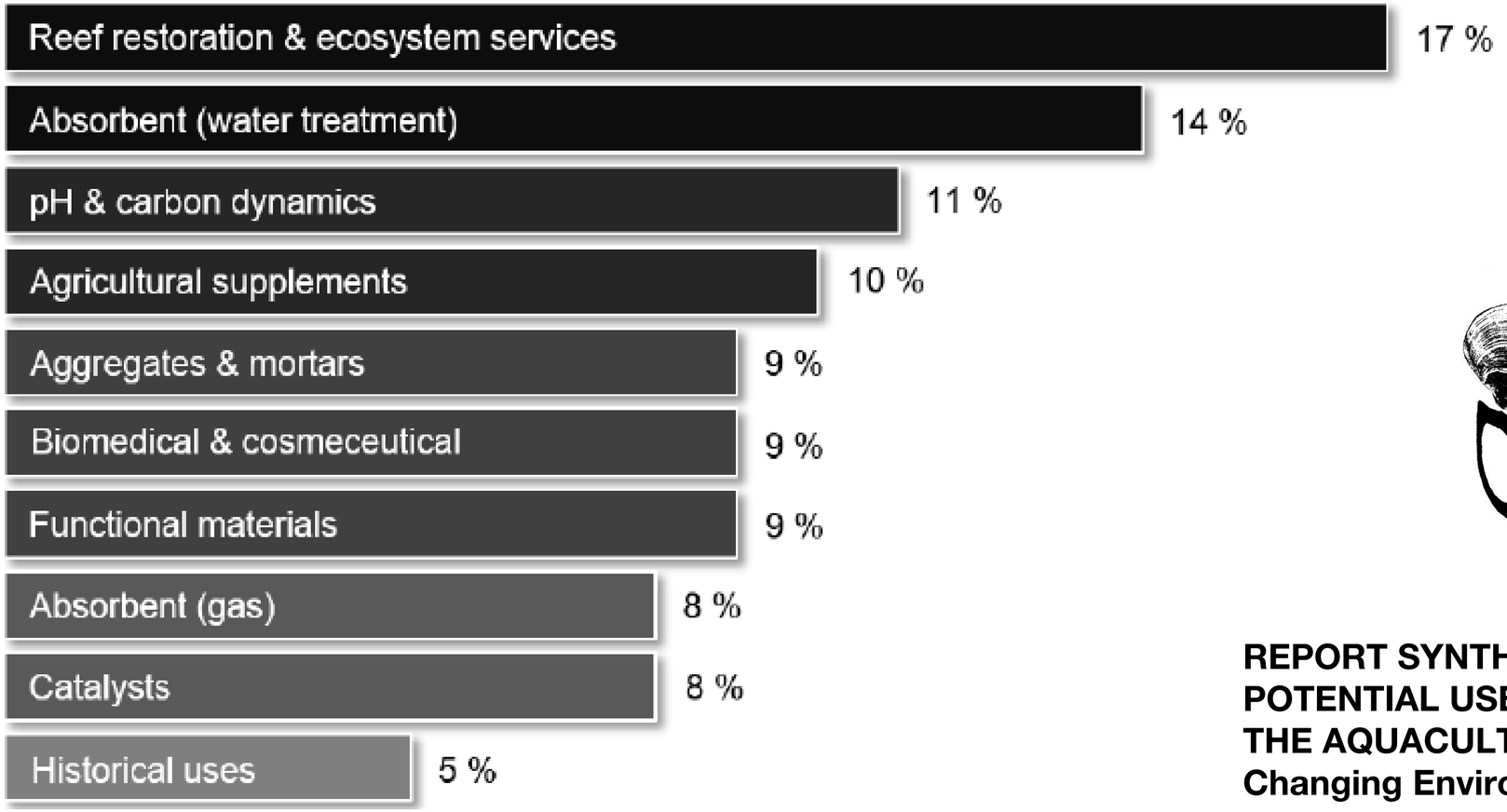


REPORT SYNTHESISING THE EXISTING AND POTENTIAL USES OF SHELLS AS BY-PRODUCTS OF THE AQUACULTURE INDUSTRY. CACHE (Calcium in a Changing Environment) PROJECT

https://zenodo.org/record/2662011/files/Shell_Waste_Report.pdf?download=1

Current uses: Livestock feed supplement

- Calcium supplementation is used to improve the health of livestock, particularly bone health

- Also in laying birds is used as a supplement to improve the quality and strength of egg shells

- Also some studies showed that it was an effective supplement for broiler chickens

References:

- Scott M.L., S.J. Hull, & P.H. Mullenhoff. 1971. The calcium requirement of laying hens and effects of dietary oyster shell upon egg shell quality. Poultry Sci., 50 (1971), pp. 1055-1063
- Miller, P. C. and Sunde, M. L. (1975). The effect of various particle sizes of oyster shell and limestone on performance of laying leghorn pullets. Poultry Science 54: 1422–1433
- Aletor VA, Onibi OE. Use of oyster shell as calcium supplement. Part 1. Effect on the utilization of gossypol-containing cotton seed cake by the chicken. Nahrung. 1990;34(4):311-8. doi: 10.1002/food.19900340403.
- Pizzolante, C. C., Saldanha, E. S. P. B., Laganá, C., Kakimoto, S. K., & Togashi, C. K. (2009). Effects of calcium levels and limestone particle size on the egg quality of semi-heavy layers in their second production cycle. Revista Brasileira de Ciência Avícola, 11(2), 79-86.
- Saunders-Blades JL, MacIsaac JL, Korver DR, Anderson DM. The effect of calcium source and particle size on the production performance and bone quality of laying hens. Poult Sci. 2009 Feb;88(2):338-53. doi: 10.3382/ps.2008-00278.

Current uses: Agricultural liming agent

- The second major market, including neutralisation of soils and metals bioremediation
- Liming is an important activity with several benefits such as reduce acidity and improve fertility.
- Liming action can be also a carbon sink mechanism
- The toxicity of the salt related to crushed shells was studied and no toxicity was demonstrated
- Use of crushed oysters improved soil status promoted microbial populations, increasing nutrient cycling

Chang Hoon Lee, Do Kyoung Lee, Muhammad Aslam Ali, Pil Joo Kim. 2008. Effects of oyster shell on soil chemical and biological properties and cabbage productivity as a liming materials. Waste Management, Volume 28, Issue 12, Pages 2702-2708. https://doi.org/10.1016/j.wasman.2007.12.005.

Current uses: De-icer grit

- Chlorine-based compounds, such as rock salt (NaCl), are commonly used as de-icing chemicals.

- Chlorine-based road grits could damage the urban environment due to their corrosive effects.

- Calcium magnesium acetate or any calcium acetate could be environmental-friendly potential alternatives as road grit

- The use of shells as calcium donor in the formation of calcium acetates could be also an alternative use for shells valorisation.

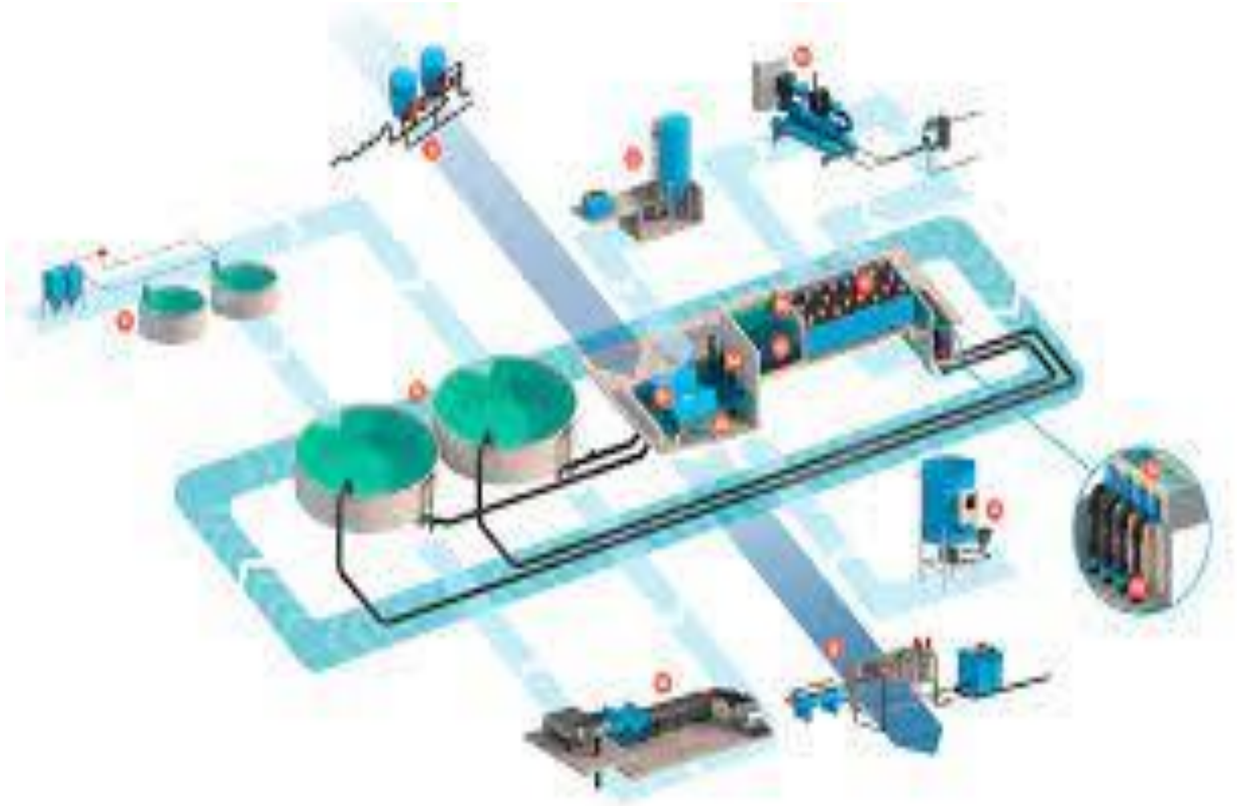
Hartl, W., & Erhart, E. (2002). Effects of potassium carbonate as an alternative road de-icer to sodium chloride on soil chemical properties. Annals of applied biology, 140(3), 271-277.

Current uses: Water treatment

The main use of shells in water treatment is for heavy metal removal in wastewater treatment facilities, since the calcium carbonate rich powder is good as a lead (Pb) sorbent, whilst the use of mixed shells rich in both calcite and aragonite should be optimised for heavy metal removal. Another study indicated that the shell dust coming from invasive snail (*Physa acuta)* could act as an effective cadmium (Cd) sorbent.

Besides heavy metals, calcareous shells have been previously tested as nitrate, sulphate and/or phosphate sorbents. For instance, the use of shells for carbon sequestration has been studied by several authors, highlighting that a previous pre-treatment via calcination or pyrolisation to convert the shells in calcium oxide (CaO) is required.

- Du, Y., Lian, F., & Zhu, L. (2011). Biosorption of divalent Pb, Cd and Zn on aragonite and calcite mollusc shells. Environmental Pollution, 159(7), 1763-1768.
- Hossain, A., & Aditya, G. (2013). Cadmium biosorption potential of shell dust of the fresh water invasive snail Physa acuta. Journal of Environmental Chemical Engineering, 1(3), 574-580.
- Ma, K. W., & Teng, H. (2010). CaO powders from oyster shells for efficient CO2 capture in multiple carbonation cycles. Journal of the American Ceramic Society, 93(1), 221-227.
- Castilho, S., Kiennemann, A., Pereira, M. F. C., & Dias, A. P. S. (2013). Sorbents for CO_2 capture from biogenesis calcium wastes. Chemical Engineering Journal, 226, 146-153.
- Monneron-Gyurits, M., Joussein, E., Soubrand, M., Fondanèche, P., Rossignol, S. (2018). Valorization of mussel and oyster shells toward metakaolin-based alkaline activated material. Applied Clay Science 162, 15–26



Current uses: Aquaculture water treatment

-Suitable material to remove nutrients from wastewaters discharged to natural environments

-Some studies showed the possibilities to be used in RAS systems

-Other studies showed the efficiency for phosphorous removal mainly using calcined shells

Yen, H. Y., & Chou, J. H. (2016). Water purification by oyster shell bio-medium in a recirculating aquaponic system. Ecological Engineering, 95, 229-236.
Buzin, F., Dupuy, B., Lefebvre, S., Barille, L., Haure, J. (2015). Storage of Pacific oysters Crassostrea gigas in recirculating tank: ammonia excretion and potential nitrification rates. Aquacultural Engineering 64, 8-14.

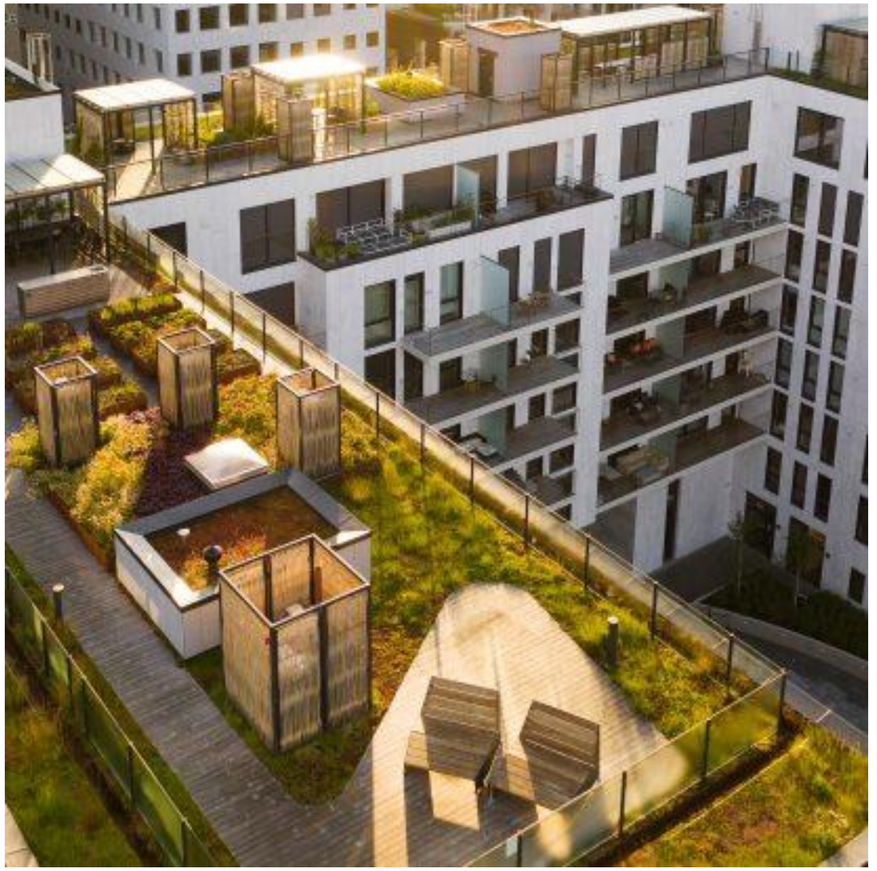
Current uses: Green roofing

- Another potential use in the building industry is the incorporation of shells into green roofing structures

- They can help with the neutralisation of acid rain, and the reduction of heavy metal contamination in drainage water

- There are two forms of green roofs: extensive and intensive.

- Both types of roof are designed with the same principal layers: vegetation, growing medium, filter membrane, drainage layer, root barrier, waterproofing membrane.



Current uses: shells in the building sector

- Whole oyster shells are used for simple wall structures in coastal villages in China, and crushed scallop shells have been used as a simple path aggregate on the Island of Mull in Scotland.

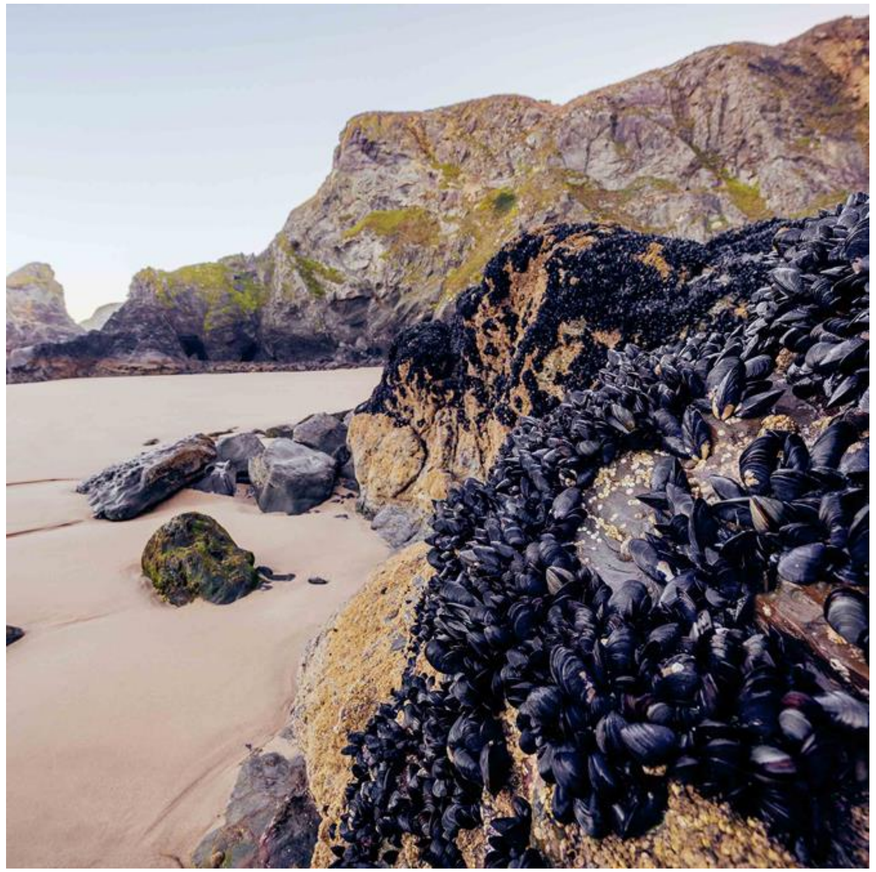
- In France, a study investigated the incorporation of crushed *Crepidula* shells into pervious concrete mixes and concluded that shell incorporation did not have an adverse effect on the concrete's mechanical strength, and increased porosity allowed for better water permeability, an important characteristic of pervious concretes.

Current uses: shells in the building sector

- More recently, In Spain, Galician mussel shells have been tested for their suitability in aggregate mixes.

- Biovalvo project (https://proyectobiovalvo.wordpress.com/) carried out by the University of Coruña has demonstrated that mussel shells thermally processed at 135°C for 30 minutes can be used as aggregates for mass concrete. Percentages of substitution of up to 25% of natural aggregates by mussel shell aggregate (sand or gravel) are suitable for structural concrete.

- A study assessing the incorporation of mussel shell waste in Spain into mortars found that differences in particle microstructure between quarried limestone and mussel waste $CaCO_3$ resulted in mussel waste-derived mortars showing improved setting times and final strength.

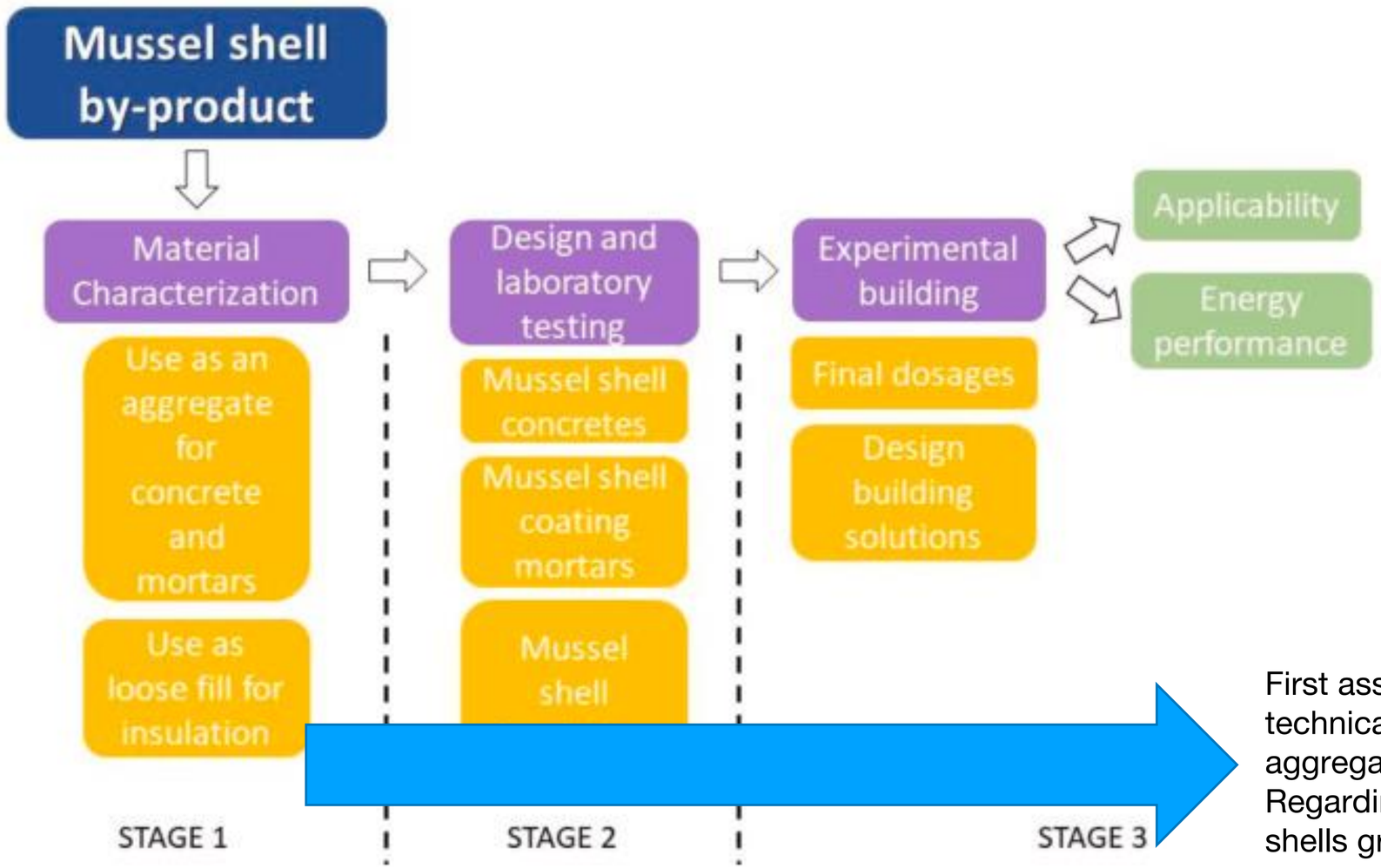


Ballester, P., Mármol, I., Morales, J., Sánchez, L. (2007). Use of limestone obtained from waste of the mussel cannery industry for the production of mortars. Cement and Concrete Research 37, 559–564.

BIOVALVO

The Biovalvo project constructed a house, located in Mariñeiros, featuring walls, floor and roof using up to 75% shell material in their composition

Since its construction, the short-term output of the project regarding resistance capacity and wall insulation seems to have been very satisfactory. Researchers are currently focused on monitoring long-term behaviour and the aging process of these mortars. To do this they are subjecting the material to extreme environmental temperatures, high and low, and significant humidity levels, in order to calculate the amount of shells needed for the structure to become completely resistant and durable.

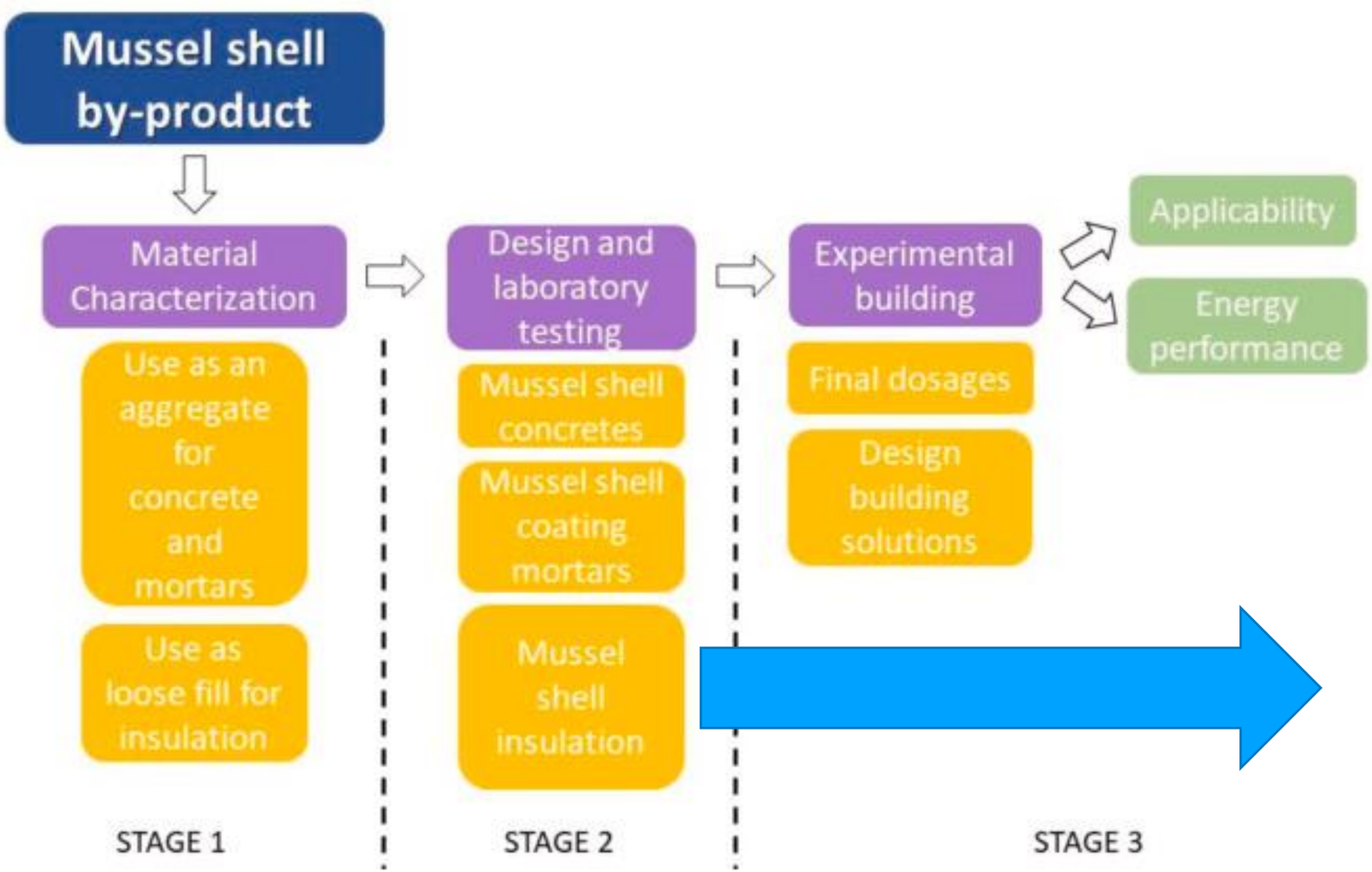


ELSEVIER
Journal of Building Engineering
Volume 44, December 2021, 102635

Assessment of mussel shells building solutions: A real-scale application

Carolina Martínez-García, Belén González-Fonteboa, Diego Carro-López, Juan Luis Pérez-Ordóñez

First assessments showed that there are no technical barriers to use mussels shells as aggregate in concretes.
Regarding their use as loose-fill material, mussel shells gravel is the best option.



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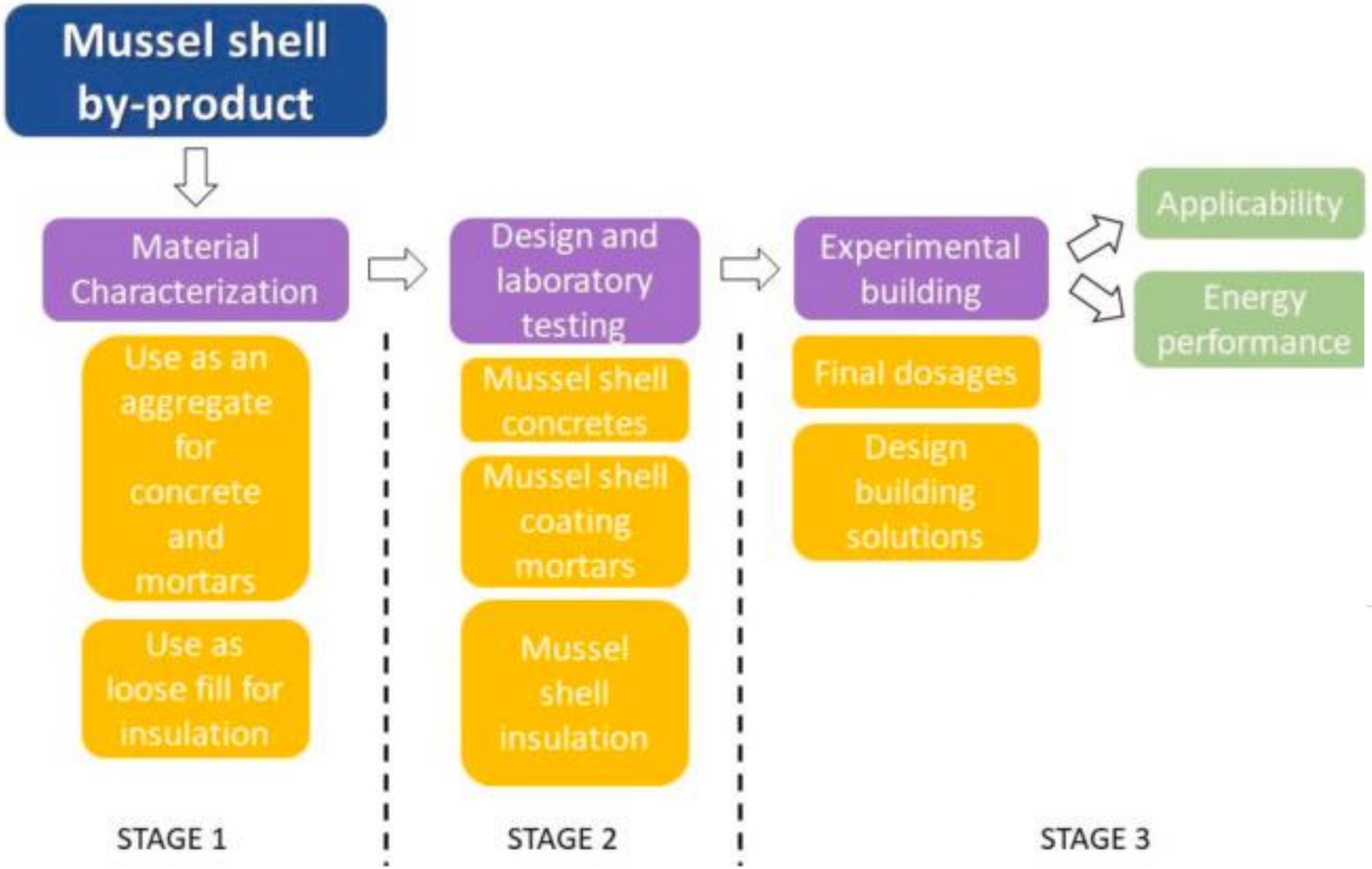
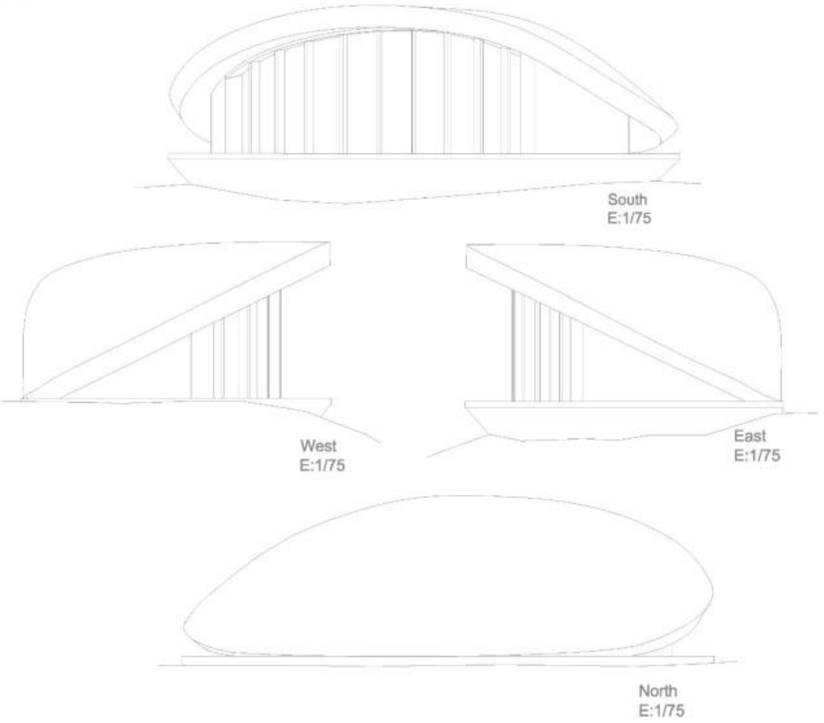
Results show the feasibility of mussel shell in cement coatings at an optimal replacement rate of 25% of the conventional aggregate.


Mussel shell by-product
Material Characterization
Use as an aggregate for concrete and mortars
Use as loose fill for insulation
Design and laboratory testing
Mussel shell concretes
Mussel shell coating mortars
Mussel shell insulation
Experimental building
Final dosages
Design building solutions
Applicability
Energy performance
STAGE 1
STAGE 2
STAGE 3


South
E:1/75
West
E:1/75
East
E:1/75
North
E:1/75

Calcium oxide catalysts

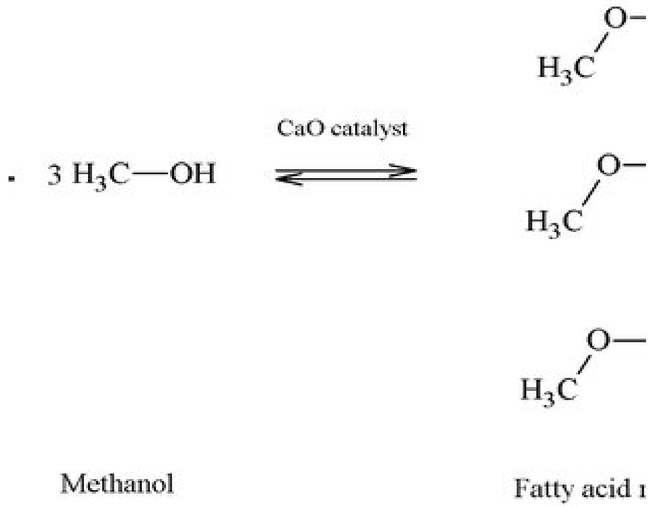
- Calcium oxide (CaO) is a widely used catalyst and chemical compound in a myriad of industrial applications. CaO can be formed via the calcination of $CaCO_3$

- Waste shells from aquaculture have been advocate as a low cost $CaCO_3$ source for the production of CaO for a variety of applications, particularly biodiesel production in developing countries.

- Oyster shells have been shown to be an effective heterogeneous catalyst in the conversion of soybean oil into biodiesel, whereas calcined waste cockle shells were good to catalyse the production of biodiesel from palm olein.

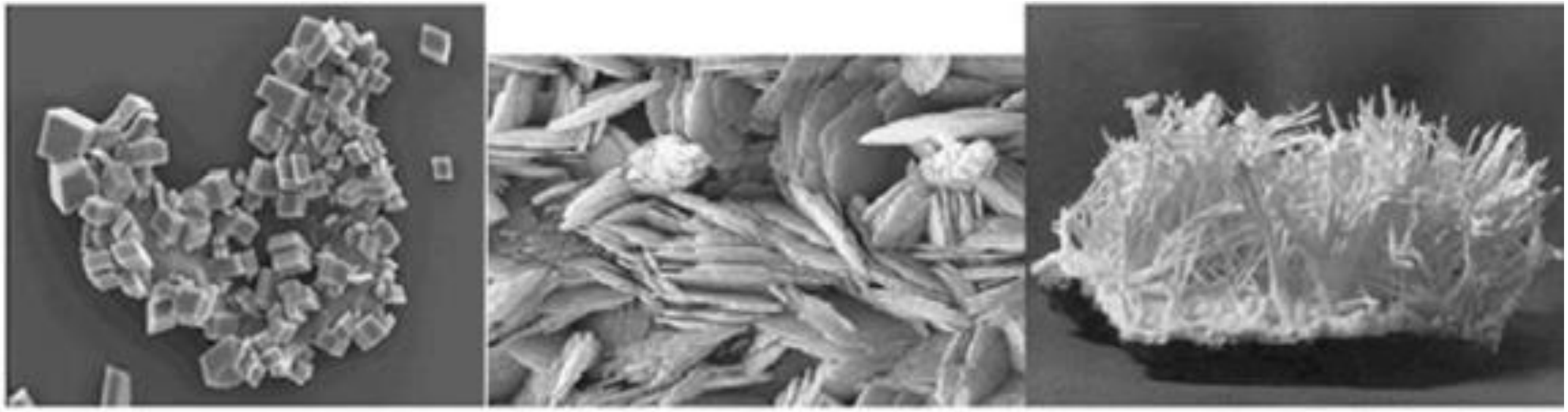
- Freshwater mussel shell was found to be a feasible catalyst in the conversion of Chinese tallow oil to biodiesel, and was able to catalyse 7 reaction cycles (Hu et al., 2011).

- Finally, oyster shell-derived calcium oxide was used to modify copper-based catalysts, and compared to commercial calcium oxide sources as a hydrogenation catalyst.



Biomedical and cosmeceutical

- Modern science has found that shells, particularly their organic protein matrix, can exhibit many properties that are of interest to the modern cosmetics and biomedical industry.
- The most widely discussed biomedical use for shells is in bone and tissue re-engineering
- The $CaCO_3$ powder acts as a substrate on which new osteoblasts can grow and secrete bone. For instance, nacre powder, from oyster *Pinctada maxima*, acted as a scaffold in bone reconstruction in human jaw defects, demonstrating their potential in bone engineering and augmentation applications.



Functional materials

- The previous slides have highlighted the potential for $CaCO_3$ shells to be converted to other compounds such as calcium oxide, calcium phosphate, or hydroxyapatite, for instance. These conversions open up further potential applications, and widen the potential uses of shells in industry.

- Nacre, in particular, has inspired the field of nanocomposite materials for several decades due to its calcium carbonate organic matrix layered arrangements.

- There are a number of articles highlighting the potential use of waste shells from aquaculture in polypropylene and polymer composites. Waste blue mussel (*Mytilus edulis*) shells were used as a base material for $CaCO_3$ filler in polypropylene manufacture.

Shells returned to the marine environment

- There is a growing body of evidence in scientific literature to suggest that shells are a valuable material from a biological perspective within the marine environment, and may provide and promote a variety of ecosystem services.

- Ocean alkalinisation has been proposed as a method of limiting atmospheric CO2 increases and ocean acidification through pH buffering.

- The project **Cquestrate** has attempted to apply this alkalinisation concept in scaled experiments, but has not had positive results or found workable solutions. This stands testament to the complexity of carbonate chemistry manipulation for CO2 sequestration techniques. Despite this, several studies have shown that more localised and confined systems that are affected by acidity could be treated in a simple and cost effective way by the addition of CaCO3

cquestrate

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SUMMER SCHOOL

Innovative processes studied in t GAIN Project: Shells as a subst for the production of seaweed seedlings

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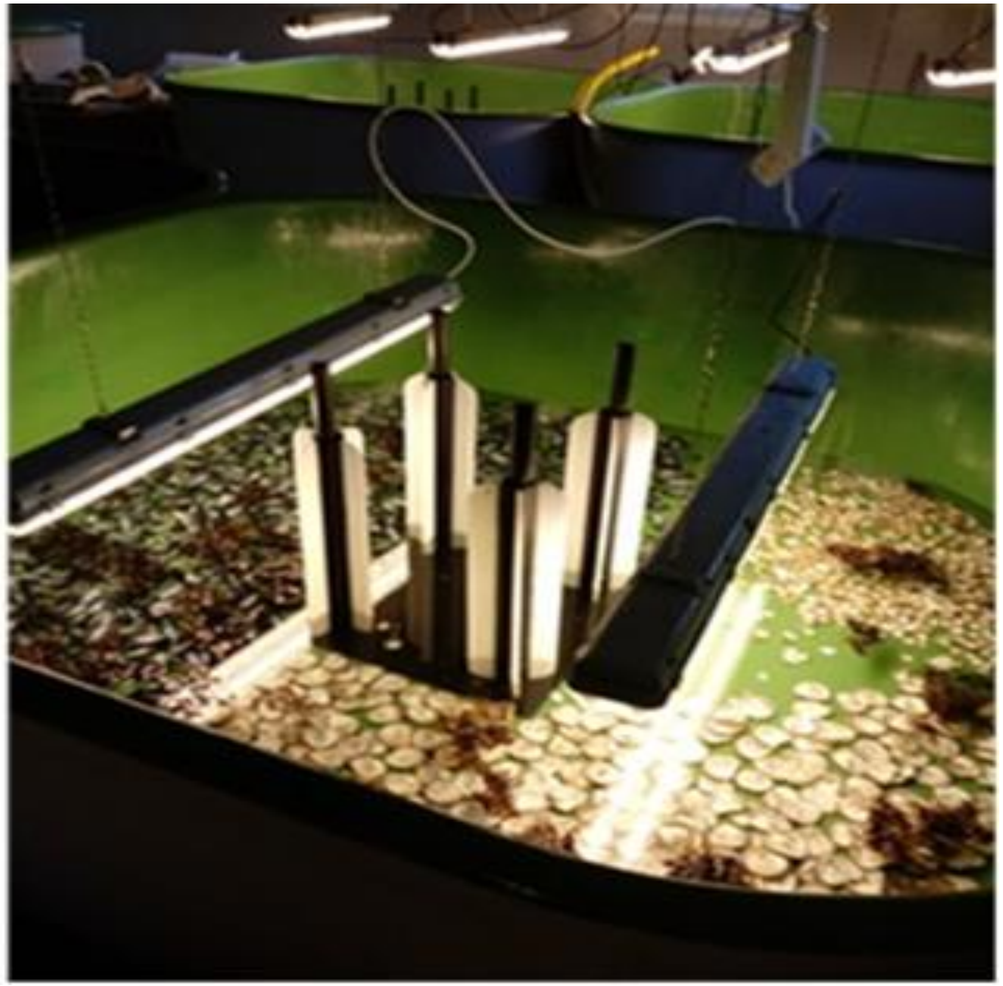
Substrate for algae

- Shells as substrate for cultivation of nori (*Porphyra spp*., *Pyropia spp*.), a high value seaweed.
- Industrial nori production relies on the availability of shells to cultivate conchocelis, a microscopic stage in the life cycle of nori algae.
- A typical Japanese seedling producer uses about 200,000 oyster shells to cultivate nori conchocelis for seedling production purposes.
- In Europe; no industrial nori production exists so far; however, a variety of local species with good food properties are native in European waters.
- Preliminary cultivation experiments at GAIN partner Salten Havbrukspark exhibit conchocelis growth on all six types of shells investigate.

Substrate for algae

In Gain experiments, cockle, oyster and blue mussel shells were inoculated with fertile *Porphyra umbilicalis* gametophyte, i.e. the conchocelis phase



Substrate for algae

- *Porphyra* conchocelis grew well in the tested shells. In the pictures you can recognize it as red to purple colour.

- Oyster shells deliver optimal results in terms of growth and ease of handling.

Substrate for algae

Conchocelis also grew on other shells, however, for practical handling, especially for the physical removal of contaminants, blue shells are too fragile and cockles too small

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Innovative processes studied in GAIN Project: shells as biofiter medium

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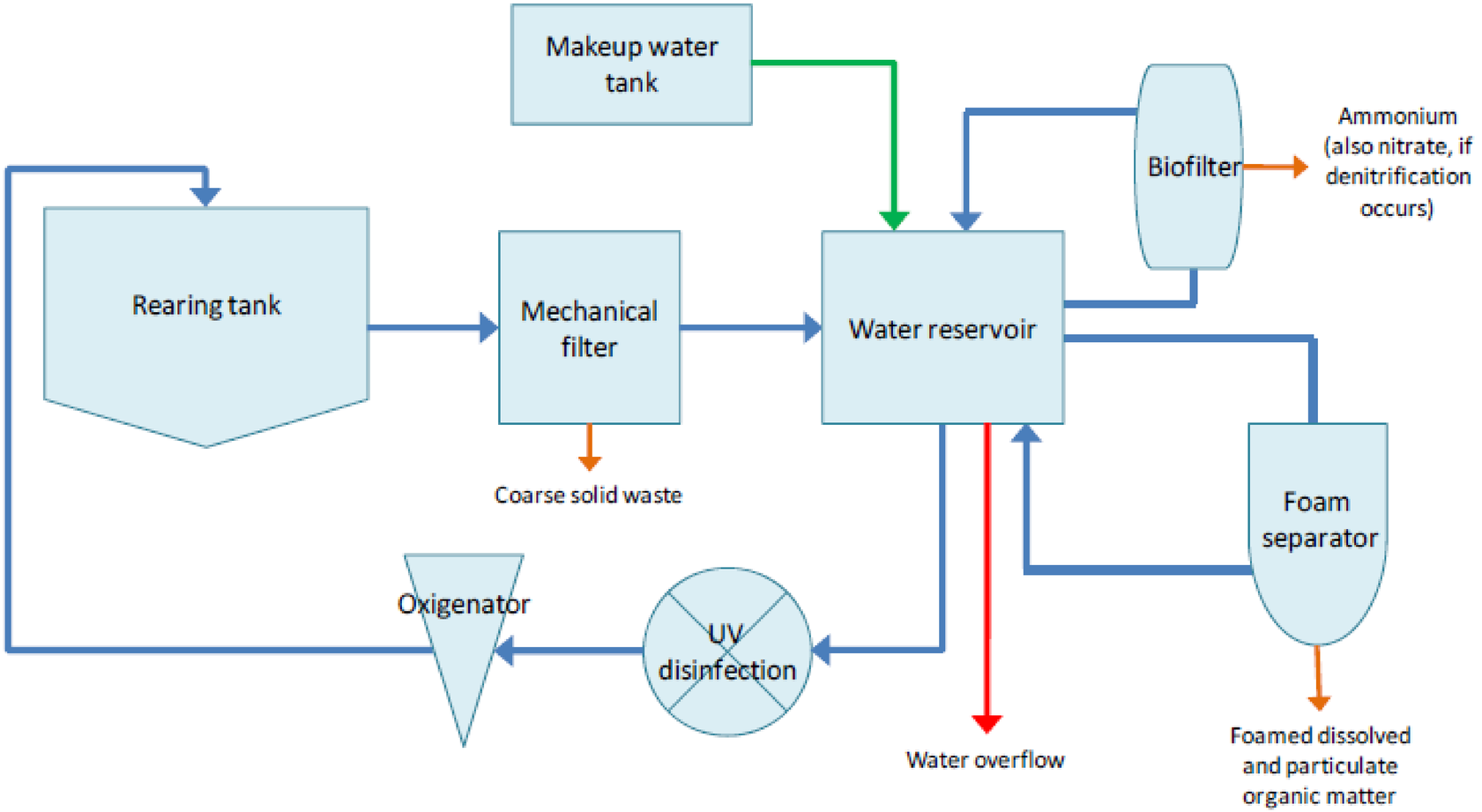
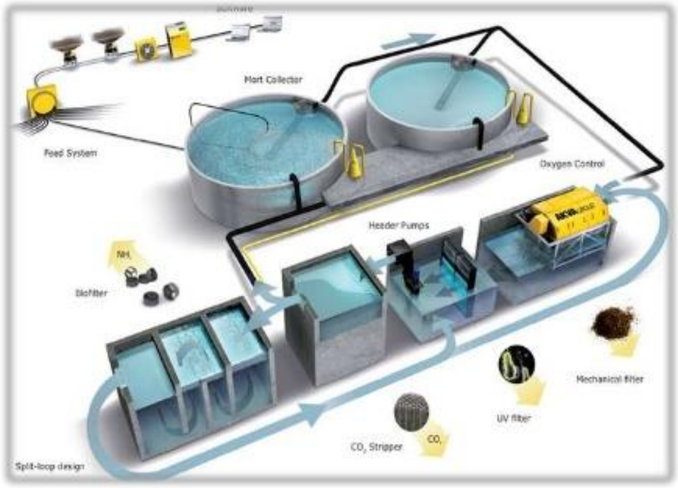
RAS


Makeup water tank
Biofilter
Ammonium (also nitrate, if denitrification occurs)
Rearing tank
Mechanical filter
Water reservoir
Coarse solid waste
Foam separator
Oxigenator
UV disinfection
Water overflow
Foamed dissolved and particulate organic matter

Reduce the use of plastics by using mussel shells as biofilter media in RAS systems

RECIRCULATORY AQUACULTURE SYSTEM



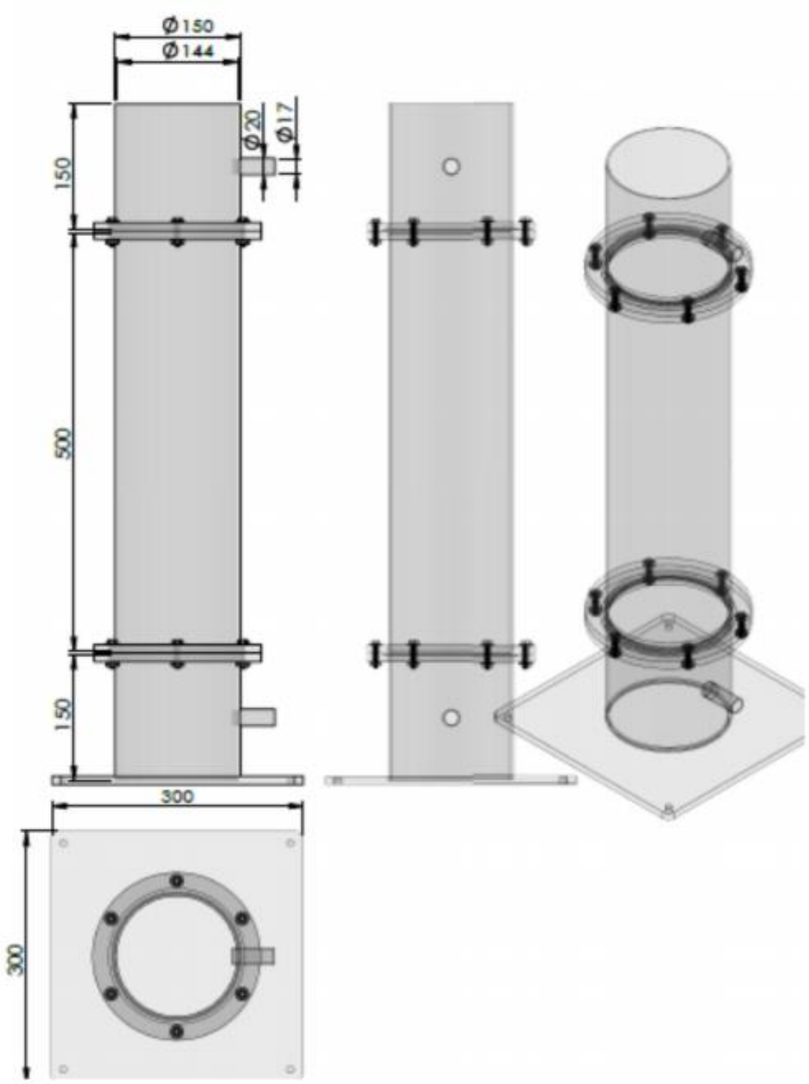
COMPARISON LAB SCALE

VS

USUAL

GAIN

Biofilter design



Biofilter reactors

Materials and methods

Three filters of 10 L volume were set up

- Different filler materials were used: plastic balls, crushed mussel shells, and whole mussel shells.
- Crushed shells would allow a greater specific surface area for the adherence of nitrifying bacteria
- Whole shells would be easier since they come directly from the industry and this would facilitate its handling if used in real plants.

Materials and methods: lab-scale

Periods	$N\text{-}NH_4$	HRT	Inocula	Parameters
1 (50 days)	5 ppm	15-20 days	Prodibio	pH, Ammonium, nitrite, nitrate, disolved oxygen
2 (175 days)	5 ppm	1 day after day 120	Sludge +prodibio, Probiobio, Bio S Aquaforest	pH, Ammonium, nitrite, nitrate, disolved oxygen

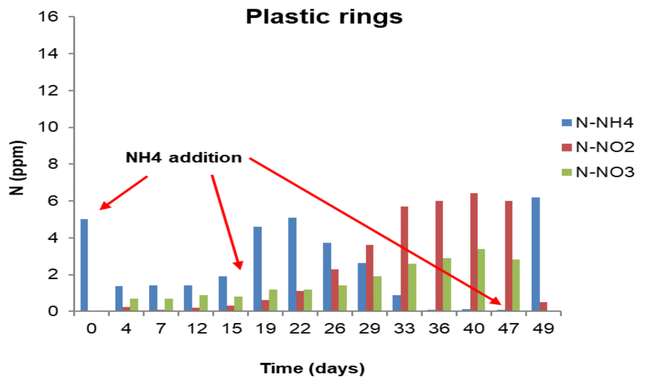
First period

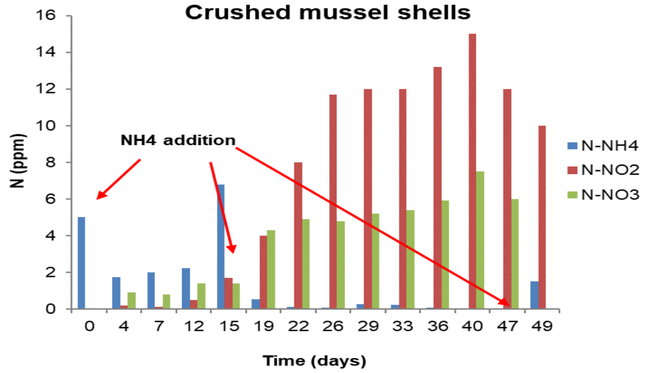
During this period the biofilters presented similar behaviour regardless the filling material.

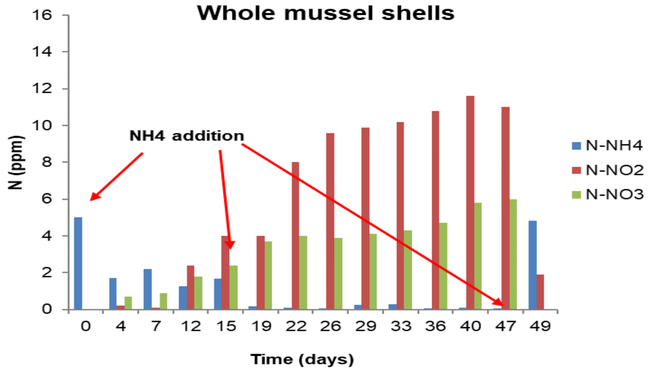
Start-up required almost 15 days.

After the second nutrient dosing, the biofilter filled with plastic material needed around 15 days more than mussel-shell biofilters to eliminate the ammonium. After this second dosing, the mussel shell filter showed a sharp decrease in the ammonia concentration and increase in nitrite (day 15 on), up to three times faster than in a biofilter packed with plastic balls.

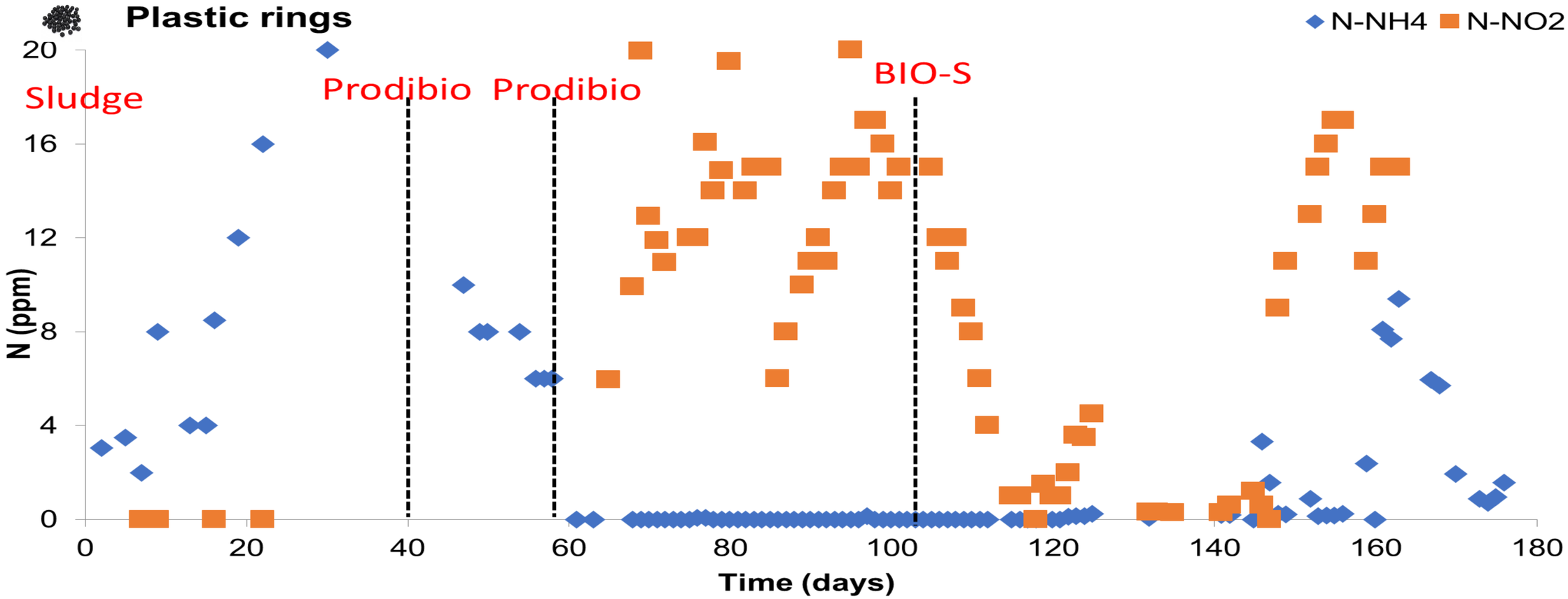
After the third ammonium acetate addition, the biofilter packed with whole mussel shells performed apparently better than other reactors in the second nitrification step.





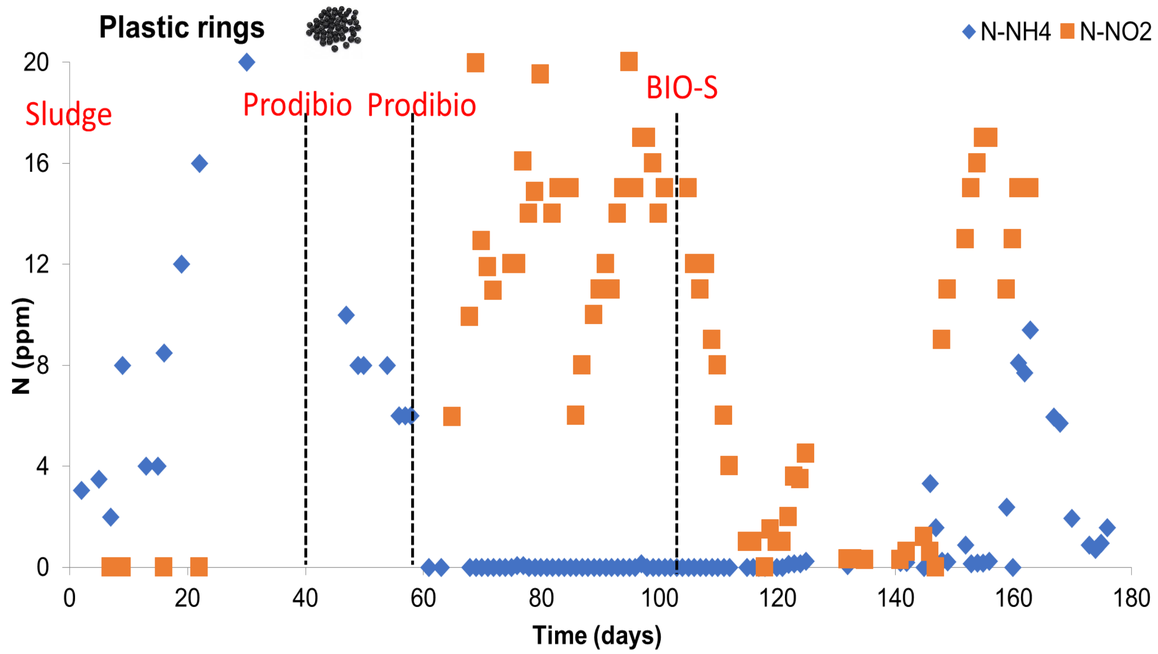


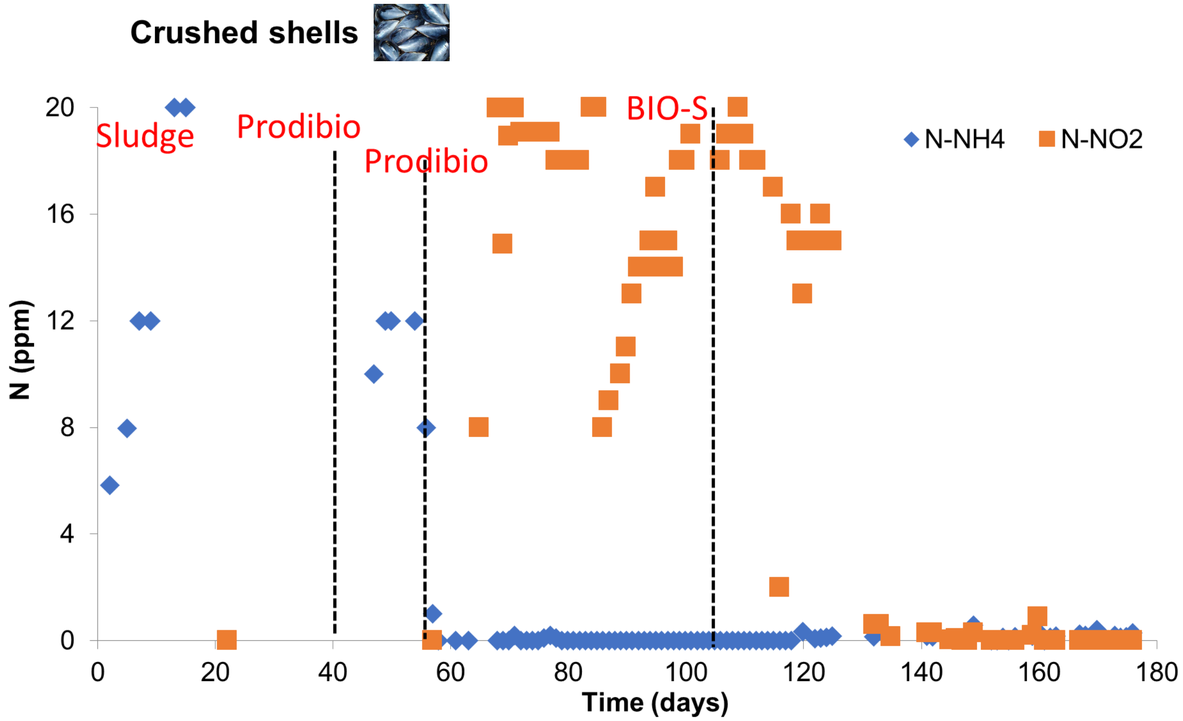
Second period



- Problems with inoculum for start-up (we use several inoculums: sludge, Prodibio, BIO-S)

Second period

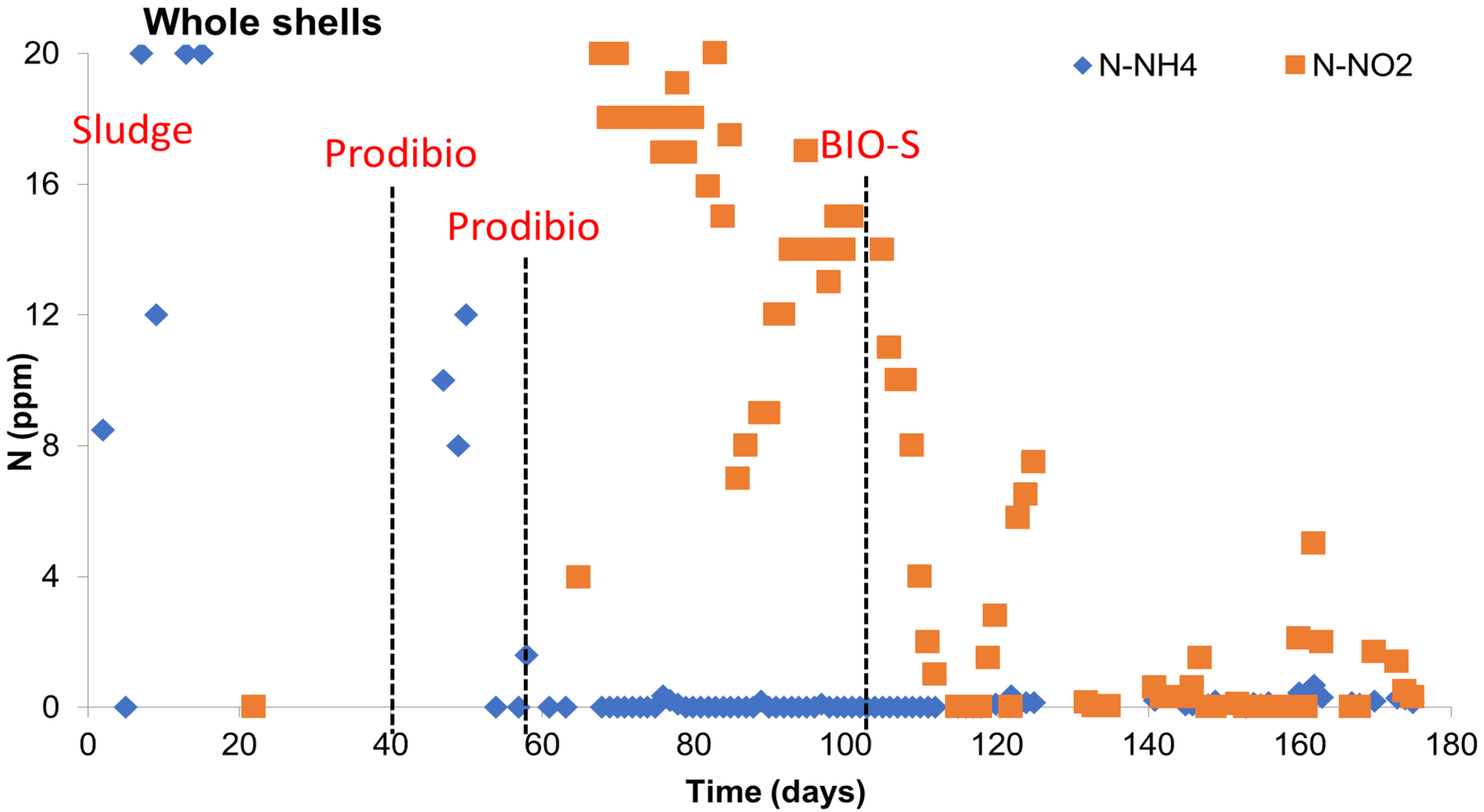




- After steady-state crushed shells had a better performance in terms of ammonium an nitrite elimination rates compared to plastic rings biofilter, mainly for pH shifts in the plastic reactor

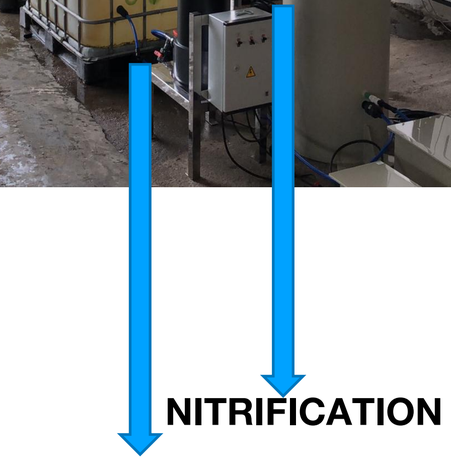
Second period

- Variation of pH was important (ranging from 6,7 to 8,4) in plastic rings biofilter. The addition of alkali was required across the operation period to control pH
- Mussels shell biofilters can maintain the pH without buffer requirements



- Similar to crushed shells, but this reactor needed more time to reach zero nitrite after BIO-S reinoculation

Materials and methods: pilot scale



Pilot-scale system installed in Grupo Tres Mares (Cee, Galicia)

5 ppm of ammonium concentration, and with a hydraulic retention time of 1 day

The initial idea was to study the possible phosphorous adsorption in the shells, but also some data coming from nitrogen elimination were obtained during this week.

Materials and methods: pilot-scale

NITRIFICATION

DENITRIFICATION

	$N\text{-}NH_4^+$	$N\text{-}NO_2^-$	$N\text{-}NO_3^-$
Feed (ppm)	5	0	0
Effluent (ppm)	0	0	1,8
Removal Efficiency (%)	100	100	66

- High nitrification (100%) and denitrification yields (more than 60%)

Materials and methods: pilot-scale

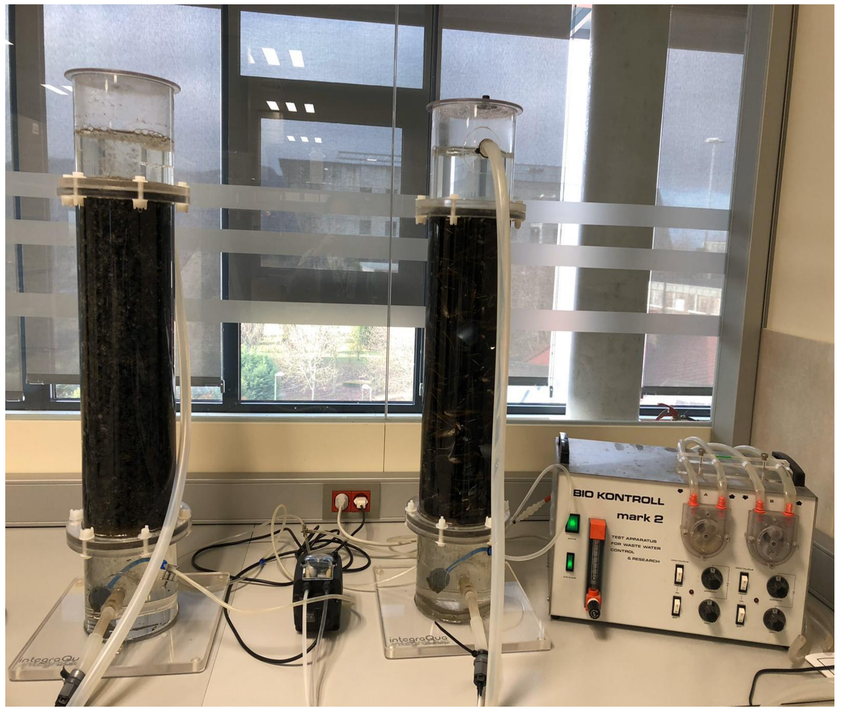
NITRIFICATION

DENITRIFICATION

	PO_4
Feed (ppm)	0,12
Effluent (ppm)	0,06
Removal Efficiency (%)	50

- The shells also allow high yields of phosphorous adsorption close to 50%

Materials and methods: phosphorous elimination

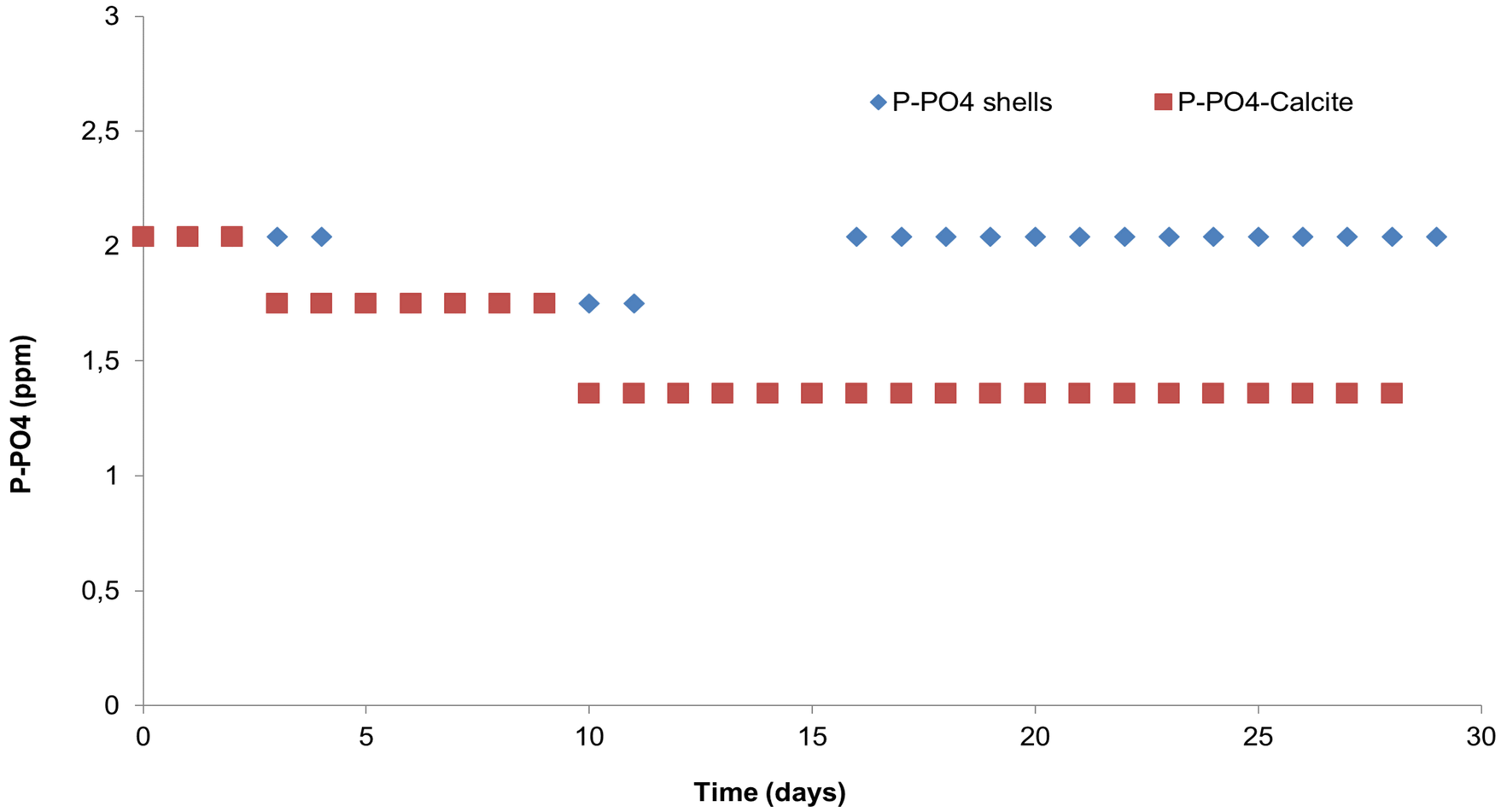


- Two of the three filters of 10 L volume were used to evaluate the phosphorus adsorption in the mussel shells.

- One of the filters was filled with crushed mussel shells, 1.0 kg, whereas the other one with the same quantity of calcite material.

- The filters were run for 30 days with concentrations of 10 ppm of phosphate. After that on day 30 we added 1 gram per litre of phosphate to evaluate the elimination at high concentrations over a 24 hour period.

Results of phosphorous elimination



- Calcite material was better since it was able to adsorb around 86% of the phosphorus, whereas in the crushed shells this value was 79%.

Results of phosphorous elimination

	Time 0 h - $P\text{-}PO_4^-$	Time 1h - $P\text{-}PO_4^-$	Time 24 - $P\text{-}PO_4^-$
Mussel shells	1	843	677
Calcite	1	843	550

- Considering a 24 hours period the removal was around 32% in the crushed mussel shells and almost 45% in the calcite filled filter

Conclusions of GAIN biofilter trials

The main conclusions are :

- Similar efficiency of shell filled systems (whole shells) compared to bioplastic fillers in ammonia and nitrite removal; reaching almost 100% working at lab-scale and treating simulated seawater.
- High removal efficiencies in nitrification (100%) and almost 70% in the denitrification step were reached using whole shells as biofilter packaging in pilot scale system.
- The selection of the inoculum seems to be important to promote both nitrification steps, i.e. both ammonia and nitrite removals.

Some considerations

Reagarding the possible use of the shells at high TRL, it would be neccessary to consider the following points for a future business plan:

- No replacement of shells was neccesary after 175 days operating the biofilters, since no excessive biofilm formation was observed. Therefore the shell waste for this use will be minimal in an aquaculture farm. For this reason the use of shells as a biofilter media can be extended to other wastewater treatment systems such as industrial or even municipal ones.
- Promotion of stricter regulations regarding effluent discharges (i.e zero ammoium and nitrite values) and water consumption for current aquaculture systems will be crucial for stimulating the development of these applications in a commercial way.
- The GAIN trials show that the use of plastic fillers in biofilters is not necessary for the correct elimination of nitrogen, clearly confirming the mussel shells as an alternative filling material. The future now involves evaluating the process over longer periods of time and on an industrial scale, to assess its economic viability and wider practical suitability.

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