STOCKHOLM JUNIOR WATERPRIZE 2026

WATER WITHOUT WORRY

DEVELOPING A COMPACT FILTER MADE FROM NATURAL MATERIALS FOR MICROBIAL WATER TREATMENT

MAYA SHARMA 9. NOVEMBER 2008
11TH GRADE MAX-PLANCK-GYMNASIUM TRIER
Germany

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Illustrations drawn by the author

Photos taken by the author

Biography

My name is Maya Sharma. I was born in 2008 and grew up in Trier, Germany, together with my older brother. Our home is located close to the Luxembourgish border, in the heart of Europe, which has given me the opportunity to experience different cultures and languages from an early age.

I am currently a student of Max-Planck-Gymnasium, Trier, and will graduate from school in 2028. My main subjects are mathematics, biology, and French. I particularly enjoy biology because it helps me understand the complexity of the human body and the whole natural world.

In my free time, I am a passionate dressage rider. I own a dressage horse that I have trained myself since she was very young. Building a strong partnership with my horse has taught me patience, responsibility, and dedication. We regularly participate in competitions, and I enjoy the challenge of constantly improving our harmony and performance.

Animals have always played an important role in my life, and I love spending time with them. Looking ahead, my goal is to become a doctor and study human medicine in Düsseldorf. I am fascinated by science and healthcare and would like to help people through a career in medicine. My ambition, combined with my determination and passion for learning, motivates me to work hard towards achieving this dream.

Acknowledgements

My deepest thanks go to the staff at the State Investigation Office, particularly Dr.Lars Jurzik. Without the opportunity to work on the premises of the State Forensic Science Institute and to use its equipment and materials, I would not have been able to carry out this work. Thank you for teaching me how to work in a laboratory and for being the most competent adressees for any questions on my journey.

I would also like to thank Jugend forscht, who sponsored an oxygen probe for my school to help bring my idea to life. I really appreciate the sponsoring of blood agar and want to thank Thermo Fisher Scientific for it. Thank you to my supervising teachers, Ms. Backes

and Ms. Pagé, who supported and guided me – and also made sure I didn't fall into the river 😉.

Technical summary

Under the title 'Water without worry', my aim is to design a cost-effective and efficient filter that will enable me to filter water from various water bodies to such an extent that it is free of bacteria and safe to drink without any risk to health. In order to better characterise the Moselle river as an example water body, parameters such as oxygen content, rainfall over the last three days, temperature and general microbiological parameters are required for this project. These parameters are relevant for a meaningful evaluation of the samples.

Motivation and research question

Actually, I wanted to investigate a completely different topic...

It all began in the summer of 2024. After searching for an S2 laboratory for a long time and speaking to many people on the phone, I finally came into contact with the State Investigation Office and Dr. Jurzik. Unfortunately, I was unable to carry out my original idea of investigating Fusobacterium necrophorum (the bacterium responsible for foot rot in horses) – this bacterium was not particularly welcome in the drinking water laboratory. Nevertheless, I was overjoyed that my contact with Dr. Jurzik had led to a collaboration with the State Investigation Office. I was offered a nine-month internship contract, and a new project idea emerged. I was then able to implement this successfully using the facilities and resources of the State Investigation Office.

The question arises as to whether a filter can be constructed from chitosan to achieve resilient drinking water production through the use of surface water in areas with low rainfall.

From 7 April 2025 to 20 October 2025, with a few exceptions, I took weekly samples from the Moselle river and analysed them for colony counts. A total of 26 samples were analysed during this period.

I began taking weekly water samples from the Moselle river in early April 2025, analysing them, and testing and refining filter designs.

Background

What is chitosan?

Chitosan is a natural polymer composed of long chains with amino groups attached. It consists of deacetylated chitin, which means that an acetyl group is removed from each building block, leaving a free amino group at that position (Younes & Rinaudo, 2015). In their normal state, these groups are neutral; in an acidic environment, they accept protons, causing the chitosan to become positively charged. Being positively charged they attract negative particles such as dirt or bacteria (Ke et al., 2021). Due to these properties, chitosan is used in medicine and, because of its antibacterial effect, is also a component of many medical plasters and wound dressings (Liu et al., 2022).

Chitosan can be obtained, for example, from crab shells, fungal cell walls or insect exoskeletons, making it a widely available, renewable raw material (Younes & Rinaudo, 2015). The synthetic production of chitosan is difficult, as each building block of the polymer would have to be produced individually, the bonds formed, and the amino groups positioned.

What is the structure of bacteria? Where does their negative charge come from?

Bacteria can be broadly divided into two groups:
Gram-negative and Gram-positive bacteria. These differ in their structure. For my filtration experiments, the structure of the bacterial cell wall is of particular relevance.

Even though individual bacteria differ greatly in structure, common features can still be identified. The general structure of a bacterium from the outside in:

Gram-negative bacteria:

- Outermost layer: a negatively charged capsule (optional) due to carboxyl groups, phosphates, sialic acids and uronic acids (Whitfield, 1988)
- outer membrane: consisting of an
    - outer layer containing lipopolysaccharides (LPS). The LPS consists of O-antigens, which are long sugar chains protruding outwards with a weak negative charge. It also consists of Lipid A molecules. These are disaccharides containing several fatty acids and many phosphate groups, giving them a strongly negative charge. The Lipid A and O-antigens are linked by core polysaccharides. These contain sugars, phosphates and carboxyl groups, which also give them a negative charge (Sperando et al., 2009).
    - Inner layer, consisting of various phospholipids.

  Consequently, the outer membrane carries a negative charge.

- Periplasm: has no net charge.
- Peptidoglycan layer: It consists of sugars linked by peptide bridges and carries a slightly negative charge due to the carboxyl groups of the peptides.
- Inner membrane: This consists of phospholipids and membrane proteins and is therefore slightly negatively charged.

The main source of negative charge in Gram-negative bacteria is the lipopolysaccharide layer.

Gram-positive bacteria have a less complex structure:

- Outer envelope: a negatively charged capsule (optional)
- Peptidoglycan layer, very thick. This consists of sugars and peptide bridges, but is permeated by long anionic polymers, the teichoic acids (Silhavy et al., 2010). These possess many phosphate groups, which results in a strong negative charge. These are followed by lipoteichoic acids, which are also strongly negatively charged as they possess many phosphate groups. They link the peptidoglycan

layer to the inner membrane, which, as with Gram-negative bacteria, is composed of phospholipids and proteins.

The main reason for the negative charge in Gram-positive bacteria is the teichoic and lipoteichoic acids.

It can therefore be concluded that bacteria generally carry a negative charge.

This explains in theory, why the filter works.

Description of the sampling site

To ensure I had comparable data, I always took all my samples from the same spot on the Moselle river. Whilst searching for a suitable location, I found a spot where a paved slope led directly down to the Moselle river. This meant that even when the water level was low, I was not at risk of falling into the river. There is a campsite near this spot. However, the area around the slope consists only of grass, trees and bushes. From the end of April onwards, a family of Egyptian geese was regularly seen spending their days there.

Figure1 : Sampling point on the Moselle

Materials and chemicals used in the project

Chitosan, vinegar essence, caustic soda, polyethylene glycol, sodium tripolyphosphate, distilled water, NaCl, sand, glass wool, nutrient agar,

filter column, Erlenmeyer flask, Eppendorf pipette, pipette tips, magnetic stirrer, reagent shaker, Petri dishes, sample bottle, oxygen probe, magnifying glass, counting grid, permanent marker, gloves, safety goggles

Procedure

For each sampling, I used the probe to measure the oxygen content and temperature of the Moselle, and also checked the „Hochwasser RLP“ website for the current flow velocity and water level. Furthermore, I noted the rainfall over the previous three days.

Once I arrived at the laboratory, I first boiled the DEV agar; once it had become liquid, I then maintained its temperature at 46 °C in a water bath. This ensured that it was still liquid when I poured the plates, but the heat had not destroyed any bacteria. I then prepared two dilutions of the sample and NaCl each time, at ratios of 1:10 and 1:100. In duplicate, I incubated the sample and the dilutions at 22 °C and 36 °C respectively.

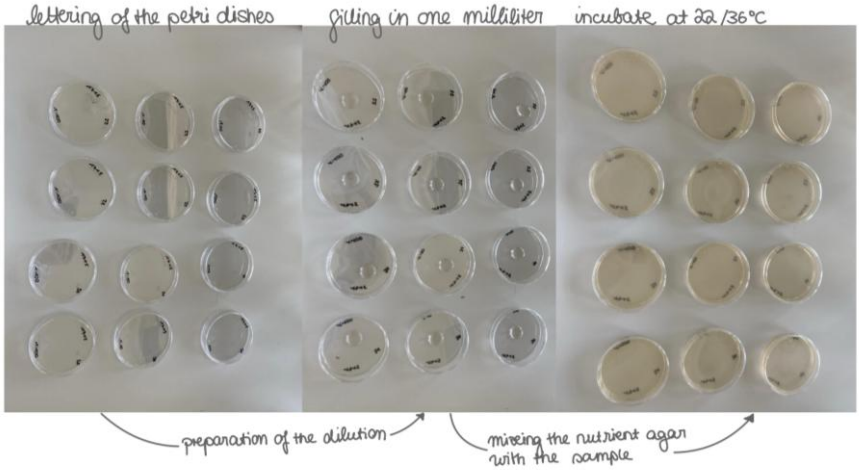


Figure 2: Preparation of the Petri dishes

The colonies that grew at 36 °C were the human-pathogenic bacteria (due to human body temperature), and everything that grew at 22 °C belongs to the group of environmentally harmful bacteria.

About 48 hours later, I read out the plates by counting every single visible colony. If too many colonies had grown, I used a counting grid; with this, I counted the colonies in a square at several points, calculated the mean value and then multiplied this by the total number of squares.

At low dilutions, it sometimes happened that a spore-forming organism had spread across the entire plate; as it was then no longer possible to count the colonies accurately, I marked these plates as n.a. (not analysable). On some days, I also tried out filter ideas.

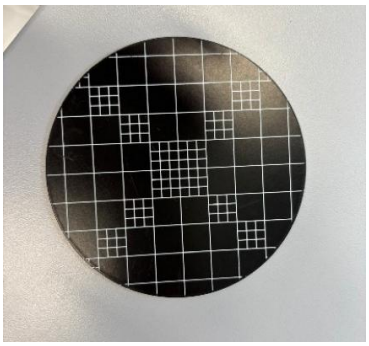


Figure 3: Non-evaluable samples & counting grids

Filter

To begin with, I tested whether simply placing the chitosan into a filter column was sufficient for filtration. To stabilise the setup, I placed glass wool at the bottom of the filter column, some sand on top of that, and the chitosan flakes on top. (Filter experiment 1)

Figure 4: Schematic diagram of filter experiments 1 & 2

In another experiment, the sample was first mixed with a little chitosan and then, after 10 minutes, placed on the filter column containing chitosan. (Filter experiment 2)

As these two experiments did not yield any additional benefit in terms of reducing the bacteria, I considered what might be the cause and, whilst researching online, took another look at the structure of chitosan.

Chitosan has a very small surface area, which means there is hardly any contact between the chitosan and other substances. Furthermore, it is insoluble in neutral or alkaline water (Aranaz et al., 2021).

It therefore made sense to investigate it in an acidic environment. To do this, I prepared a 1% acetic acid solution using distilled water and acetic acid. I then added 2 g of chitosan

to 200 ml of this solution. To ensure everything mixed well, I stirred the mixture for several hours using a magnetic stirrer. Right from the start of stirring, it was clear that a gel had formed from the solution, which became increasingly viscous over time. The chitosan dissolved completely.

In the next experiment, I then tried to use the gel for filtration, but unfortunately without success. Even at a concentration of 0.001% chitosan, the gel did not allow any water to pass through. I therefore tried to see if the gel would mix with the water and thus reduce the bacteria. That would certainly have been a good idea, but unfortunately I was unable to separate the gel from the water afterwards. My idea of drying the gel and then using this film as a filter was also not feasible. (Filter experiment 3)

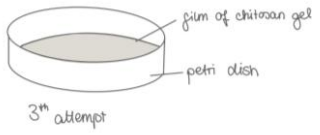


Figure 5: Set-up for filter experiment 3

So I thought about how I could continue to use the gel, but still prepare it in such a way that I could use it for filtering.

After much thought, I came up with the idea of making beads, similar to vegan caviar. This was intended to increase the surface area whilst still remaining permeable. So I looked into how vegan caviar is made and then applied this process to my chitosan gel. To better adapt the process, I looked into how ionotropic gelation works (Li et al., 2023). This led me to the chemical sodium tripolyphosphate, and I decided to use this as an anionised solution.

For this experiment, I prepared a new gel by dissolving 0.4 g of chitosan in 200 ml of 1% acetic acid. This new formulation was more advantageous as, whilst it still formed a gel, the gel was significantly more fluid.

I then used caustic soda to adjust the pH of the gel from approximately 3.5 to 5. In another container, I mixed distilled water with sodium tripolyphosphate to produce a TPP solution. I then dripped the chitosan gel into this solution whilst stirring slowly. This resulted in beads which, although still relatively uneven, were otherwise exactly as I had imagined.

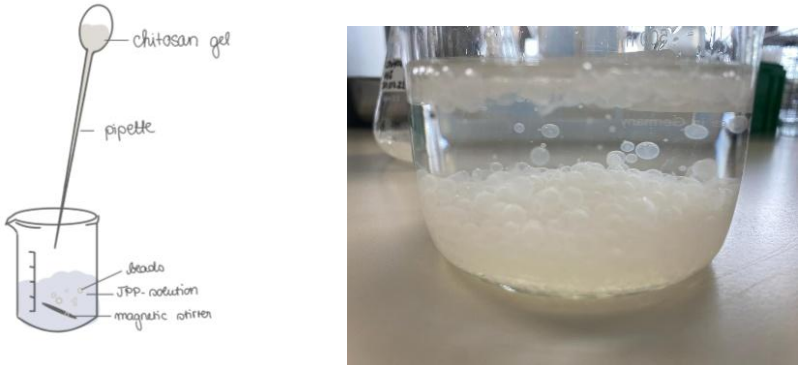


Figure 6: Production of the beads & finished beads

To remove the resulting beads from the TPP solution, I washed them. To do this, I first centrifuged the beads with a small amount of TPP solution, so that the beads settled and I could simply pour off the rest of the solution. I then added distilled water and shook the mixture. This allowed the beads to mix evenly in the distilled water again, and I was able to centrifuge the whole thing once more. Finally, I poured off the distilled water, and the beads were ready.

For filtering, I placed the beads on the filter column instead of the chitosan flakes and then ran the water through. (Filter experiment 4)

Figure 7: Setup of Filter Experiment 4

Although the results were already considerably better, they were still not good enough to drink the water.

So I considered what I needed to change to improve my results. To ensure more water came into contact with my chitosan, my aim was to increase the surface area. That is why I wanted to make fibres. These have a significantly larger surface area for the same volume. I optimised the process slightly by setting the magnetic stirrer to rotate much faster whilst I dripped the gel into the TPP solution. The result was thin filaments which,

after washing, formed a net-like structure, leading me to expect a significantly better filtration result. (Filtration experiment 5)

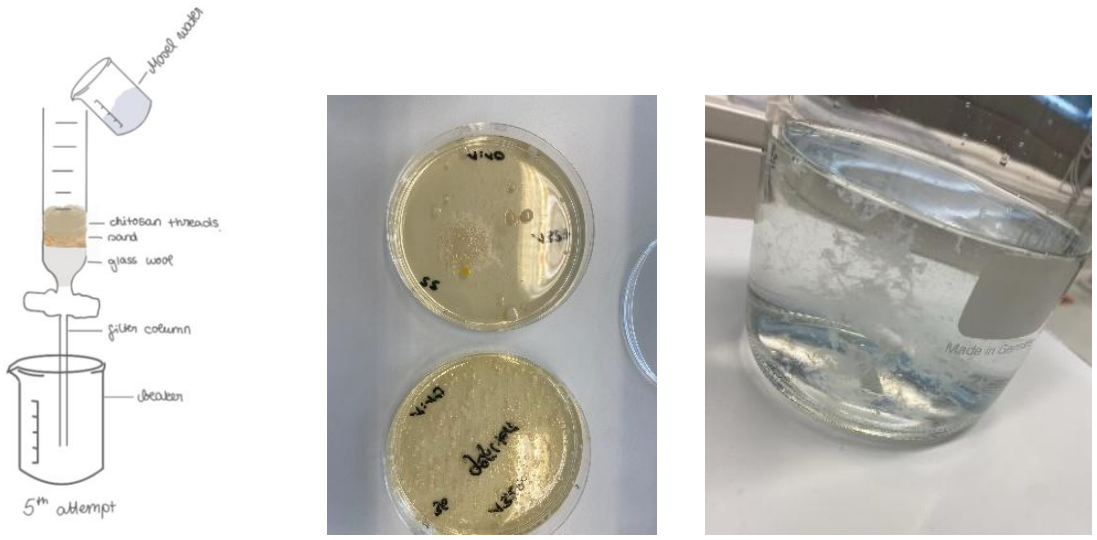


Figure 8: Set-up for Filter Test 5; poor filtration result; filaments in the beaker

However, as the water had unexpectedly formed significantly more colonies after filtering than before, I set about (dejectedly) looking for the problem.

My mistake was that, after the experiments, I had only rinsed the filter column with water and had not sterilised it. Over time, colonies had grown inside the column, meaning that, because of the contaminated filter, more bacteria grew after filtering than before.

Control

After autoclaving the column, I carried out a few additional controls. I ran sterile water through it and nothing grew on the plates afterwards, so I knew that the autoclaving had been successful. Furthermore, I always sterilised the sand and glass wool as well, to ensure they could not carry any germs either. I then ran river water through the filter column packed with glass wool and sand and observed that, after this 'filtering', there were just as many bacteria in the sample as in the 'unfiltered' one. This control demonstrated that any success was due to my chitosan and not to the sand or the glass wool.

Once I knew why the last experiment had failed and had re-sterilised the filter column, I made some new gel, as the old one was now almost empty and quite old. I repeated the

entire experiment with the threads using the new gel. To make the threads a little more stable and less prone to breaking, I added 5g of polyethylene glycol during the gel preparation in a later experiment, as this substance forms filamentous structures in water – which gave the threads even more structure. This filter was then my finally optimised model.

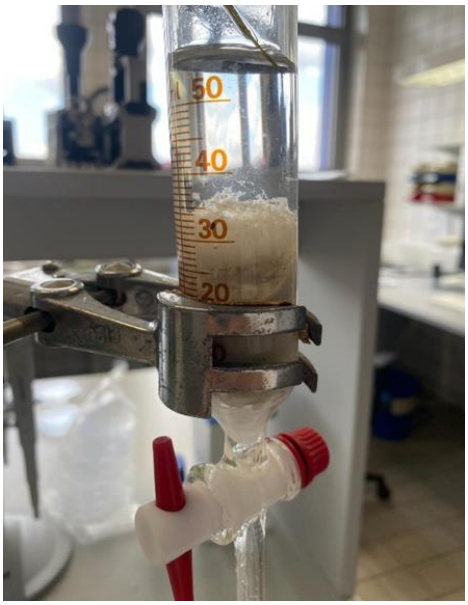


Figure 9: Photo of the final optimised filter

Result

Results of the filter experiments

Now that I had autoclaved everything before each experiment, the filter was able to work its magic and the previously undrinkable Moselle river water was of drinking water quality after filtering.

As I was still used to the high colony count from the previous filter tests, I chose a dilution of 1:10 for the first filter evaluation.

However, as almost all bacteria had been filtered out, I did not apply any further dilution in any of the subsequent tests. To see if there would be an even better result if I mixed the sample with a few strands of thread before filtering, I tried both methods with the same sample. Furthermore, I focused solely on human-pathogenic bacteria (36 °C).

The result differed by only 3 colonies, although even the filtrate that had not been mixed with the fibres contained fewer colonies. Generally speaking, however, these 3 colonies

make no difference, as despite thorough shaking, there could be more bacteria in one millilitre than in another. Nevertheless, the experiment showed that it is not necessary to mix the sample with the threads beforehand.

Date	unfiltered 22 °C	filtered 22 °C	unfiltered 36 °C	filtered 36 °C
08/09/2025	5073	0	3919	10
15 September 2025	8910	4	8965	1
6 October 2025	4227	not tested	2997	9

The permitted limit for colony count in drinking water at 22 °C and 36 °C is 100 cfu/ml (colony-forming units per millilitre). Before filtering, this value was in some cases exceeded by a factor of 90. By filtering, I managed to get well below the limit. The water was therefore perfectly safe to drink following my filtering tests.

Further investigations (following the completion of the original project):

To be able to use my filter in everyday life exactly when it is needed, it needs to be durable. I therefore dried it to enable it to be stored for a longer period of time.

To do this, I placed my freshly made threads in a drying cabinet at 36 °C for 24 hours.

The result was very stable, hardened threads. I then placed them in a filter column and, as before, tested the bacterial load before and after filtering. Using this method, I managed to reduce the bacterial count by approximately 50%. However, as this was still far too many bacteria for my liking, I tried to see what would happen if I first re-moistened my dried fibres in the filter column so that their surface area increased again and they softened. After moistening them for about 5 minutes with distilled water, I then ran microbiologically contaminated water through them. Using this method, I managed to reduce the bacteria to negligible levels or eliminate them completely. Through this series of experiments, I realised that the filter can be stored and retains its full effectiveness if moistened again before use.

Sustainability

However, as I would like to use the filter more than once, I considered how – or indeed whether – I could regenerate it. Since chitosan burns during autoclaving, I tried drying the

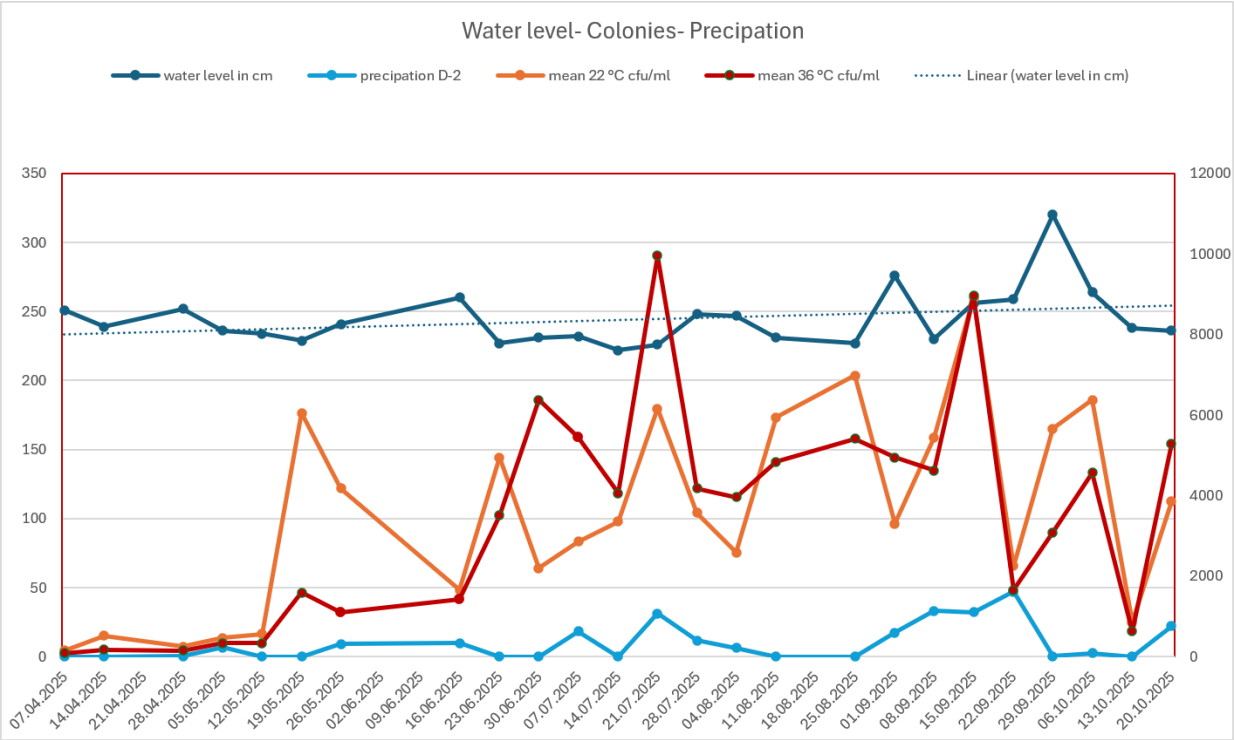
contaminated fibres at 60°C. After re-moistening these dried fibres, I was able to restore the filter to its full performance. The result is no different from that achieved with freshly made fibres. This is because most bacteria found in water die at 60°C, especially if this temperature is maintained for a few hours. Whilst this 'regeneration' does not guarantee sterilisation or the elimination of all germs, it does kill many of them, thereby extending the filter's lifespan.

Results of the Moselle water analysis

A total of 26 samples were analysed. For the colony count (22 °C), the minimum and maximum values were 150 and 8,910 CFU/ml. At 36 °C, the minimum and maximum values were 90 and 9,915 CFU/ml. The water level of the Moselle fluctuated between 222 and 320 cm during the study period.

The oxygen content I measured varied between 7 and 12.65 mg/l and the temperature between 10.3 and 24.9 °C.

I analysed the Moselle water samples throughout the year using a 1:10 dilution, as the undiluted sample became too imprecise as the year progressed. If I had extrapolated the results using the counting grid, even the slightest miscount could have led to significant inaccuracies and discrepancies of several hundred colonies. With a dilution of 1:100, on the other hand, only a very small amount of the sample is present. Here too, measurement inaccuracies are to be expected due to the high dilution. I therefore chose a dilution of 1:10.



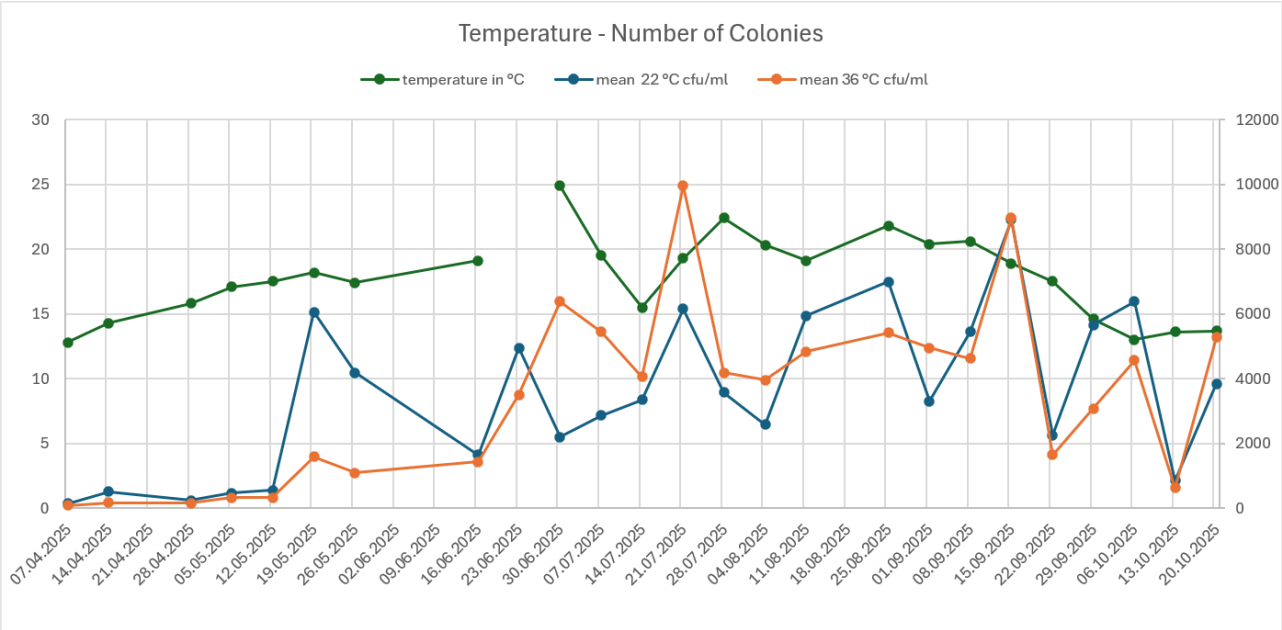
This diagram shows the mean number of colonies at 22 °C and 36 °C, the water level and the precipitation D-2. The colony count is based on a dilution of 1:10.

My values for the colony count show significant fluctuations. From April to mid-May, the values for the colonies that grew were very low.

In mid-May, both values suddenly rose sharply, with the number of colonies at 22 °C being significantly higher than that at 36 °C. Over the course of the summer, both values rose even further. However, this rise was not uniform, and there were also days in between when the colony counts decreased, although they never reached their April low. At 10,000 colonies, my counts at 36 °C reached their peak on 21 July 2025 . Generally, the colony counts at 36 °C were significantly higher than those at 22 °C for most of the summer. It was not until autumn that both values were almost equal, and as winter approached, the number of germs grown at 22 °C finally increased and exceeded the number of germs at 36 °C. Unfortunately, when comparing all the parameters shown, there is still no clear indicator of when the colony count is high and when it is low. There were days when it rained heavily and the colony count was high, yet after some other rainfall there was no increase in the number of colonies. In the summer, following a long dry spell, there were

also peaks in colony numbers. On other days, however, when it had been dry for some time, there were no spectacularly high values.

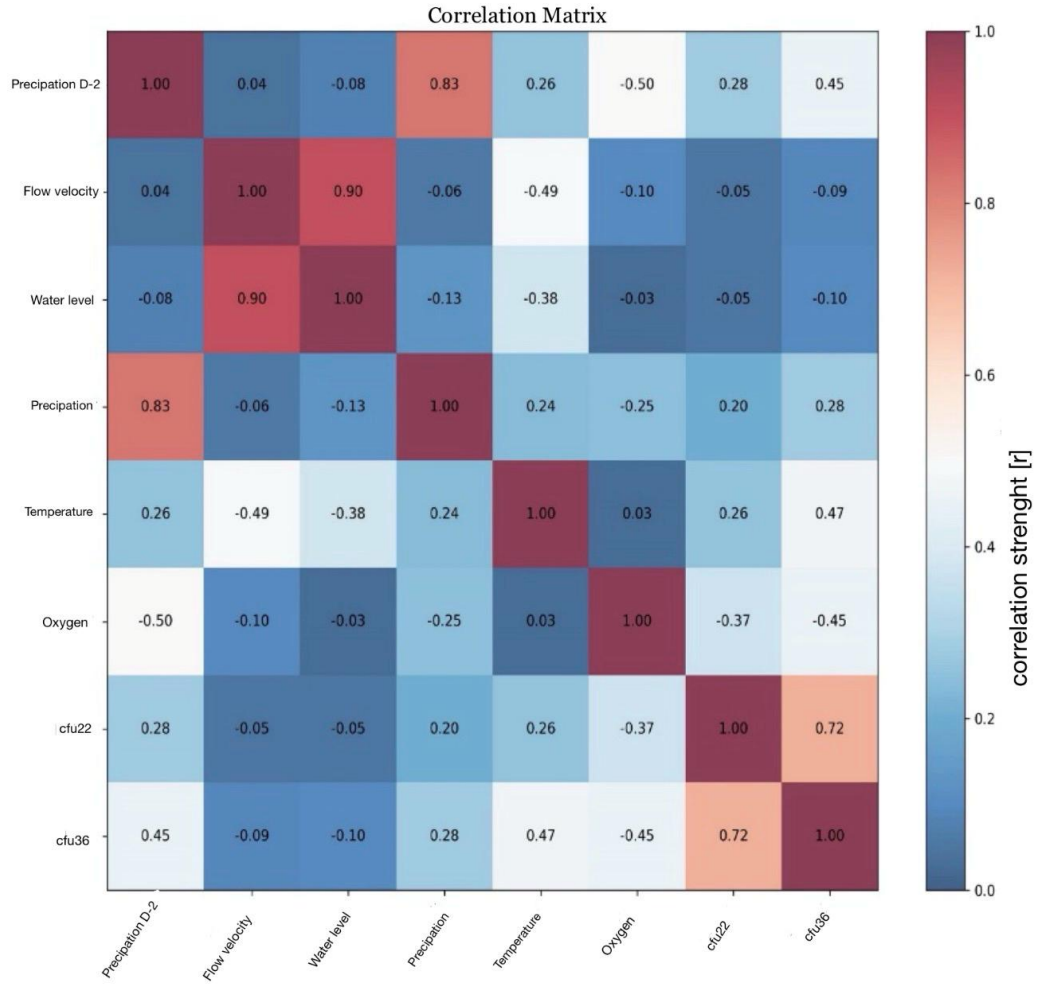
I was able to identify a correlation between temperature and the number of colonies.



As the temperature rose, the number of colonies also increased; the bacteria at 36 °C in particular behaved in this way. This became clearly visible in July: when there was a significant drop in temperature, there was also a sharp drop in the bacteria at 36 °C; when it got warmer again, these bacteria increased immediately.

However, as it is sometimes difficult to identify correlations from diagrams – since the values are not always easy to represent – I have plotted all parameters using a correlation matrix.

Correlation is a measure that indicates the strength and direction of a linear relationship between two variables. Here, relationships are immediately apparent and there is no room for misinterpretation. If r = +1, there is a perfect positive linear relationship. If r = -1, there is a perfect negative linear relationship. If r = 0, there is no relationship at all. The values in the matrix are calculated using the Pearson formula; for simplicity, this can also be done in Excel using the =*CORREL* command.



Magnitude of r	Interpretation
0.00 -0.019	No or negligible correlation
0.20 -0.39	Weak correlation
0.40 -0.59	Moderate correlation
0.60 -0.79	Strong correlation
≥0.80	Very strong correlation

The correlation matrix now clearly shows that there is a very strong correlation between water level and flow velocity. This relationship is logical, as a higher flow velocity means more water in the riverbed, which automatically results in a higher water level. A moderate correlation is visible between temperature and germination at 36 °C. This shows that my assessment of a relationship based on the diagram was correct.

My assumption of a relationship between growth at 36 °C and precipitatio-2 was also correct, as there is likewise a moderate correlation between these two parameters. Furthermore, there is a moderate correlation between oxygen and bacterial growth at 36 °C. This time, however, it is negative. When there was little oxygen present, bacterial growth at 36 °C was higher. This shows that my bacteria at 36 °C prefer low oxygen levels and are therefore more anaerobic. The bacteria show a strong correlation at 22 °C and at 36 °C. This relationship was already visible in the graphs. At the start of the sampling, both values were low and increased over the course of the summer.

Discussion of results

Chitosan filter

My chitosan filter proved suitable in the experiments for significantly reducing bacteria. It therefore represents a simple, cost-effective and sustainable method for surface water treatment. In further experiments, one could investigate the reduction of chemicals or the elimination of turbidity using my chitosan fibres instead of bacterial load.

Moselle water analysis

I have not yet found a single, entirely clear factor that explains why there was an occasional rise or fall in bacterial levels. However, I have several hypotheses which I believe, when considered together, could offer an explanation. During a long dry spell, the bacterial load is particularly high at 36 °C. At this time, the water level is lower and the contaminants in the remaining water are more concentrated. Consequently, for example, any residual faecal matter is also significantly more concentrated, which is why the extreme increase occurred primarily in the bacterial count at 36 °C . Furthermore, the bacterial load is significantly higher after heavy rain, which could be due to large amounts of water entering the Moselle via fertilised fields or through the overflow of sewage systems, water that would not otherwise reach the river. At particularly warm temperatures, the number of colonies increases significantly, especially at 36 °C. At warmer temperatures, it is much easier for bacteria to thrive, so they also multiply more

quickly. As dry spells and warm temperatures usually go hand in hand, the bacterial load is greatly increased at these times.

In my opinion, however, it is not only rainfall and temperature that play a decisive role.

At the end of April, I noticed that a Nile goose had hatched a large brood of goslings near my sampling point. Goslings were hatching all over the area at that time; they were scattered along the riverbank and, over time, learnt to swim in the Moselle river. With the increase in the number of ducks, there was also a sharp rise in the amount of (Nile goose) droppings. I therefore think that my sudden rise in bacteria levels is linked, amongst other things, to all the Nile geese in the Moselle river, as the number of birds living on the river has simply multiplied so rapidly.

Outlook

Through my analysis of bacterial levels in the Moselle river, I have found that it is particularly important, especially during the summer months, not to drink Moselle river water without filtering it first.

To make better use of the filter I developed, particularly in areas where a reliable supply of drinking water cannot be taken for granted, I decided to design a bottle adapter to fit my filter. The standard plastic and, often, glass bottles available in shops all have screw caps of the same diameter. That is why I developed an adapter that can be screwed onto these bottles. The water flows through this and comes out on the other side of the adapter as drinking-quality water. This allows you to connect a bottle containing water to be purified to an empty collection bottle, thereby obtaining drinking-quality water ready to drink straight from the bottle.

These could be taken anywhere, allowing you to use any water and enjoy fresh, clean drinking water after a short time. This would be a particularly good way of protecting people from diseases spread via drinking water in countries with poor drinking water supplies. Furthermore, filtering is a good way of maintaining the drinking water supply in the event of potential flooding scenarios, as a reliable drinking water supply is also unavailable in such situations.

Furthermore, my filter is very sustainable. The chitosan required can be obtained from chitin, which can be cultivated in the form of fungal cell walls or extracted from crab shells, which serve as a waste product in the food industry. Chitin could also be extracted from the bee carcasses found around hives during beekeeping to produce chitosan.

Going forward, I am looking for an industry sponsor who would like to work with me on further developing my idea for the filter and the adapter, with a view to bringing it to market readiness.