(04-018) - Nature-based solutions for sanitation in the Chinampas in Mexico City

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The agrohydrological system "Las chinampas in Mexico" has been a model of preservation of natural resources for decades. However, the water in the area is highly contaminated by anthropogenic activities, deteriorating the quality of the water resource, putting the environment and public health at risk. The present study evaluated the effect of cork-base biofilters functionalized (cork-f) with M. olieifera seeds extracts (MoSe) and adapted in Nutrient Film Technique (NFT) hydroponic system as tertiary wastewater. A 27-4 fractional factorial design and biofiltration system were used to evaluate the effect of gravel functionalized with MoSe on the inhibition of E. coli (InhEc). The results showed that treatment 4 indicated the greatest inhibitory effect on E. coli, with a maximum bacterial reduction of approximately 99.10% (p<0.05). The relevant conditions were established in treatment 4: (646 ml/min) aeration, (3 mm) gravel particle and (6 h) lysis time. In conclusion, the functionalization of cork-based biofilters with an NFT hydroponic system increased the potential for antimicrobial activity being a promising strategy for wastewater treatment.

Keywords: cork-based biofilter; water treatment; chinampas; functionalization; hydroponic system; Moringa oleifera

Soluciones basadas en la naturaleza para el saneamiento en las Chinampas en Ciudad de México

El sistema agrohidrológico "Las chinampas en México" ha sido un modelo de preservación de los recursos naturales durante décadas. Sin embargo, el agua de la zona está altamente contaminada por actividades antropogénicas, deteriorando la calidad del recurso hídrico, poniendo en riesgo el medio ambiente y la salud pública. El presente estudio evaluó el efecto de biofiltros a base de corcho funcionalizados (corchof) con extractos de semillas de M. oleifera (MoSe) y adaptados en un sistema de hidroponía de película nutritiva (NFT) como tratamiento terciario de aguas residuales. Se utilizó un diseño factorial fraccionado 27-4 y un sistema de biofiltración para evaluar el efecto de grava funcionalizada con MoSe sobre la inhibición de E. coli (InhEc). Los resultados mostraron que el tratamiento 4 indicó el mayor efecto inhibidor sobre E. coli, con una reducción bacteriana máxima de aproximadamente 99,10% (p<0,05). Las condiciones relevantes se establecieron en el tratamiento 4: (646 ml/min) aireación, (3 mm) tamaño de partícula de grava y (6 h) tiempo de lisis. En conclusión, la funcionalización de biofiltros a base de corcho con un sistema hidropónico NFT aumentó el potencial de actividad antimicrobiana siendo una estrategia promisoria para el tratamiento de aguas residuales..

Palabras clave: biofiltro a base de corcho; tratamiento de agua; chinampas; funcionalización; sistema hidropónico; Moringa oleifera

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Agradecimientos: We thanks to Mexico-CONAHCYT, grant number 3006060, Banco Santander and Universitat Politècnica de Catalunya-BarcelonaTech for the postdoctoral fellowship.



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1. Introduction

The agro-hydrological system practiced for many generations and developed by the Aztecas "The Chinampas in Mexico" provide food, employment and preservation of natural resources (Robles *et al.*, 2018). Chinampas are ecologically and culturally recognized as a Ramsar site, a World Heritage site by UNESCO, and a Globally Important Agricultural Heritage System (FAO, 2017). The water resource is the fundamental element for the proper functioning of the ecosystem, which have evolved under the specific ecologic opportunities. However, the water quality of the zone was severely affected and deteriorated due to some factors, among them high levels of contamination physicochemical a biological pollution generating negative impacts in the environment and the public health (González *et al.*, 2016).

Currently, wastewater from houses surrounding the lake from chinampas often goes directly into the water body used to irrigate crops, representing a threat to the construction of a food security system for the benefit of the population in the Chinampera Zone and Mexico City (Gómez *et al.*, 2020; Rosas *et al.*, 2015). Some major health risks are caused by microorganisms such as bacteria or pathogen the best-known coliforms to indicate the fecal contamination because it can be found almost exclusively in human and animal feces (Wu *et al.*, 2018). Additionally, because fecal bacteria may survive for long periods, reproduce and disperse in water systems (Pereira *et al.*, 2014; Jang *et at.*, 2017). There are important fecal indicator and opportunistic pathogens bacteria as *Escherichia coli* and *Enterococcus faecalis*, causing serious diseases of clinical importance transmission across the human-agriculture-environment continuum (Zaheer *et al.*, 2020). Likewise, these bacteria could be found in the nature of hydrological processes that transport the organisms to and within the aquatic environment (Pereira *et al.*, 2014; Nurliyana *et al.*, 2018; Ramos *et al.*, 2020).

In this sense, one of the biggest challenges with sanitation is the development and implementation of environmentally friendly, easy-to-use and low-cost systems. (Arockiam *et al.*, 2020; Hyun *et al.*, 2019). Nature-based solutions (NBS) can protect, manage and restore natural or modified ecosystems that address societal challenges effectively and adaptively, simultaneously providing human well-being and biodiversity benefits (Browder *et al.*, 2019; Cohen-Shacham *et al.*, 2016; Ramirez-Agudelo *et al.*, 2020). Water pollution control nowadays is predominantly carried out with various types of NBS mainly constructed wetlands and green infrastructure can be effectively used in combination with biofiltration systems for water's reuse and its sustainable sanitation (Oral *et al.*, 2020).

The combination of applied biotechnologies is essential as wastewater treatment to obtain microbiological high quality agricultural products, avoid potential of intestinal infections through the fecal-oral transmission between the farmer and wastewater and to provide additional treatment for safe discharge to the environment (Srivastava *et al.*, 2022; Ukaogo *et al.*, 2020).

Biofiltration and the hydroponic system Nutrient film technique (NFT) are considered as one of the alternatives for the elimination of contaminants and pathogens involving a combination of physical-chemical and biological processes with microorganisms, plants, and media-based interactions (Cifuentes-Torres *et al.*, 2020; Szekely & Jijakli, 2022). Likewise, the application of natural coagulants such as plant seed extracts has been shown to reduce and/or neutralize colloid loads and eliminate pathogens (Choy *et al.*, 2014; Camacho *et al.*, 2017).

Thus, the functionalization process has been established to retain the active protein with antimicrobial activity of *Moringa oleifera* (*M. oleifera*) seed extracts (*Mo*Se) on porous materials such as cork, increasing the inhibition potential and effectively reducing the growth of *E. coli* (Infante *et al.*, 2021). For this reason, the objective of this study was to evaluate the effect of gravel biofunctionalized with antimicrobial compounds of *Mo*Se (*f*-gravel) on the inhibition of *E. coli* by using the fluorescence technique. Likewise, a hydroponic NFT system was

development and coupled to cork base-biofilters system functionalized of *Mo*Se (*f*-cork) as a tertiary treatment for improving *E. coli* removal of water pretreated in the Chinampera Zone.

2. Material and Methods

2.1 Chinampa water characterization

Water samples were collected from Apatlaco channel and in each part of a constructed biofiltration system in the Chinampa Aurora of the Chinampera Zone, Mexico (19°16'07.2"N 99°05'01.6"W) (Rodríguez–Vázquez, 2018). Electrical conductivity (EC), pH, dissolved oxygen (DO) and total dissolved solid (TDS) of water samples were determined *in situ* using a standard multiparameter probe. The samples were stored at 4 °C until reach the lab for further analysis (APHA, 2005). Microbiological analysis of water samples was performed using commercial media to quantify the content of *E. coli*. Briefly, water samples were cultured on Chromocult coliform agar, Eosin Methylene Blue Agar (EMB) and Luria-Bertani (LB) with serial dilutions using the micro-drop technique and incubated at 37 °C for 12-24 h (Pitkanen *et al.*, 2007).

2.2. Functionalization of gravel with *MoSe*

The *M. oleifera* seeds were provided by the Autonomous University of Sinaloa, Sinaloa, Mexico. The extraction operation of bioactive components from *M. oleifera* seeds, the stock solutions and the biofunctionalization process of the filter material was following the work carried out by Infante *et al.* (2021) with some modifications. Briefly, a seed particle size of 0.9 mm was used and two stock solution concentrations of 5 and 10% were prepared. According to the study of Jerri *et al.* (2012) with some modifications, the gravel cleaning process was carried out by three washes with tap water, 3 washes with distilled water and left for 24 hours with deionized water.

2.3. Biofiltration system and antimicrobial activity

Biofiltration system was operated in glass columns of volume (188.5 cm³), diameter (4 cm) and a length (16 cm) as is shown in Figure 1. The factors and conditions were established according to the 2^{7-4} fractional factorial design (see section 2.5). The body of the filter was constituted by an elongated glass cylinder, with a circular opening with a hollow connector in the center, in which a tube was connected through, and the water inoculated with *E. coli* cells above 1 x 10⁸ CFU/mL (aliquots of 50 mL) was treated in the filtering system filled with *f*-gravel functionalized with *Mo*Se and without gravel functionalized (control), in which the viability of *E. coli* was evaluated with a hydraulic retention time at room temperature for 0, 6 and 12 h. Then, water samples were taken and plated on Chromocult agar and incubated for 12 h at 37 °C. The results of the inhibition of *E. coli* were expressed in reduction percentage of microorganisms, using the following formula: the difference between the CFU/mL at time "Zero" and the CFU/mL at the end of 6 and 12 h (ASTM, 2001).

Likewise, the air flow in the column was 646 mL / min and the water treated was simulated from the "Chinampera" zone; pH 7, EC in a range of 800 and 1500 µs/cm.



Figure 1: Functionalization of gravel with MoSe

2.4. Live / Dead bacterial viability assay

The best treatment was selected to observe the effect of gravel with *Mo*Se on the inhibition of *E. coli*. The samples stained with a BacLight double staining kit to determine the viability of adhered and unadhered bacteria. BacLight uses two nucleic acid stains: green SYTO 9 fluorescent stain (can penetrate all bacteria) and propidium iodide red fluorescent stain (can penetrate only bacteria with damaged membranes), samples obtained before and after of the treatments evidenced by fluorescence microscope (Jerri *et al.*, 2012).

2.5. Fractional 2⁷⁻⁴ experimental design using gravel as filter material

The effect of gravel functionalized with *Mo*Se on inhibition of *E. coli* (Inh*Ec*) was tested using a 2⁷⁻⁴ fractional factorial design according to study of Infante *et al.*, (2021) with some modifications, for a total of eight treatments (Montgomery, 2012) (Table 1). The fixed factors were seed drying temperature (DT), aeration (AR), seed extract concentration (CN), electric conductivity (CD), hydraulic retention time (HRT), gravel particle size (PS) and lysis time hours (LT). The matrix between treatments and independent factors is presented in Table 2.

Factors	Levels		
		Low	High
		-1	1
Seed drying temperature (°C)	DT	60	100
Aeration	AR	0	646
(mL/min)			
Concentration (%)	CN	5	10
Electric Conductivity	EC	800	1500
(µs/cm)			
Hydraulic retention time (h)	HRT	6	12
Gravel particle size (mm)	PS	3	5
Lysis Time (hours)	LT	2	3

Table 1: Fractional factorial design 2⁷⁻⁴ between the levels of the independent factors

Table 2: Matrix of the fractional factorial design 2 ⁷⁻⁴							
Treatment	DT	AR	CN	EC	HRT	PS	LT
	(°C)	(mL/min)	(%)	(µs/cm)	(hours)	(mm)	(hours)
T1	60(-1)	0(-1)	5(-1)	1500(1)	12(1)	5(1)	6(-1)
T2	100(1)	0(-1)	5(-1)	800(-1)	6(-1)	5(1)	12(1)
Т3	60(-1)	646(1)	5(-1)	800(-1)	12(1)	3(-1)	12(1)
Τ4	100(1)	646(1)	5(-1)	1500(1)	6(-1)	3(-1)	6(-1)
Т5	60(-1)	0(-1)	10(1)	1500(1)	6(-1)	3(-1)	12(1)
Т6	100(1)	0(-1)	10(1)	1500(1)	12(1)	3(-1)	6(-1)
Τ7	60(-1)	646(1)	10(1)	800(-1)	6(-1)	5(1)	6(-1)
Т8	100(1)	646(1)	10(1)	1500(1)	12(1)	5(1)	12(1)

DT (drying temperature), AR (aeration), CN (seed extract concentration), EC (electric conductivity), HRT (hydraulic retention time) PS (gravel particle size) and LT (lysis time).

2.6. NFT hydroponics system installation

The hydroponic system was installed in the Chinampa Aurora, Cuemanco of the Chinampera Zone, Mexico. Initially, a biofilter composed of inorganic supports and reeds from the region was used as a secondary treatment coupled to the tertiary treatment for water purification (Rodríguez-Vázguez, 2018). The NFT hydroponic system selected was a closed system to avoid disturbances due to environmental conditions such as sun, rain, strong winds and cold that could disturb its operation. The system comprised three sections, the first a container where the water treated by a system of biofilters is placed, in this container there is a first filtration that isolates large dissolved solids, the second section is a channel covered with PVC material that presents a slope of 1% with a filtration system consisting of a section of gravel with two types of particle size, the smallest size (3 mm) was provided at the beginning of the system and the largest size (5 mm) distributed from the middle to the end, this system presented a length of 12 meters where lettuce plants (*Lactucca sativa*) were cultivated because they are those crops of easy and fast growth (Bawiec *et al.*, 2016). Finally, the tertiary section was adapted to cork-base filters functionalized with *Mo*Se. In each section NFT system, physical-chemical parameters and *E. coli* were monitored for 3 months.

2.7. Statistical analysis

The analysis of variance (ANOVA) was used to test the hypothesis. The alpha value was determined for a 95% reliability (Alpha = 0.05). The null hypothesis always postulated equality between treatments of a 2^{7-4} fractional design, while the alternative hypothesis always postulated that at least one of the treatments produces a different effect. The experiments were performed in triplicate. Additionally, the generalized minimum regression analysis and the test of comparison of means of LSD significant minimum difference were used after having used the student's t-test for the comparison of two means with weighted variance when in the ANOVA they presented significant differences. The SAS program (The SAS System version 9.1, SAS Institute, 2002) was used for ANOVA and LSD testing.

3. Results

3.1. Microbiological characterization of water from the Chinampera zone

According to the presumptive bacterial isolates of fecal coliforms from the water channels of the Chinampera Zone, the molecular analysis was carried out of the sequence of the 16S ribosomal fragment of the strain, in which there was a 99% similarity with the sequence of the strain NBRC 102203 (accession number: NR_114042.1) of the species *E. coli*. The presence of this strain is of great clinical importance, compromising the urinary tract system, being resistant to different antibiotics (ampicillin, erythromycin and vancomycin) (Infante *et al.*, 2021).

3.2. Functionalization of gravel using seeds extract of *M. oleifera* on *E. coli*

In the present study, a biofilter with gravel functionalized of *Mo*Se was evaluated using a 2^{7-4} fractional factorial design to increase the reduction of the logarithmic units of *E. coli*. Initially, a percentage above 90% reduction of the bacteria was obtained in most of the treatments, as is shown in Table 3.

The statistical analysis was obtained in the ANOVA of the generalized linear procedure (GLM), a significance level of p<0.0001, presenting a moderately high positive correlation in which the independent factors that presented a significant effect were aeration, particle size and lysis time. Additionally, the fit of the linear regression model was p<0.0001 and R² =0.81 and the model equation was the following: Inh(%)=98.42+0.20 Air-0.12PS-0.35LT with the significant independent factors being aeration (level high), particle size (low level) and lysis time (low level). When comparing means between treatments using the LSD test, differences were observed, forming 3 groups: (4 and 7), (3, 1 and 6) and (2, 8 and 5). In which treatment 4 was the one that obtained the highest % removal of *E. coli* compared to the other treatments with a percentage of bacterial inhibition of 99.10%.

Relevant conditions in treatment 4 were established by the following factors: (100 °C) seed drying temperature, (646 ml/min) aeration, (5%) seed extract concentration, (1500 μ s/cm) electric conductivity, (3 mm) gravel particle size and (6 h) lysis time. The control without gravel functionalized presented a bacterial amount with an approximate value of ~2.9 x 10⁵ CFU/mL while the gravel functionalized presented a reduction of 3 logarithmic units with a value of 8.9 x 10² CFU/mL, evidencing the functionalization of the material with antimicrobial properties from *Mo*Se on *E. coli* inhibitory effect.

These results are similar than those performed by Schulze-Makuch (2002) showing that adsorbent materials used in wetland systems such as biosurfactant modified zeolite presented adsorption and inhibition effect between 99-100% of *E. coli*, while the coarse granular material with a particle size of (5-25 mm) inactive between 0.1 - 2.7 logarithmic units of *E. coli*. However, it was shown that the finer the material with a particle size between 2-13 mm can reach a bacterial inactivation of 0.7 to 3.4 logarithmic units. Finally, these conditions reached the permissible parameters to use water for animal consumption and for irrigation of crops.

Treatments	Gravel CFU/mL	<i>f</i> -gravel UFC/mL	Inh <i>Ec</i> (%)	
 T1	3.7x10⁵	8.0x10 ³	97.95	
T2	2.4x10 ⁵	9.7x10 ³	96.01	
Т3	3.5x10 ⁵	7.4x10 ³	97.59	
Τ4	2.9x10 ⁵	8.9x10 ²	99.10	
Τ5	2.2x10 ⁵	9.9x10 ³	95.61	
Т6	4.1x10 ⁵	7.7x10 ²	98.43	
Τ7	1.2x10 ⁵	7.5x10 ²	98.90	
Т8	1.1x10⁵	9.2x10 ³	91.85	

Table 3: Percentage of inhibition of <i>E. coli</i> using biofilters with gravel porous material
functionalized

DT (Drying temperature), AR (aeration), CN (Concentration *of MoSe*), EC (Electrical conductivity), HRT (Hydraulic retention time) PS (Gravel particle size), LT (Lysis time) and Inh *Ec* (Inhibition of *E. coli*).

3.3. Live/Dead bacterial viability Assay

In Figure 2, staining of the bacterial strain was observed to determine the effect of cell inhibition using *M. oleifera* seed extract functionalized with gravel. In Figure 2a, the control treatment is found without gravel functionalized while in Figure 2b, the treatment was with the gravel functionalizated of *MoSe*, in which the inhibitory effect on *E. coli* was evidenced. The studies carried out by Xiong *et al.* (2017) in which they functionalized the extract of *M. oleifera* in biofilters with sand as a filtering material, evidencing that the cationic protein present in the seeds could be adsorbed in the sand through the electrostatic attraction, maintaining the coagulant-flocculant capacity and the antimicrobial activity improving the removal of pathogens and avoiding the release of organic matter. Thus, that strategy obtained an improvement with the functionalized sand in the elimination of 3-4 log of 1 μ m particles, these particles of 1 μ m in size were chosen because they represent many targets microbial contaminants (such as coliform bacteria) and the efficiency of the collector is usually the lowest in this size.

Likewise, when comparing the present study with the investigations carried out by García-Varela *et al.* (2015), in which they evaluated the antimicrobial activity of phenolic compounds of plants for the inhibition of *E. coli*, they indicated that there are several possible mechanisms by which phytochemical compounds can produce an antimicrobial effect: possible alteration of the physicochemical properties of the membrane plasma, pore formation, DNA gyrase and inhibition of nucleic acid synthesis and toxicity through hydrogen peroxide formation. However, the effect observed by the extracts on the tested microorganisms suggests that they could directly interfere with the membrane structure by creating pores, due to the time required to produce inhibition. This mechanism could very well explain the images in Figure 2, obtained by fluorescence microscopy where the permeability of the cell membrane is needed for propidium iodide (PI) to enter the cell and adhere to the DNA, indicating that the microorganisms are compromised.

Figure 2: Live/Dead Baclight test carried out with fluorescence microscopy from samples with gravel functionalized with *Mo*Se



(a)

(b)

Biofilter without *f*-gravel (**a**), Biofilter functionalized with *f*-gravel (**b**). Dead cells are stained in red, while green-stained cells are alive.

3.3. Biofilter system and NFT hydroponics system

Different stages of the Chinampa channel and biofilter system were monitored in the physicochemical and microbiological parameters. The treatment operation of the biofilters were evaluated for a period of 3 months as is shown in Table 4. The physicochemical parameters that had a greater effect with the use of the systems, they were: pH, (DO) dissolved oxygen, (EC) electric conductivity, (TDS) total dissolved solids, and *E. coli*.

Physicochemical and microbiological analyses were carried out on the treated water samples from each part of the hydroponic system, mainly observing that the system favours the reduction of *E. coli*. Likewise, it tends to improve pH stability with cork, which is an indicator that influences the absorption of nutrients in the crop.

The inhibition of bacteria with cork biofilters could be due to its lignin, suberin and tannin molecules that have antimicrobial characteristics (Olivella *et al.*, 2013). Therefore, to demonstrate the effect of the functionalization of cork with *Mo*Se extract, the studies carried out by Infante *et al.* (2021) confirmed the decrease in copies of the LacZ gene and the percentage of viability due to the improved inhibition of *E. coli*, potentiating the antimicrobial activity together with some components of the cork that generate synergy and a greater chemisorption effect.

Parameters	ters Water Biofilter		NFT	Cork	Cork-f	* Allowable
	Channel		System			limits
рН	7.79	7.66	9.11	7.28	7.6	7.0

Table 4: Evaluation of physicochemical and microbiological parameters of the Chinampa and monitoring of the system

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$\text{DO}~(\frac{mg}{L})$	0.85	3.01	2.87	1.86	1.87	>6
EC $(\frac{\mu S}{cm})$	1115	1191	2708	1832	1972	0-300
$\textbf{TDS}\;(\frac{mg}{L})$	567	570	1354	916	986	<1000
<i>E. coli</i> (CFU/mL)	2.5x10 ¹	1.5x10 ¹	1x10 ¹	<10 ¹	<10 ¹	1x10 ²

*Permissible limits for use in agricultural irrigation in accordance with the standard (NOM-001-SEMARNAT-1996). Dissolved oxygen (DO); Electrical Conductivity (EC) and Total dissolved solids (TDS).

In relation to the microbiological analyses of the hydroponics system plants selected, *E. coli* was detected in the root, leaves and phenolic sponge (Table 5). These results indicated that in the plant where the gravel filtration system began, it had a higher density and abundance of roots as its length compared to other plants, showing a high inhibition of *E. coli* in the confirmatory medium (methylene blue eosin) EMB.

In contrast, the roots the bacteria grown in the Luria-Bertani LB medium, being one of the most used for the culture of *Escherichia coli* and other bacterial species, because it is rich in nutrients, mainly tryptone, which is the product of the digestion of casein from milk with pancreatic enzymes, so all those lactose-fermenting bacteria grew in this medium, These results are consistent with the studies carried out by (Ottoson *et al.*, 2005) who reported the inhibition of pathogenic microorganisms in the root system by absorption and adsorption processes of the plant contributing to the microbial elimination in the system.

The process of elimination of pathogenic microorganisms involves a series of mechanisms, such as filtration, which is provided by the gravel where microorganisms are eliminated and through antibiosis, a process where microorganisms interact with each other biologically resulting in the reduction of their population (Martin *et al.*, 2013).

The capacity of the hydroponic system for the elimination of pathogens in wastewater presented the elimination of fecal coliforms. Furthermore, the capacity of the hydroponic wastewater treatment system for the elimination of microorganisms was evidenced in the present study and was consistent with the results obtained by Ottoson *et al.* (2005) their study included the analysis of untreated samples, in treated and untreated wastewater for fecal microbial indicators (Coliforms, *E. coli, Clostridium perfrigens* spores and somatic coliphages) indicating that hydroponics wastewater treatment removed microorganisms satisfactorily.

Lactucca	sativa	<i>E. coli</i> (CFU/mL)					
Samples		I	Root	Lea	ives	Phenolic Sponge	
	Total weight (g)	LB	EMB	LB	EMB	LB	EMB
1	9.9	36 x 10 ²	<10 ¹	21 x 10 ²	<10 ¹	50 x 10 ²	30
2	3.1	17 x10 ¹	<10 ¹	25 x 10 ³	<10 ¹	21 x 10 ¹	<10 ¹
3	4.5	13 x 10 ³	<10 ¹	50 x 10¹	<10 ¹	32 x1 0 ¹	<10 ¹

Table 5: Detection of *E. coli* cells in lettuce plants grown in the hydroponics system

4. Conclusions

Nature-based solutions as strategies in the near future for food security through water conservation and elimination of pathogenic microorganisms. This study showed the maximal inhibitory effect of gravel functionalized with *Mo*Se on *E. coli* above 99.10%, evidencing antimicrobial properties by fluorescence microscopy. Likewise, the NFT hydroponic system was accoupled to cork-base filters system functionalized with *Mo*Se to increase the potential for antimicrobial activity and effectively reduced the growth of *E. coli* with a value <10¹ CFU/mL in water pretreated in a channel of Chinampera Zone. It was evidenced that the NFT hydroponic system was able to reduce pathogenic microorganisms such as *E. coli*. Likewise, the clarification of the water improved, which indicates the efficiency of the system with a high potential in the treatment of wastewater to reuse in agricultural systems and for vulnerable communities similar to the intervened in the Chinampera Zone, providing agri-food security, being a biotechnology response strategy for the advancement of sustainable development goal 6 (SDG6) to guarantee the availability and sustainable management of water and sanitation for all.

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