

## Eco-efficiency analysis of a continuous two-phase partitioning bioreactor treating 2,4-dichlorophenol synthetic wastewater

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### ABSTRACT

Continuous two-phase partitioning bioreactors are a promising solution for removing poorly biodegradable pollutants, such as phenolic compounds, from wastewater. However, their combined economic and environmental feasibility has not yet been jointly evaluated, and it is crucial to ensure a minimum incorporation of sustainability aspects during design.

For this purpose, four methodologies were employed. Standalone analyses were first conducted using Life Cycle Assessment and Life Cycle Costing, followed by an integrated environmental–economic sustainability assessment. The latter consisted of an eco-efficiency approach and a qualitative evaluation based on the principles of the EU Taxonomy, which functions as a key instrument within the European sustainable legal framework. The methodologies were applied to four scenarios in which the concentrations of 2,4-dichlorophenol, as targeted model phenolic compound, in the influent and the recycling ratio of the treated effluent were varied. The findings revealed that integrating recycling within the reactor resulted in a significant environmental burden across categories, with exception of ecotoxicity (with 98.2% decrease). Although the technology successfully achieved its removal objectives, the higher consumption of utilities required for proper reactor operation reduced the expected benefits. As for operating expenses, differences between scenarios were minimal due to the dominance of fixed costs, which remain constant in all cases. Comparatively, the technology aligned with performance metrics found in existing literature (around 13.7 kg CO<sub>2</sub>eq./m<sup>3</sup> of treated wastewater and costs above 7.62 €/m<sup>3</sup>). To enhance the competitiveness of the technology, it is essential to manage energy demands for heating and aeration and to increase process automation, thereby reducing associated labor costs.

### 1. Introduction

Industrial effluents often contain xenobiotic pollutants, such as pharmaceuticals, pesticides, microplastics, endocrine disruptors and aromatic compounds, which are highly recalcitrant and partly degraded in conventional wastewater treatment systems [1]. Within this broad spectrum of organic pollutants, phenolic compounds, such as phenol, cresols, chlorophenols, nitrophenols and alkylphenols, represent a significant threat to the environment. This is due to the high volumes produced and released by various anthropogenic activities (approximately 10 million tons annually), their high organic load and the toxicological effects associated with the formation

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of free radicals [2,3]. These compounds are widely present in industrial wastewater from different sectors such as textiles, tanneries, coke ovens, rubber manufacturing, pulp and paper, fiberglass and pharmaceuticals, among others [4].

The effectiveness of conventional wastewater treatment of these compounds has been called into question, as maximum reported removal efficiencies reach only 60% [5]. Consequently, innovative alternatives have been explored. Among these options, wet air oxidation and catalytic wet air oxidation processes have been shown to be effective for phenol removal, although they require high pressure and temperature conditions (20–200 bar, 200–320 °C), which represent a large energy demand and operating costs [6]. On the other hand, systems based on physical processes, such as solvent extraction, adsorption, pervaporation, membrane extraction, reverse osmosis and nanofiltration, only transfer the contaminant from the main flow to another phase [7]. However, these technologies are generally less energy efficient and encounter higher cost and environmental impact compared to biological alternatives [8]. Nature-based solutions, such as constructed wetlands, represent feasible alternatives for the treatment of the recalcitrant organic fraction of wastewater, achieving average removal efficiencies exceeding 80%. Their effectiveness is attributed to the combined action of several mechanisms, including phytodegradation, rhizofiltration, microbial degradation, and sorption [9]. However, these systems also present inherent limitations, such as limited tolerance to high phenol concentrations, slow treatment kinetics, large land requirements, sensitivity to environmental conditions, restricted process control and optimization, and the potential accumulation of phenols in plant tissues and sediments [10].

Regarding aerobic and anaerobic biological processes, their effectiveness can also be affected by substrate inhibitions due to decreased biocatalytic activity in the presence of high contaminant concentrations. In this context, two-phase partitioning bioreactors (TPPBs) arise as a promising technology to effectively control the exposure of biomass to high concentrations of pollutants [11]. This technology has shown satisfactory results in the treatment of waste containing phenols, biphenyls, indole, polyaromatic hydrocarbons and polychlorinated biphenyls [12].

However, most of the previous research on this technology has been conducted in batch mode [13]. Aiming to enhance performance from this operating mode in terms of process control flexibility and removal efficiency, continuous operation has been less frequently explored across literature. Some related practical examples of treatment have been applied for fracking fluids, hypersaline streams, tannery wastewater and chlorophenol-containing effluents [14–18]. In this framework, the continuous two-phase partitioning bioreactor (C-TPPB) is operated with a polymeric tubing immersed in the bulk phase of the bioreactor containing the microbial culture with the toxic wastewater flowing tubing-side. This approach enables the gradual transfer of the contaminant to the microbial culture, maintaining microorganism exposure at sub-inhibitory concentrations [16,18].

While the technological performance of this C-TPPB—particularly its removal efficiency—has been demonstrated, it remains essential to identify the factors influencing the environmental profile of the system from the earliest design stages since it serves as a direction-setting and risk-screening tool that guides process choices before scale-up decisions become locked in and, thus ensuring that the technology effectively minimizes the environmental impact from an integral perspective, including not only environmental indicators but also the potential social and economic benefits. TPPBs have already been analyzed from both economic and environmental perspectives [19]. However, that study focused on a separate enviro-economic analysis, comparing aerobic and anaerobic conditions for industrial wastewater treatment while assessing the potential use of a polymer to control the diffusion of the contaminants to the biomass. Beyond this distinct objective, the reactor has operated in sequencing batch mode, targeted a different pollutant from the textile industry, functioned at a larger scale (full industrial rather than pilot), and utilized an amorphous polymer in beads instead of tubing).

To develop a comprehensive sustainability assessment applicable to TPPBs, various methodologies and evaluation frameworks can be considered [20]. Among the possible tools for a combined economic-environmental approach are the following: System of Environmental-Economic Accounting (SEEA), Eco-Efficiency Analysis (EEA), EU Taxonomy, Contingent Valuation Method (CVM) and the use of composite indicators [21–23]. Eco-Efficiency is a multidimensional and flexible methodology that can be applied through a life cycle approach, following the guidelines of ISO 14,045, or by integrating tools such as Life Cycle Analysis (LCA) and Data Envelopment Analysis (DEA), or even by comparing performance indicators, among others. In the wastewater treatment sector, the use of indicators for EEA can be found in several specialized publications in this field [24–27]. An extended literature review for eco-efficiency analysis in wastewater treatment is reported in Supplementary Materias. Eco-Efficiency has been applied primarily to urban wastewater treatment, with dairy and petroleum refinery effluents as examples of industrial wastewater [25,28]. It is important to note that most EEA studies on wastewater treatment technologies have focused on large-scale applications, with a few exceptions, such as studies on fluidized bed anaerobic membrane bioreactors and aerobic membrane bioreactors for black and grey water treatment [29,30]. Studying the TPPB for treating phenolic wastewater will offer valuable insights to enhance eco-efficiency understanding in industrial wastewater treatment at pilot and small scales. Applying eco-efficiency analysis during scale-up helps reveal environmental-economic trade-offs while also identifying eco-efficient design pathways and supporting defensible, long-term infrastructure decisions. This is particularly important because costs and environmental impacts at non-full scales are disproportionately high caused by different orders of magnitude of the material and energy flow data. There is no predictable straightforward relationship guaranteeing that cost or environmental impacts increases more rapidly than the others since they are governed by different mechanisms [31].

While eco-efficiency serves as a methodology for assessing processes and products, the EU Taxonomy provides a framework that links investment decisions and economic activities with environmental criteria at the company level. Because of this, few studies address the qualitative and/or quantitative implications of implementing the EU Taxonomy at process level [32]. On the qualitative side, the EU Taxonomy has been considered a key instrument for the development of environmental, social and governance (ESG) ratings, assessing their contribution to the transition towards climate neutrality [33]. Regarding its applicability at company level, the effects of incorporating the EU Taxonomy in the wastewater treatment sector and the suitability of its criteria at pilot, benchmark or

process scale have not yet been analyzed.

In this context, this study aims to assess whether implementing an effluent recycling rate that partially returns the effluent to the bioreactor is a viable strategy, considering not only effluent quality but also overall enviro-economic sustainability. The analysis was approached from conceptual design using experimental data at different concentrations to process modelling, which should be able to provide scale-up inventory data for impact and cost assessment. This scaled up is necessary for any sustainability analysis since the goal is to determine critical parameters and comparative of process routes or scenarios of operation and thus supports in the shaping of better design trajectories rather than producing accurate impact results. With a life cycle approach, the EEA methodology was applied to comprehensively identify the key environmental and economic factors influencing the operation of the phenolic wastewater treatment process. EEA indicators were developed based on category results and product value assessments derived from LCA and LCC analyses. The rationale behind this step-by-step validation is the limited availability of eco-efficiency studies beyond domestic treatment and large-scale applications. Finally, to demonstrate the practical relevance of these methodologies in shaping real-world policies, the findings from the previous stages were aligned with EU Taxonomy environmental eligibility qualities. Both substantial contribution and Do Not Significant Harm (DNSH) criteria have been taken into account for a lower technology-readiness level (TRL) for its six objectives (i.e., climate change mitigation, climate change adaptation, sustainable use and protection of water and marine resources, transition to a circular economy, pollution prevention and control, protection and restoration of biodiversity).

## 2. Methodology

### 2.1. Experimental scenarios

The C-TPPB system employed in this study was composed of a 3-L glass vessel comprising a permeable polymeric tubing (Hytrek G3548) submerged in microbial culture. The affinity between the target pollutant and the polymer enabled the contaminant diffusion through the tubing walls into the aqueous phase of the bioreactor, where the biodegradation occurred [34]. The mass transfer process was governed by the concentration gradient across the tubing walls, which is influenced by substrate biodegradation rate. When its biodegradation occurs, to restore equilibrium, the contaminant was gradually transferred from wastewater to the microbial culture, resulting in the microorganisms being exposed to sub-inhibitory concentrations.

The bioreactor was equipped with a thermostatic probe to control the temperature, a magnetic stirrer and a cylindrical grid support for the tubing, as described in Tomei and Mosca Angelucci [18].

The target contaminant was 2,4-dichlorophenol (2,4-DCP), used as a model phenolic wastewater; therefore, the study does not represent complex real wastewater but serves as a preliminary assessment of critical parameters. Operating conditions included influent aqueous solution of 2,4-DCP concentrations of 700–900 mg/L and effluent/feed recycle ratios (ERR) of 0, 0.3, and 0.5. These concentrations fall within literature ranges (9–6800 mg/L) [8], and four experimental scenarios were evaluated to identify optimal conditions.

- **Scenario BL (baseline):** Operation at 700 mg/L of 2,4-DCP without recirculation.
- **Scenario BR03:** Same concentration conditions as BL (700 mg/L), but with an ERR of 0.3.
- **Scenario HR03:** High concentration of 2,4-DCP (900 mg/L) and ERR of 0.3.
- **Scenario HR05:** 2,4-DCP concentration of 900 mg/L with an ERR of 0.5.

All scenarios were run at 28 °C and pH 7.5, using a biomass yield coefficient of 0.633 gCOD/gCOD and an endogenous decay rate of 0.06 g/g·d, as estimated by Tomei and Mosca Angelucci [18]. The data gathered allowed evaluating the impact of organic loading and ERR on the environmental performance (see Table 1). Information from laboratory experiments incorporating changes in the hydraulic residence time and operational other parameters were not studied in this research and thus, the enviro-economic sustainability of scenarios related to these aspects could not be proposed.

### 2.2. Scale-up of the C-TPPB-based system

Based on the laboratory-scale operation data from Table 1, a model was designed for industrial scale-up considering the principles of geometric, kinematic, dynamic, thermal and chemical similarities to perform mass and energy balances. The scaled reactor had a

**Table 1**

Operational laboratory data needed for process design and Life Cycle Impact Assessment [18].

Technical parameters	BL	BR03	HR03	HR05
Organic loading rate of the 2,4-DCP (kg/m <sup>3</sup> ·d)	0.185	0.186	0.225	0.230
Concentration of 2,4-DCP in the influent (mg/L)	721.52	726.58	878.48	898.73
Concentration of 2,4-DCP in the tubing effluent (mg/L)	9.20	4.73	4.81	4.85
Mineral stock solution (mL/L influent)	141.81	142.80	172.66	176.64
Biomass concentration (gVSS/L)	1.16	1.26	1.17	1.06

Note: BL: Baseline scenario; BR03: 700 mg/L and 0.3 recycling ratio; HR03: 900 mg/L and 0.3 recycling ratio; HR05: 900 mg/L and 0.5 recycling ratio; COD: Chemical Oxygen Demand; VSS: Volatile Suspended Solids; 2,4-DCP: 2,4-Dichlorophenol.

volume of 100 L, a height/diameter ratio of 3.3, a headspace of 15 % of the total volume and was equipped with thermal jacket to maintain the temperature [35,36]. The fluid velocity inside the tubing located within the bioreactor was kept constant, and thus, it resulted in 3.46 cm inner diameter and 3.4 m length. Synthetic phenolic wastewater (with the target contaminant indicated in the previous Section 2.1.) was continuously fed to the tubing with a flow rate of 25.6 L/d, increasing in scenarios with recirculation to 33.28 L/d (BR03 and HR03) and 38.4 L/d (HR05). The sludge outflow was assumed to be negligible due to the high solid retention time, according to Tomei and Mosca Angelucci (2019)

As shown in Fig. 1, the system included two pumps (for feed and recirculation), a blower, an influent storage tank and a buffer tank. The influent pump provided the feed at the set flow rate and the recycle pump was used in the last three scenarios (BR03, HR03 and HR05). The blower was another energy consuming device and was necessary to maintain aerobic conditions as well as for efficient mixing, as the implementation of mechanical stirrers was limited by the use of the in-vessel polymeric tubing. In addition to energy, a mineral medium was constantly dosed into the reactor to ensure a C/N/P ratio of 100:5:1. The proportion of the different nutrients fed to the system was chosen based on the optimal generally formulated ratio for heterotrophic bacteria which is enough to preserve community structure, prevent opportunistic species from dominating and stabilize functional performance when using synthetic wastewaters and continuously stirred tanks [37].

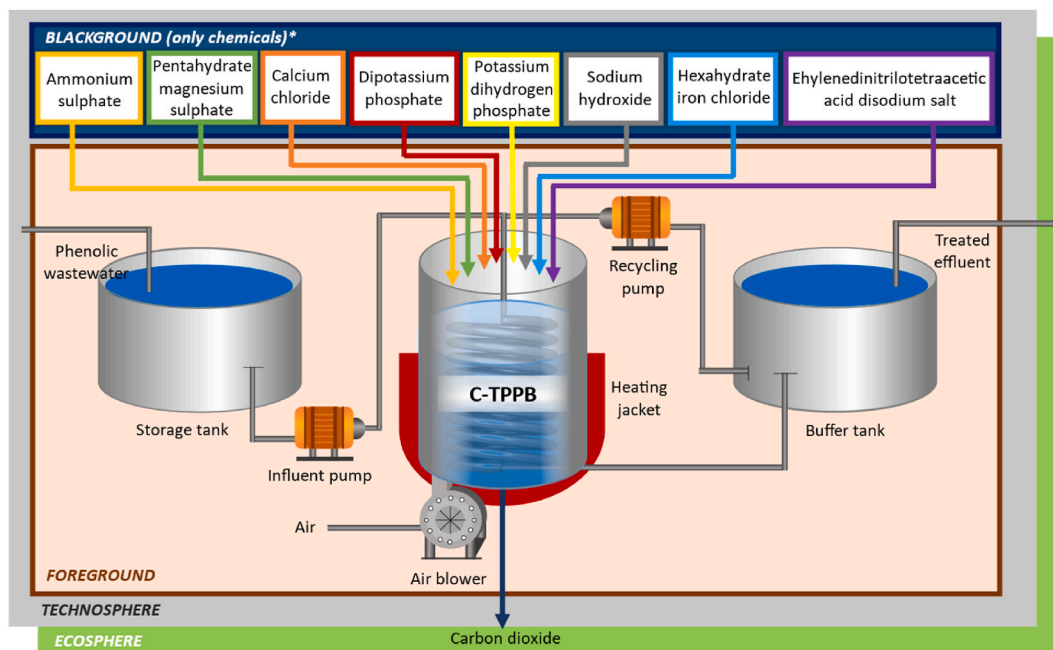
### 2.3. Life Cycle Assessment

The LCA methodology was performed to assess the environmental impacts of the different alternative scenarios and thus, to be used in decision-making criteria for risk and priority technical solutions identification. In accordance with ISO 14,040/14,044:2006 standards, the four LCA stages were followed: definition of the goal and scope, life cycle inventory analysis, life cycle impact assessment and interpretation of the results [38,39]. Therefore, the methodological choices (i.e., system boundaries, functional unit, selection of impact categories ...) were discussed across this section. Summarized information is shown in Table S7 (Supplementary Materials).

#### 2.3.1. Goal and scope

The C-TPPB treatment system was designed to reduce or eliminate the pollutant load of a 2,4-DCP industrial effluent, considered representative of phenolic wastewaters, to enable safe discharge. All environmental impacts associated with the system boundary elements were allocated exclusively to the wastewater treatment function. Consequently, the LCA applied allocation at the point of substitution (APOS), did not include system expansion, and could also be classified as attributional. Consistent with the process function, the volume of wastewater treated was selected as the functional unit (FU), facilitating comparison with treatment processes reported in the literature.

With respect to the technical system boundaries, the assessment was limited to a *Cradle-to-gate* life cycle, excluding construction,



**Fig. 1.** Configuration of the treatment process and definition of the system boundaries. Although the background sub-system in the figure only displays the chemicals, the asterisk indicates that the electricity for the pumps and the blower and the heat of the jacket are also part of the sub-system.

maintenance, and end-of-life phases. This is in line with approximately 57% of LCAs on wastewater treatment technologies since they have only focused on the operational phase [40]. Material and energy inputs, as well as emissions, were classified into foreground and background subsystems (Fig. 1). Background inventory data were obtained from the Ecoinvent® database, adopting European geographical boundaries.

### 2.3.2. Life cycle inventory

The Life Cycle Inventory (LCI) was built on the scaled-up data from Section 2.2. The compilation was performed with a "bottom-up" approach, with in-out flows quantified for the foreground subsystem. Table 2 shows the LCI disaggregated into inputs from the technosphere and outputs to nature. Indirect emissions to nature were calculated as part of a background process taken from Ecoinvent®. Direct *gate-to-gate* emissions, on the other hand, were only CO<sub>2</sub> from biodegradation of organic matter and residual 2,4-DCP in the effluent. Since the CO<sub>2</sub> emissions originated from an industrial effluent, they could not be assumed to be biogenic and were therefore estimated from the degradation of organic compounds, as this parameter could not be measured directly during the laboratory experiments.

### 2.3.3. Life cycle impact assessment

Life Cycle Impact Assessment (LCIA) represents the phase of the LCA in which the elementary flows listed in Table 2 were translated into environmental impact indicators. This stage was performed using SimaPro 9.4.0.2 software and the ReCiPe 2016 Midpoint and Endpoint (H) V1.07/World (2010) impact assessment methods. Although several LCIA methods are available and ISO standards do not prescribe a specific method, ReCiPe 2016 was selected due to its widespread application in wastewater treatment studies.

Within the ReCiPe Midpoint framework, only the 7 most relevant impact categories were considered, each contributing more than 5% relative to the dominant impact category, human carcinogenic toxicity. Accordingly, the LCA results were interpreted based on the following indicators: Global Warming Potential (GWP, kg CO<sub>2</sub> eq.), Ionizing Radiation (IR, kBq Co-60 eq.), Freshwater Eutrophication (FE, kg P eq.), Freshwater Ecotoxicity (FET, kg 1,4-DCB eq.), Marine Ecotoxicity (MET, kg 1,4-DCB eq.), Human Carcinogenic Toxicity (HCT, kg 1,4-DCB eq.), and Fossil Resource Scarcity (FRS, kg oil eq.).

In addition, the ReCiPe Endpoint method was applied to obtain a single, aggregated damage score, enabling a comprehensive and comparable evaluation of the overall environmental consequences of the treatment system.

## 2.4. Life Cycle Costing

LCC was conducted following ISO 15,686–5:2017 and ISO 15,663:2021 in four steps: goal and scope definition, structured cost breakdown, modeling and analysis, and interpretation of results [41,42]. Since the first and last steps align with LCA, this section focuses on the cost breakdown and modeling.

The LCC primarily estimated operational costs, consistent with the European Green Public Procurement Criteria for Wastewater Infrastructure, which note that operational expenses often exceed initial investments [43]. Construction costs were included only to determine fixed costs as a proportion of the initial investment (Table S13). Operational costs encompassed labor, consumables, energy, insurance, supervision, land rent, plant maintenance, and environmental expenses (Table S14–S15). Annual cost increases were

**Table 2**

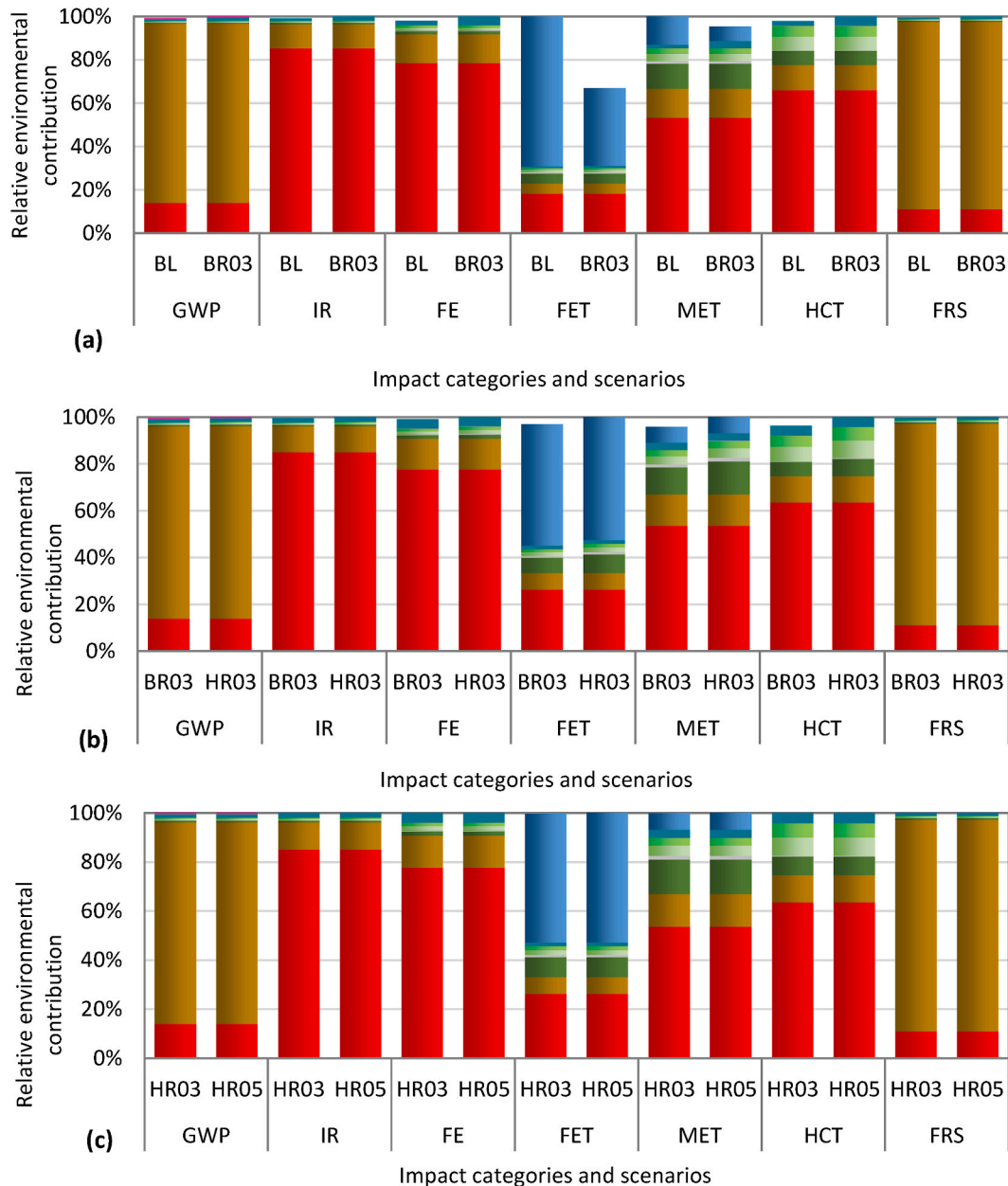
Life cycle inventory of the selected four 2,4-Dichlorophenol treatment scenarios expressed per functional unit or 1 m<sup>3</sup> of treated wastewater. Chemicals and emissions are expressed in g and energy in kWh.

LCI materials	BL	BR03	HR03	HR05
<b>Inputs from Technosphere</b>				
<i>Chemicals</i>				
Ammonium sulphate	709.04	709.04	863.29	863.29
Pentahydrate magnesium sulphate	141.81	141.81	172.66	172.66
Calcium Chloride	7.09	7.09	8.63	8.63
Dipotassium phosphate	155.99	155.99	189.92	189.92
Potassium dihydrogen phosphate	120.54	120.54	146.76	146.76
Hexahydrate iron chloride	0.29	0.29	0.35	0.35
NaEDTA	0.29	0.29	0.35	0.35
Sodium hydroxide	913.12	1826.25	1826.25	1826.25
<i>Energy</i>				
Blower	5.15	5.15	5.15	5.15
Heating	41.56	41.56	41.56	41.56
Influent pump	5.24·10 <sup>-7</sup>	5.24·10 <sup>-7</sup>	5.24·10 <sup>-7</sup>	5.24·10 <sup>-7</sup>
Recycling pump	0.00	1.07·10 <sup>-8</sup>	1.07·10 <sup>-8</sup>	2.98·10 <sup>-8</sup>
<b>Outputs to the Nature</b>				
<i>Emissions</i>				
Carbon dioxide (CO <sub>2</sub> )	45.07	45.07	55.29	55.28
2,4-Dichlorophenol	9.20	4.73	4.81	4.85

Note: BL: Baseline scenario; BR03: 700 mg/L and 0.3 recycling ratio; HR03: 900 mg/L and 0.3 recycling ratio; HR05: 900 mg/L and 0.5 recycling ratio; LCI: Life Cycle Inventory NaEDTA: Ethylenedinitrilotetraacetic acid disodium salt.

calculated using a 2.23% European inflation rate [44].

Construction costs were estimated using factorial methods or as a percentage of initial investment for items such as land, equipment maintenance, and insurance, with a 15% amortization rate [45–48]. End-of-life and decommissioning costs were excluded, and the facility's operating life was assumed to be 30 years. Methodological choices and results are summarized in Table S13.



**Fig. 2.** Relative impact (MidPoint) environmental profile comparison and contribution analysis between BL and BR03 (a), BR03 and HR03 (b), and HR03 and HR05 (c). ■ Blower; ■ Ammonium sulphate; ■ Potassium dihydrogen phosphate; ■ 2,4-Dichlorophenol; ■ Recycling pump; ■ Calcium chloride; ■ Ethylenedinitrilotetraacetic acid; ■ Influent pump; ■ Pentahydrated magnesium sulphate; ■ Hexahydrated iron chloride; ■ Carbon dioxide; ■ Reactor heating; ■ Dipotassium phosphate; ■ Sodium hydroxide. BL: Baseline scenario; BR03: 700 mg/L, 0.3 recycling ratio; GWP: Global Warming Potential; IR: Ionizing radiation; FE: Freshwater Eutrophication; FET: Freshwater Ecotoxicity; MET: Marine Ecotoxicity; HCT: Human Carcinogenic Toxicity; HR03: 900 mg/L, 0.3 recycling ratio; HR05: 900 mg/L, 0.5 recycling ratio; and FRS: Fossil Resource Scarcity.

## 2.5. Eco-efficiency analysis

The EEA sought to correlate the environmental impacts estimated through the LCA with the value product; in this case, the economic value of the scaled-up scenarios and taken from the LCC. Conducted in accordance with ISO 14,054:2012, followed the life cycle approach already considered in LCA and LCC and was limited, as these two analyses, to the operation of the treatment facilities, excluding infrastructure [49].

The results of the analysis are presented using a multi-perspective approach with four quantitative indicators: GWP, FET, FE and an environmental single score per unit cost. The indicators were selected to capture the influence of greenhouse gas emissions, as energy use dominates the environmental conclusions of this research; the potential harm of toxic effluents, a critical category for distinguishing among scenarios (Fig. 2); and the sensitivity of the system to nutrient releases, which is a desirable characteristic for wastewater effluents. In addition, the selected indicators facilitate simplified communication with non-LCA experts and enable the ranking of alternative design scenarios. Moreover, as shown in Table S6, GWP, FE, and FET are the three most frequently studied impact categories in eco-efficiency assessments of WWTPs. Table 3 summarizes some of the methodological choices made for the analysis.

## 2.6. Sensitivity analysis

The sensitivity analysis conducted in this research aimed to explore potential changes in the critical environmental and economic factors that could impact the eco-efficiency profile. These included both technical and methodological approaches (both environmental and economic) to understanding how results may vary depending on the assumptions made. In the first case, the BL scenario was modified to explore the extent to which modifications influence the two critical environmental impacts of phenolic wastewater treatment using the C-TPPB: heating and the simultaneous air supply and stirring with the use of blowers. Four additional scenarios have been provided, considering only one change between each one of them.

- **Baseline scenario (BL):** The heating process was carried out for an open-air reactor, and the electricity demand of the blowers was estimated in accordance with standard recommended practices [50]. The influent stream temperature was assumed to be 28°C, which was the same as the operating temperature.
- **Baseline scenario with changes in the blower energy (BL-B):** The BL scenario was modified to use an electricity demand estimated following the energy balances procedures indicated in Metcalf & Eddy [51].
- **Baseline scenario with a closed-reactor (BL-CR):** The influent wastewater stream entered at 28 °C, and no energy losses occurred as the reactor was assumed to be adiabatic.
- **Baseline scenario with open-reactor and heated influent (BL-HI):** Unlike the BL scenario, the influent was fed at 20 °C and heated to the operating temperature of 28 °C.
- **Best performing scenario (BPS):** For the environmental analysis, the electricity demand of the blower was considered, the reactor was assumed to be adiabatic, and the influent wastewater was already at the operating temperature.

Beyond this technical benchmarking, the environmental assessment of the BL scenario included a methodological sensitivity analysis, incorporating variations in the LCIA methods (ReCiPe and USEtox). This approach aimed at mitigating biases stemming from different characterization factors, which was particularly crucial given the limitations in LCA databases for recalcitrant pollutants. The use of these methods is justified by the significance of the studied compound in toxicity-related impact categories. Also, USEtox stands out for being using European characterization factors and thus, regional specific [52]. Notably, USEtox is the default LCIA method

**Table 3**  
Summary of methodological eco-efficiency choices as described in Section 2.5.

Attribute	Description
Goal of the study	To promote the operation of the facility considering both optimal environmental and economic aspects while removing 2,4-Dichlorophenol
Type of wastewater	Industrial wastewater (with 2,4-dichlorophenol)
Technology	Continuous Two-Phase Partitioning Bioreactor (C-TPPB)
Technical system boundaries	Cradle-to-gate, only operation
Environmental assessment methodology	Life Cycle Assessment
Impact assessment method	ReCiPe 2016 V1.07
Environmental categories selected	Global warming potential, freshwater ecotoxicity and Endpoint ReCiPe single score.
Product system	Technology under development for the removal of phenolics in wastewater
Function of the system	Removal of a recalcitrant compound (2,4-dichlorophenol) present in phenolic wastewater
Product system value	The operational costs of operation of the technology
System value assessment methodology	Life Cycle Costing
Choice of the eco-efficiency indicator	<ol style="list-style-type: none"> <li>1. Global warming potential divided by operational costs</li> <li>2. Freshwater ecotoxicity divided by operational costs</li> <li>3. Freshwater eutrophication divided by operations costs</li> <li>4. Environmental single score divided by operational costs (both ReCiPe and USEtox).</li> </ol>

recommended by the International Reference Life Cycle Data System (ILCD) for assessing toxicity categories [53].

When performing the economic analysis for the C-TPPB, the fixed operational costs were the most contributing element to the profile being caused by labor costs (more information in Section 3.2). Since this is caused by economies of scale, the daily maintenance of the operators was modified from 2 h/d to 2 h/week. The goal was to achieve a fixed cost/investment ratio of 3, comparable to a large-scale facility [54]. This aligns with Jung et al. [55], who proposed part-time labor of 2 h per week, plus 4 h monthly and 16 h annually for maintenance.

Finally, some evidence suggests that the environmental impact of the construction is not negligible, contributing 5%–23% to global warming potential [56]. To assess how eco-efficiency evolves with improved operational environmental performance, the BL and BPS LCA scenarios were compared, with and without fixed costs. Additionally, three alternative scenarios were examined to determine: (1) the impact of labor cost reductions on eco-efficiency, (2) the significance of at least 5% of construction impacts and costs (as a percentage of total investment costs), and (3) the eco-efficiency trend when construction-related environmental costs reach 23%.

## 2.7. Application of the EU taxonomy

In this work, the EU Taxonomy was used to identify economic activities and their related criteria for C-TPPB-based treatment technology. For this study, three of the steps considered by the Regulation (EU) 2020/852 were followed. In this regard, taxonomy-eligible (1) and activities that meet with technical screening criteria (both substantial contribution and DNSH) related to each of the EU taxonomy objectives (2) were identified along with supplementary information for the transition of eligible to aligned activities (3). The selection of these activities was supported by the Taxonomy Compass tool, which digitizes the criteria of the Commission Delegated Regulation (EU) 2021/2139 and Commission Delegated Regulation (EU) 2023/2486.

## 3. Results

### 3.1. Environmental impact analysis

#### 3.1.1. Outcomes of the baseline scenario

The first step in the environmental diagnosis for the selection of the best scenario was the identification of the hotspots in the process. In this way, a complete picture could be provided of how the change in parameters such as effluent recycling ratio and influent concentration were influencing the environmental profile and results. Fig. 2 gives an overview of environmental profile comparison and contribution analysis in pairs of the analyzed scenarios.

Three groups of processes contributed to the BL scenario: energy, chemicals, and direct emissions to air and water. However, the profile was mainly characterized by one of them for more than 66.6% (MET) except for the FET category (23.0%). The intrinsic design of the treatment system has rejected the removal of 2,4-DCP from industrial wastewater in an energy-driven process, which for some impact categories such as FRS accounted for more than 98%. As already described in Section 2.2, energy was consumed inside the treatment process by 4 elements: influent and recirculation pumps (turned off for the BL scenario), heating jacket and air blower. However, the last two yielded results above 99.9% of the total representativeness of the overall energy score of the category indicators.

The air blower was more relevant in 5 of the 7 categories analyzed, with heating penalized only for GWP (83%) and FRS (86%). This is due to the LCI results and the midpoint LCIA characterization factors associated with the ReCiPe method. For GWP and FRS, the characterization factors for heat and electricity were similar and, therefore, the results were determined by the heating energy demand (almost 90% of the total consumed).

However, the Ecoinvent database reported a much more polluting effect for the electricity production process than heat production in the remaining categories. Because of that, the categories of IR, FE, FET, MET and HCT were penalized for electricity use. Therefore, mixing with air inside the reactor vessel is a major problem and thus a parameter worth controlling. The estimated energy for agitation was dependent only on the reactor volume, while the oxygen demand was related to the organic contaminant removal. Therefore, the electricity demand during aeration was dependent on the limiting activity (agitation or oxygen supply). For the proposed scenario, the energy consumption in the blower was governed by agitation, with the required power being much higher (96% of electricity use) than that for oxygen supply.

Although not as important as energy, the influence of chemicals stood out in the toxicity categories of FET (7.2%), MET (18.8%) and HCT (18.5%). With the exception of ammonium sulphate, dipotassium phosphate, potassium diphosphate and sodium hydroxide, chemicals had a minor contribution since their relative share in the profile was less than 1%. Among these four, the greatest impact comes from ammonium sulphate in the medium category of MET (11.5% of the total score). In terms of direct emissions, CO<sub>2</sub> and 2,4-DCP were emitted to the atmosphere and aquatic environments, respectively.

Although assumed to be fossil emissions, CO<sub>2</sub> from the degradation of the 2,4-DCP was much less of a threat to the environment than the consumption of utilities such as energy or chemicals. For example, GWP was only 0.33% affected by direct CO<sub>2</sub> emissions, while the other 97.4% and 1.5% were related to the other two groups.

In contrast to these, the emission of 2,4-DCP to water became a hazard of considerable magnitude. This was particularly true in the case of FET, since almost 70% of the profile was associated with the emission of non-degraded pollutants. Although with a removal efficiency of 98.7% of the target pollutant, the C-TPPB should improve its kinetic performance and energy demand in the future to improve its friendliness.

One example of a strategy could be the enhancement of the agitation/oxygen supply devices by using mechanical mixers specifically designed for the bioreactor, instead of blowers, or by considering configurations other than the coiled spiral for the tubing, such

as parallel vertical modules. However, the research conducted so far has been able to result in a technology capable of reducing the impact on FET by 98.2% (from 16.7 to 0.43 kg 1,4-DCB).

### 3.1.2. Comparison of impacts between scenarios

The analysis of the LCI inventory in [Table 2](#) indicated that the amount of chemicals consumed increased (1.02 times) only with the rise of the concentration of 2,4-DCP in the influent and remained constant with changes in the recycling ratio. On the other hand, energy demand remained unchanged across scenarios, as heat and electricity are capacity-dependent parameters. The exceptions occurred when effluent recycling was applied. The primary difference in the LCI was attributed to emissions to the environment, as each scenario's measures directly impact disposal efficiency and, consequently, effluent composition. Despite having the lowest chemical consumption, BL exhibited the highest emission of the target phenolic pollutant. In contrast, HR03 showed the lowest emissions among all scenarios. Additionally, sodium hydroxide was the most consumed chemical, primarily used for pH control during operation.

Regarding energy demand, heating and electricity for the blowers were the most critical energy-related factors. Therefore, a continuous 0.5 L/(m<sup>3</sup>·s) was assumed, using therefore 5.15 kWh/m<sup>3</sup> of electricity [50]. Although for this research the presented heuristic was taken as baseline scenario, estimations done considering Metcalf and Eddy [51] and Judd & Judd [57] resulted in an electricity demand of 0.17 kWh/m<sup>3</sup>, which was 96.6% lower.

Estimating the heating energy required for wastewater treatment depends on factors such as ambient temperature, influent temperature (urban or industrial), and reactor configuration (closed/open-air, adiabatic/isothermal). Thermal energy demand can reach 5.8 kWh/m<sup>3</sup> for a 5 °C drop in wastewater temperature [58] and up to 9.3 kWh/m<sup>3</sup> for an 8 °C difference between ambient and operating temperatures. Despite its high energy cost, temperature control is crucial for phenol biodegradation, particularly below 10 °C [59].

Industrial wastewater, often discharged at high temperatures (e.g., 90 °C), could eliminate the need for additional heating if an adapted cooling process is implemented before treatment. Additionally, open-air reactors experience heat dissipation, leading to a heating demand of 41.56 kWh/m<sup>3</sup> ([Table 2](#)) based on a heat loss of 0.5 kWh/m<sup>2</sup> [60]. However, in a fully adiabatic system, energy demand could drop to 5.15 kWh/m<sup>3</sup> or even lower, depending on blower electricity consumption, as estimated by Metcalf & Eddy [51].

Despite the importance that heating and electricity use in the blower may have, the scenarios were only energetically different in the consumption of the recycling pumping. Therefore, when assessing the environmental feasibility with LCA the use of effluent recycling for the baseline concentration (700 mg/L of 2,4-DCP), scenarios BL (without recycling) and BR03 demonstrated how recycling was not a good strategy from a general environmental point of view. However, it was more than satisfactory if the goal was to prevent the toxicological impact on the discharged effluent (from 9.20 g/m<sup>3</sup> to 4.73 g/m<sup>3</sup> of 2,4-DCP in the inventory [Table 2](#) for the respective scenarios). Therefore, and caused by the energy use in the recycling pump, the environmental impact then increased in the range of 0.6%–2.1% (FRS and HCT) except for FET (more related with the effluent toxicity) which is reduced by about 33.2% ([Fig. 2a](#)). However, the intensification of the activities of phenolics-related industries may lead to a higher concentration of flows and thus to a loss of efficiency in treatment. For example, the HR03 scenario was 200 mg/L higher in 2,4-DCP concentration, which was reflected in a decline of 0.29%–4.2% (FRS and MET) of the impact. Despite the fact that the recirculation pump was already operational in the HR03 scenario and electricity was not a differentiating hotspot, the process design of a 0.5 ERR (HR05) did not improve the FET and MET, maintaining similar values by about 0.46% and 0.06% for each category, respectively ([Fig. 2c](#)).

If the scenarios were compared with other phenolic wastewater treatment technologies, the variability of the results was not so significant. The results obtained for the proposed baseline C-TPPB alternatives were between 13.61 and 13.77 kg CO<sub>2</sub>eq./m<sup>3</sup> of treated wastewater (shown in [Table 4](#)). This narrow range may be attributable to the low variability of outcomes across the different scenarios and conditions considered. However, its practical significance depends on the underlying data uncertainty and on whether the observed differences exceed the margin of error of both the assessment method and the measurements. Typically, this issue can be addressed through uncertainty analysis. The challenge arises when secondary data with unknown statistical errors are used, as this increases outcome variability due to reliance on expert judgment and assumptions in subjective methods (e.g., the Pedigree matrix).

In any case the obtained results are in line with those reported by literature in the range between 8.8·10<sup>-3</sup> and 1334 kg CO<sub>2</sub>eq./m<sup>3</sup>

**Table 4**

Absolute comparison of the results of the scenarios for the MidPoint category of climate change and for a damage single score.

Type of impact	BL	BR03	HR03	HR05
<b>Midpoint approach</b>				
Per cubic meter of wastewater treated (kg CO <sub>2</sub> eq./m <sup>3</sup> ) <sup>(1)</sup>	13.609	13.717	13.772	13.772
Per cubic meter of wastewater treated (kg CO <sub>2</sub> eq./m <sup>3</sup> ) <sup>(2)</sup>	13.564	13.672	13.717	13.717
Per cubic meter of wastewater treated (kg CO <sub>2</sub> eq./m <sup>3</sup> ) <sup>(3)</sup>	13.293	13.293	13.303	13.303
Per cubic meter of wastewater treated (kg 1,4-DCB/m <sup>3</sup> ) <sup>(1)</sup>	0.306	0.205	0.211	0.212
<b>EndPoint approach</b>				
Per cubic meter of wastewater treated (Pt/m <sup>3</sup> ) <sup>(1)</sup>	0.211	0.213	0.214	0.214
Per cubic meter of wastewater treated (Pt/m <sup>3</sup> ) <sup>(2)</sup>	0.211	0.212	0.213	0.213
Per cubic meter of wastewater treated (Pt/m <sup>3</sup> ) <sup>(3)</sup>	0.205	0.205	0.205	0.205

Note: BL: Baseline scenario; BR03: 700 mg/L and 0.3 recycling ratio; DCB: Dichlorobenzene; HR03: 900 mg/L and 0.3 recycling ratio; HR05: 900 mg/L and 0.5 recycling ratio. (1) For fossil carbon dioxide emissions, (2) for biogenic carbon dioxide emissions, (3) without considering the chemicals.

(Table 5). For example, Magdy et al. [61] compared five chemical methods (activated carbon, electro-Fenton, solar photo-Fenton, solar photocatalysis by TiO<sub>2</sub>, and photocatalysis by a composite of TiO<sub>2</sub> and activated carbon) for the removal of phenols in wastewater. For a functional unit of 1 m<sup>3</sup> with an initial concentration of 200 mg/L, the GWP varied between 0.88 and 26.77 kg CO<sub>2</sub>eq./m<sup>3</sup> (for photo-Fenton and electro-Fenton respectively). As done by Magdy et al. [61], Ou et al. [62] also performed an LCA for technologies removing phenolic compounds from wastewater. A *Cradle-to-gate* system boundary was also adopted for the wastewater treatment of critical water oxidation and ozonation coupled with coagulation/flocculation and microfiltration. The environmental profile for GWP was in this case much higher, 86 and 1334 kg CO<sub>2</sub>eq./m<sup>3</sup> for each of the previously mentioned methods respectively. In this regard, the C-TPPB seemed to operate better than catalytic wet oxidation processes but worse than Fenton, photocatalytic and biological with the use of alkali resistant strains.

The C-TPPB results seemed to align with the expected outcomes of other advanced technologies. However, the comparability of these results with those from the aforementioned publication faced significant limitations. This is because the research was conducted as an independent operational analysis of a large-scale pilot system, rather than a full-scale facility, which would also encompass the construction phase. Furthermore, the C-TPPB system achieved similar removal efficiencies (98.7% compared to 81–99.9% reported for the other technologies) for a much more polluted stream (700–900 mg/L).

### 3.1.3. Environmental damage single score

The total single damage score of the BL, BR03, HR03 and HR05 scenarios was 208.02 mPt, 209.64 mPt, 210.705 mPt and 210.706 mPt, respectively (Table S9). Therefore, this translated to a 0.77% increase in environmental damage due to the operation of the pump installed for effluent recirculation, 0.52% due to the 200 mg/L increase in influent 2,4-DPC concentration over the baseline scenario, but there was not improvement due to the use of a higher ERR (from 0.3 to 0.5). Despite the higher intrinsic electricity requirements for the recirculation flow at HR05, the achieved removal efficiency is not able to compensate for the damage caused.

Unlike the category analysis described in Section 3.1, the ReCiPe damage score was not represented by effluent emissions to water, as their percentage is less than 0.04% in all scenarios. By far the main hotspot was indirect energy use, at over 95.6%. Given its large contribution to the profile, reductions in blower or heating energy demand translated into proportionate improvements. For example, a 20% decrease in the environmental impact of energy translated into a 19.1% improvement in the overall profile.

### 3.1.4. Towards the optimization of energy use

When comparing the different energy-modified scenarios BL, BL-B, BL-CR, BL-HI and BPS, Fig. 3 was obtained. The worst outcomes were achieved by BL-HI with a relative difference with BL of 1.2%–22% (FET and FRS) among categories. Among the other two proposals or alternatives of improvement (BL-B and BL-CR), the best option was BL-B. This is because the reduction strategy suggested for the electricity demand in the blowers endorsed larger enhancements in the profile (18%–83%) of all the categories under study except GWP and FRS, whose improvements (87%) were more related to the minimization of the heating energy use. The simultaneous implementation of both measures resulted in an overall midpoint profile improvement between 22% and 98%. Therefore, and when comparing the contribution profiles of BL with BPS, the energy was no longer represented between 23% (FET) and 98% (FRS) but instead was reduced up to 6.2%–50%.

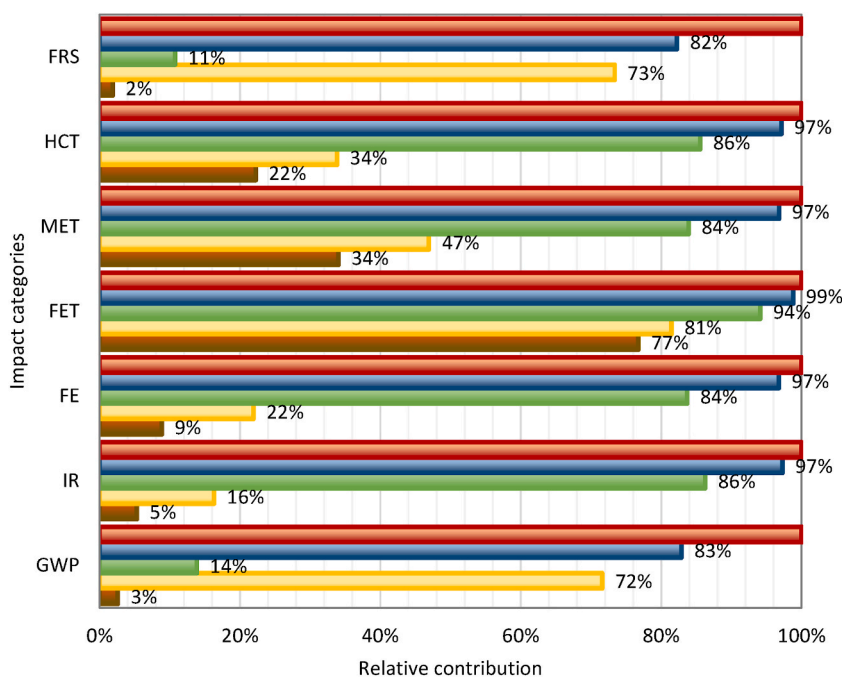
Because of this, energy was no longer the hotspot of the process. Under these circumstances, a next stage in design should be to optimize the system with a subsequent decrease in the use of the chemicals. Only in terms of GWP, the carbon emissions could be between 0.43 kg CO<sub>2</sub>eq./m<sup>3</sup> (BPS) and 16.42 kg CO<sub>2</sub>eq./m<sup>3</sup> (BL-HI). Therefore, and if compared again with the literature outcomes of Table 5, the C-TPPB may be competitive with all options except with the adsorption process proposed by Frascari et al. [63]. If that was not all, technology showed also results that are in line with the outcomes expected for full-scale domestic wastewater treatment. Some

**Table 5**

Global warming results of different literature technologies for the treatment of wastewaters with phenolic compounds.

Technology	Reference	Carbon footprint (kg CO <sub>2</sub> eq./m <sup>3</sup> )
Coagulation with CWO	[62]	85.5
Microfiltration with CWO	[62]	86.1
Coagulation with ozonation	[62]	1334
Microfiltration with ozonation	[62]	1334
Adsorption	[61]	5.12
Adsorption	[63]	8.8·10 <sup>-3</sup>
Electro-Fenton	[61]	26.78
Photo-Fenton	[61]	0.88
Photocatalysis	[61]	4.08
Photocatalysis	[64]	6.86
Photocatalysis with activated carbon	[61]	5.65
Photocatalysis with UV-LED/TiO <sub>2</sub>	[65]	5.92
Photocatalysis with UV-BL/TiO <sub>2</sub>	[65]	8.8
<i>Bacillus halotolerans</i> ACY treatments <sup>(1)</sup>	[66]	6.5
PMB600 treatments <sup>(1)</sup>	[66]	4.2
BIPB treatments <sup>(1)</sup>	[66]	1.9

Note: BIPB: Immobilized pig manure biochar; BL: Black Lightning; CWO: Catalytic Wet Oxidation; LED: Light Emitting Diode; PMB: Pig Manure Biochar; UV: Ultraviolet. (1) Alkali-resistant and thermophilic strains.



**Fig. 3.** Relative impact (MidPoint) environmental profile comparison analysis between BL-HI (■), BL (■), BL-CR (■), BL-B (■) and BPS (■). BL-HI: Baseline scenario with open reactor and heated influent; BL: Baseline scenario; BL-CR: Baseline scenario with a closed-reactor; BL-B: Baseline scenario with changes in the blower energy; BPS: Best performing scenario; FE: Freshwater Eutrophication; FET: Freshwater Ecotoxicity; FRS: Fossil Resource Scarcity; GWP: Global Warming Potential; HCT: Human carcinogenic toxicity; MET: Marine Ecotoxicity.

examples could be the research of Besson et al. (2021) (0.9 kg CO<sub>2</sub>eq./m<sup>3</sup>), Bisinella de Faria et al. [67] (1.05 kg CO<sub>2</sub>eq./m<sup>3</sup>), Gupta et al. [68] (0.51–1.14 kg CO<sub>2</sub>eq./m<sup>3</sup>), Sun et al. [69] (0.76–1.09 kg CO<sub>2</sub>eq./m<sup>3</sup>) and Chen et al. [70] (0.65 kg CO<sub>2</sub>eq./m<sup>3</sup>).

In line with the midpoint outcomes, the single score assessment resulted in an overall reduction of the environmental damage of 97% with relative decreases between the different alternatives as high as 84% and as low as 17%.

### 3.1.5. Change of LCIA method for impact assessment

Both ReCiPe and USEtox methods showed consistent results since freshwater ecotoxicity was predominantly affected by the release of 2,4-DCP to the aquatic environments. Also in line, the main hotspot of the human carcinogenic toxicity was energy consumption. However, and since the characterization factors differ from method to method, the importance of direct emissions changed between them. While the expected proportion of emissions to the environment accounted for 69.4% in FET for BL and ReCiPe (shown in

**Table 6**

Absolute outcomes and relative contribution of the ecotoxicity category for the ReCiPe and Usetox methods.

Method	Scenarios	Unit	Energy	Chemicals	Emissions
USEtox	BL	CTUe	1.50·10 <sup>-2</sup>	1.26·10 <sup>-3</sup>	82.05
		%	0.02	0.00	99.98
	BR03	CTUe	1.50·10 <sup>-2</sup>	2.22·10 <sup>-3</sup>	42.15
		%	0.04	0.01	99.96
	HR03	CTUe	1.50·10 <sup>-2</sup>	2.27·10 <sup>-3</sup>	42.91
		%	0.03	0.01	99.97
HR05	CTUe	1.50·10 <sup>-2</sup>	2.27·10 <sup>-3</sup>	43.28	
	%	0.03	0.01	99.97	
ReCiPe	BL	kg 1,4-DCB	7.04·10 <sup>-2</sup>	2.35·10 <sup>-2</sup>	0.21
		%	22.97	7.68	69.36
	BR03	kg 1,4-DCB	7.00·10 <sup>-2</sup>	2.50·10 <sup>-2</sup>	0.11
		%	34.37	12.29	53.33
	HR03	kg 1,4-DCB	7.00·10 <sup>-2</sup>	3.00·10 <sup>-2</sup>	0.11
		%	33.28	14.15	52.56
	HR05	kg 1,4-DCB	7.00·10 <sup>-2</sup>	3.00·10 <sup>-2</sup>	0.11
		%	33.13	14.09	52.78

Note: BL: Baseline scenario; BR03: 700 mg/L and 0.3 recycling ratio; HR03: 900 mg/L and 0.3 recycling ratio; HR05: 900 mg/L and 0.5 recycling ratio; CTUe: Comparative toxic units for ecotoxicity, 1,4-DCB: 1,4-Dichlorobenzene.

Table 6), the 2,4-DCP found in the effluent was almost the only parameter of concern in USEtox (99.98%). This was due not only to the 2,4-DCP characterization factor, but also to the lower energy input caused by the impact of the different substances involved in this upstream process. For example, the use of electricity in the blower and pumps evaluated with ReCiPe would be mainly defined by the emissions to water of zinc, nickel and copper among others. USEtox, however, considered terbufos, diflubenzuron and captan as its three most important substances. Regarding human toxicity, the relevance of chemicals decreased from 7.7% in ReCiPe to <0.01% in USEtox. According to the above described, chromium VI, nickel and arsenic are the hotspots for electricity in ReCiPe, while formaldehyde, 2,3,7,8-tetrachlorodibenzo-p-dioxin and chloroethene, for USEtox.

Analyzing not only the energy but the entire process emissions, the results were still congruent when it comes to the diversity of the most relevant indirect substance emissions to the environment in ecotoxicity. Zinc, Nickel, copper and silver were important for ReCiPe and phenol, formaldehyde, terbufos and chlorpyrifos, for USEtox. It is worth mentioning that the substances highlighted for ReCiPe have no characterization factors for USEtox. The biggest characterization factors in both methods were associated with the indirect emissions of terbufos and chlorpyrifos. In the third position for USEtox can be found the DCP, which differs from ReCiPe since it is a substance occupying the sixth position on a ranking based on the largest characterization factor.

### 3.2. Costs of the process

Table 7 presents the values for each type of cost (investment, working capital, variable and fixed costs, and replacement of obsolete equipment), expressed in €/m<sup>3</sup> of treated wastewater. According to these results, operational costs accounted for approximately 97% of the total investment required for the operation of the technological treatment scenarios, with only 0.05% attributed to variable costs. When fixed costs—estimated based on literature—were excluded from the total breakdown, operational costs represented merely 2%. Consequently, the initial investment became the most significant component (68% excluding fixed costs), while fixed costs were 420 times higher than operational costs. Among the initial investment expenditures (Table S14), the reactor tank was the most expensive item, accounting for around half of the budget in the BL scenario (50.6%), followed by the storage tanks (23.5% each for BL). Pumps and blowers constituted the remaining 2.4%, despite being duplicated to ensure continuous operation in the event of mechanical failures. The addition of a pump to enable partial recycling of effluent wastewater to the reactor (in scenarios BR03, HR03, and HR05) increased the relative importance of these components by only 0.5%.

Concerning operational costs (Table S15), general plant overhead was the largest contributing factor, followed by the hiring of personnel for the operation of the facility. It is worth noting that both of these were fixed costs, which aligns with the aggregated results shown in Table 7. Regarding variable costs, the significance of energy consumption (92.6%) surpassed that of chemicals and was followed by the LCA environmental costs (6.2%). Among energy expenses, reactor heating accounted for 67% of the variable costs, which contrasts with the outcomes reported by the LCA. Since the LCA and LCC share similar inventories, it would logically follow that their findings are related or closely aligned. However, the characterization factors and market prices also play a significant role in these differences.

Regarding chemicals, the use of ammonium sulphate (0.5% of variable costs), dipotassium phosphate (0.3%), and sodium hydroxide stands out. Their greater impact compared to other chemicals (0.4%) was due to their higher consumption during the process operation. Despite having the highest market price (3.02 €/kg), ethylenediaminetetraacetic acid disodium salt has the second lowest price.

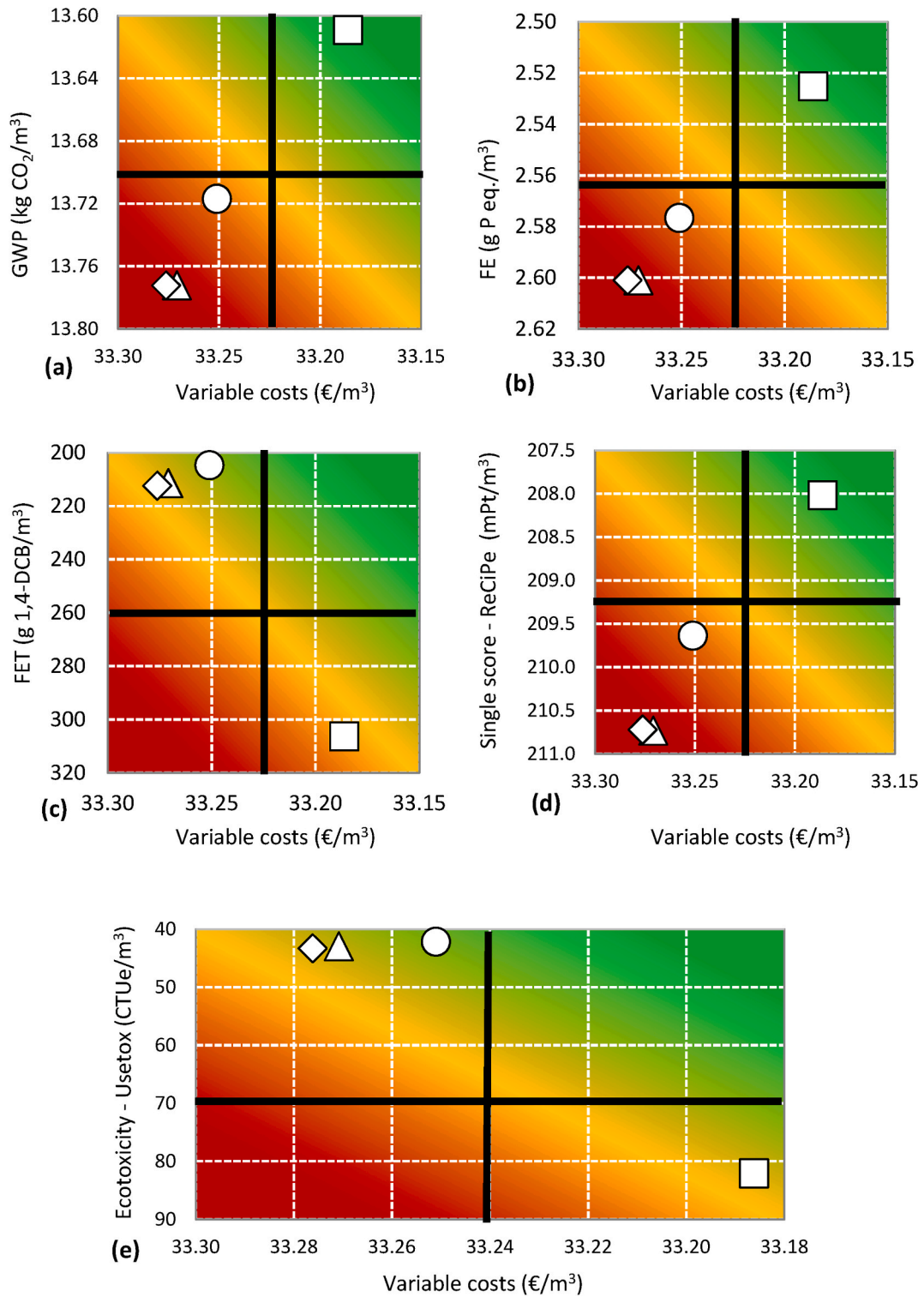
Until now, state-of-the-art cost accounting has primarily focused on the analysis of large-scale domestic wastewater treatment, with sizes ranging from 10 to 1000 times larger than those of the four selected scenarios of this research. For example, Vuori and Ollikainen [71] indicated that the total costs of the conventional activated sludge (CAS) could be around 1.71 € and 0.33 € per treated cubic meter depending on the treatment plant size (from 10,000 to 500,000 population equivalent) and without considering the sludge treatment. Also, according to their results, the use of a membrane bioreactor (MBR) could have costs around 0.08–0.27 €/m<sup>3</sup>. Combinations of the MBR with a high rate activated sludge (HRAS) systems have reported to be around 0.195 €/m<sup>3</sup> for a 550,000 m<sup>3</sup>/d treatment. However, other systems with lower land and electricity requirements have proven to perform better. This is the case of the anaerobic-anoxic-oxic (A2O) and aerobic granular sludge (AGS) processes, with costs of 0.113 €/m<sup>3</sup> and 0.091 €/m<sup>3</sup>, respectively [72].

**Table 7**

Specific costs breakdown, expressed in euros, for 30 years of operation and per functional unit or 1 m<sup>3</sup> of wastewater treated.

Variable	BL	BR03	HR03	HR05
Initial investment in time zero	257.18	258.43	260.61	260.95
Initial working capital	78.44	78.44	78.45	78.45
Variable costs	7.62	7.68	7.70	7.71
Fixed costs	13,924.78	13,925.70	13,927.32	13,927.57
Variations in working capital	25.57	25.57	25.57	25.57
Acquisition of assets in time	5.50	4.76	6.70	6.86
TAEC - Total	14,299.08	14,300.59	14,306.36	14,307.11
TAEC - Only investment	341.12	341.63	345.77	346.26
TAEC - Operational costs	13,957.97	13,958.95	13,960.59	13,960.85
TAEC - Variable costs	33.19	33.25	33.27	33.28

Note: BL: Baseline scenario; BR03: 700 mg/L and 0.3 recycling ratio; HR03: 900 mg/L and 0.3 recycling ratio; HR05: 900 mg/L and 0.5 recycling ratio; TAEC: Total Annual Equivalent Cost.



**Fig. 4.** Representation of eco-efficiency analysis results, considering environmental impacts and economic values (variable costs) for Global Warming Potential (a), Freshwater Eutrophication (b), Freshwater Ecotoxicity (c), damage single score with ReCiPe (d) and ecotoxicity damage score for the USEtox method (e). Symbols: square corresponds to the BL (Baseline) scenario, circle for the BR03 (700 mg/L and 0.3 recycling ratio), triangle is the HR03 (900 mg/L and 0.3 recycling ratio) and diamond is the HR05 (900 mg/L and 0.5 recycling ratio) scenario. GWP: Global Warming Potential; FE: Freshwater Eutrophication; FET: Freshwater Ecotoxicity.

However, the competitiveness of A2O with novel processes such the MBR is still under discussion. In contrast with the results of Cicekalan et al. [72], the LCC of Cankaya and Pekey [73] reported higher values when analyzing 10 WWTPs between capacities of around 3500 - 98,000 m<sup>3</sup>/d and A2O, anoxic-oxic (AO) and CAS processes. The results achieved varied between 0.21 and 0.53 €/m<sup>3</sup>. Even lower results were found in the literature, but in this case were more related to sections or specific technologies rather than complete WWTP. This is the case of preliminary and tertiary treatments, whose costs were reported to be around 0.024–0.131 €/m<sup>3</sup> [74].

Apparently for domestic large-scale wastewater treatment the costs rarely surpass 1 €/m<sup>3</sup>. However, industrial wastewater treatment has been providing controversial results depending on the type of wastewater, technologies and scales.

Çapa et al. [75,76] have calculated an overall cost for an industrial WWTP of 4000 m<sup>3</sup>/d of 0.36–0.46 €/m<sup>3</sup>. Higher costs were reported for brine effluents from a chemical industry producing silica derivatives (from 2.6 to 24.9 €/m<sup>3</sup>) and 1327 m<sup>3</sup>/d of capacity [76]. Even higher values were observed for a 24 m<sup>3</sup>/d facility treating olive mill wastewater in continuous mode. The results achieved were as high as 253 and 292 €/m<sup>3</sup> [77]. Despite the variability of results among types of wastewaters, processes for industrial treatment are reporting bigger costs than domestic, probably because the technologies used need to deal with pollutants that are often recalcitrant and difficult to remove. On the other hand, it should be noted that the outcomes of the cited studies were provided for facilities of different scales, whose changed was analyzed by Dogot et al. [78] for the annual operation and maintenance costs of domestic wastewater treatment. Finally, it should be noted that each study has different considerations when it comes to the fixed cost to be included under analysis, giving to a large variability of the possible results to be obtained. For example, for this study the economic costs may vary between 7.62 €/m<sup>3</sup> and 14,299 €/m<sup>3</sup> depending if only variable costs or all costs (based on the baseline assumptions taken for the research were considered).

### 3.3. Outcomes of the baseline scenarios of eco-efficiency

The eco-efficiency profile of the BL, BR03, HR03, and HR05 scenarios is shown in Fig. 4. The product value and variable costs are consistent across all graphical representations (Fig. 4a–e), while the environmental indicators, expressed in terms of midpoint impacts and endpoint damages, differ. When comparing the scenarios, four possible eco-efficiency tendencies emerged: the top-right quadrant represents lower costs and environmental impacts, the bottom-left quadrant indicates the highest cost-impact correlation, and the other two quadrants reflect combinations of both low-high costs and environmental impacts.

Using BL (square icon) as a reference, Fig. 4a, b and d demonstrate that the strategies used in this scenario resulted in the lowest variable costs and environmental impacts/damages in the GWP and FE impact categories. The exception was the FET indicator on which, for the same variable costs shown in the other categories, the environmental impact was the largest among all scenario alternatives. This was caused (as shown in Table 2) by the 2,4-DCP concentration for the BL scenario. The introduction of recycling pumping into the system (BR03) lead to a direct increase in costs and environmental impacts across all categories, except for FET. However, this relationship was not entirely straightforward: while costs rose proportionally, the environmental impact followed an inverse trend. In this sense, the facility successfully reduced effluent pollution, but at the expense of an unfavorable economic outcome.

An increase in influent concentration negatively affected both sustainability pillars—economic and environmental. It is important to recognize that impact was not independent of concentration. This is particularly relevant when comparing a technological solution with state-of-the-art processes. Once recycling pumping was implemented for highly concentrated streams, further adjustments to the recycling ratio resulted in negligible changes to eco-efficiency.

The single-score analysis of the eco-efficiency graphs (Fig. 4d and e) presented ongoing contradictions. The ReCiPe method favored technologies with the lowest energy demand, regardless of the primary goal of wastewater treatment—removing toxic contaminants. In contrast, the USEtox method focused solely on ecotoxicity, yielding results that align with the ReCiPe FET indicator. These findings suggested that optimizing the technology at the laboratory scale to improve 2,4-DCP removal efficiency was effective, enhancing both eco-efficiency and effluent toxicity-related indicators.

While a qualitative analysis helped to identify eco-efficiency trends among the alternatives studied, the quantitative analysis—using indicators that divide environmental impact by costs—was less straightforward. A lower indicator value did not necessarily mean better system performance, as the lowest values occurred when environmental impact was minimized but costs were highest. Achieving an optimal system requires balancing both aspects.

Therefore, selecting the most eco-efficient scenario required normalizing environmental and economic results to minimize both impacts and costs. Under this approach, the BL scenario was the most eco-efficient for FE and GWP, while BR03 performed best in the FET category. For highly concentrated streams, the lowest recycling ratio offered the best performance, though the difference was minimal (approximately 0.02%).

Apart from technical limitations in the comparison of treatment processes caused by a different influent concentration, there were other numerous methodological limitations in comparing the eco-efficiency results of the C-TPPB technology with outcomes from other studies. These limitations included differences in system boundaries, product value (e.g., cost with or without the inclusion of construction expenses), environmental impact categories, and technologies. Additionally, this study did not analyze large-scale processes, introducing the impact of economies of scale on the eco-efficiency, which influences both economic and environmental indicators. An effort was made to correlate the findings of this research with others by considering global GWP and the ReCiPe single score as environmental impact categories.

As shown in Table S17, the eco-efficiency for GWP was approximately 0.98 g CO<sub>2</sub>/€ for operational costs, 413 g CO<sub>2</sub>/€ for variable costs. Although these results differed significantly from those reported by Lorenzo-Toja et al. [79] which range from 1063 to 2163 g CO<sub>2</sub>/€ for operational costs, the discrepancy may arise from differing assumptions regarding cost components. Notably, Lorenzo-Toja

et al. [79] included personnel, fees, maintenance, and lab analysis, whereas this study also accounted for land rent, salaries, plant overhead and insurance costs.

For the ReCiPe single score, the eco-efficiency was approximately 0.015 mPt/€ for operational costs and 6.3 mPt/€ for variable costs. These values were again lower than the average of 119 mPt/€ reported by Lorenzo-Toja et al. [79] for similar reasons. Eco-efficiency result for the single score appear to vary widely across other publications, ranging from below 1.16 mPt/€ to nearly 1160 mPt/€. For example, Petit-Boix et al. (2018) reported a value of 140 mPt/€ for the Calafell WWTP, which aligned with the findings of Lorenzo-Toja et al. [76], despite it included construction costs in the eco-efficiency analysis. In contrast, Alizadeh et al. [80] conducted a study on a large-scale WWTP (Al-Teymour plant), reporting an eco-efficiency value of 0.0172 mPt/€ total costs (including chemical, energy, personnel, and maintenance costs). The highest values were reported by Abdalla et al. [30], who estimated eco-efficiency impacts ranging from 298 mPt/€ to 1122 mPt/€ for conventional and on-site treatments, respectively. One key difference between this publication and the previously mentioned is the inclusion of the environmental impacts associated with constructing both the sewer network and the wastewater treatment plant.

### 3.4. Sensitive parameters in eco-efficiency

The eco-efficiency results may be biased by the independent outcomes of the LCA and LCC considering both technical and methodological assumptions. As mentioned in Section 3.2, a reduction in environmental impact lead to a decrease in the indicator value. In this context, and considering only the variable operation of the facility, energy optimization could reduce the indicator by 96%. In absolute terms, eco-efficiency improved from 410 to 16.2 g CO<sub>2</sub>/€. When fixed costs were included in the analysis, the result is 0.98 g CO<sub>2</sub>/€. However, reducing labor costs by hiring an operator for 2 h per week instead of 2 h per day would raise the indicator to 8.9 g CO<sub>2</sub>/€. Including only construction costs, with a corresponding 5–23% impact on the LCA, the indicator would range from 0.99 to 1.2 g CO<sub>2</sub>/€ of investment. Implementing all three of these economic measures would increase the value to 9.3 g CO<sub>2</sub>/€. If the BPS scenario was considered instead of the BL, the eco-efficiency indicator would be the lowest, at 0.29 g CO<sub>2</sub>/€. Therefore, the outcomes of this research felt within a range of 0.29–413 g CO<sub>2</sub>/€.

### 3.5. The EU taxonomy

A total of 39 activities within the EU taxonomy were identified related to the water, wastewater, and waste sectors (code NACE E) across the six objectives. Among these, only 7 were found to be applicable or require amendment through the implementation of a C-TPPB technology (as shown in Tables 8 and 9) to address the objectives of climate change mitigation and adaptation, sustainable use and protection of marine resources, and pollution prevention and control. Since the technology did not produce reclaimed water or products that could be marketed, the objective of circular economy was not relevant. Similarly, the restoration of habitats and species was outside the scope of this study. To date, the study has addressed energy efficiency technical criteria, climate change projections and assessments, compliance with discharge requirements, and the presence of organic resistant pollutants. Detailed explanations of each element, determining whether the technology aligned with the EU Taxonomy, are provided in Tables 8 and 9

However, the EU Taxonomy did not provide specific thresholds or limitations for the criteria that can be applied to industrial wastewater treatment technologies. In addition to the development of guidelines to support compliance, actionable steps could be classified differently depending on whether the wastewater technology was part of a larger company infrastructure and based on its technology readiness level. For instance, EU Taxonomy criteria related to the location of the facility (e.g., sewer system and geographical climate factors) may not be relevant for pilot or decentralized testing.

Regardless of whether the technology was integrated into a larger EU-Taxonomy-aligned project or process scale, its primary goal was to align with the EU Taxonomy's key principles: continuous minimization and renewability of energy consumption, reduction of the carbon footprint, and the decrease of organic and persistent pollutants that contribute to eutrophication and toxicity. These factors were analyzed through a two-pillar sustainability approach using eco-efficiency, LCA, and LCC methodologies. The results and conclusions drawn from the previous sections contribute to the development of a technology that is aligned with the EU Taxonomy and can be improved over time with measures such as: the use of adiabatic reactors, integration of the manufacturing process with wastewater treatment to improve energy efficiency, implementation of an optimized aeration system, and the adoption of a low-energy recycling system to enhance pollutant removal efficiency.

## 4. Conclusions

Four scenarios were proposed for the optimization of a continuous two-phase bioreactor for pilot-scale treatment of phenolic, aqueous solution with 2,4-dichlorophenol, industrial wastewater. In these scenarios, effluent recirculation and influent concentration were modified in order to analyze how technical decisions influence integrally environmental and economic outcomes.

Regardless of the concentration, scenarios with an effluent recycling flow into the reactor showed the highest environmental impact across all categories, except for those related to ecotoxicity (with a 98.2% reduction). As intended, the C-TPPB performed better with the inclusion of recycling, leading to improved effluent quality. However, the associated utility consumption required for proper reactor operation diminished the overall benefits. Increasing concentration amplified the environmental impact, which cannot be offset by raising the recycling ratio.

When it comes to the operational costs, the differences between the scenarios were minimal due to the dominance of fixed costs, which remained the same across all cases. Finally, the technology's performance aligned with others reported in the literature

**Table 8**  
Selection of substantial contribution EU Taxonomy technical screening criteria for wastewater treatment for the objective of climate change mitigation.

EU Taxonomy objectives	Name of the eligible activity	Summarized goal of the criteria	Technical screening criteria (Substantial contribution)	Explanation of the technical criteria for the technology	Aligned activity
Climate change mitigation	Construction, extension and operation of wastewater collection and treatment	Energy demand of the wastewater treatment system	The net energy consumption of the wastewater treatment plant equals to or is lower than 35 kWh per p.e. per annum for treatment plant capacity below 10,000 p.e.	While not a complete wastewater treatment plant, the technology consumes 4.49–5.54 kWh/p.e., significantly less than the 35 kWh/p.e. expected for facilities serving 10,000 people. However, this estimate does not account for integration with centralized treatment processes.	Y
Climate change mitigation	Renewal of wastewater collection	Energy efficiency in wastewater collection	The renewal of a collection system improves its energy efficiency with a reduction of the energy consumption by 20% compared to the baseline performance averaged over three years	The sewer or wastewater collection has not been considered. Besides, this criterion is transitory and cannot be evaluated for the technology provided.	NA
Climate change mitigation	Renewal of wastewater treatment	Energy efficiency in wastewater treatment	The net energy consumption of the system is annually calculated to demonstrate a reduction in energy demand. However, this improvement should only occur if the operator demonstrates that there are no material changes or modifications to discharge.	Energy consumption has been estimated for each scenario: 41.74 kWh/m <sup>3</sup> for BL, 41.70 kWh/m <sup>3</sup> for BR03, 41.99 kWh/m <sup>3</sup> for HR03 and 42.04 kWh/m <sup>3</sup> for HR05. The results show that changes in influent load led to energy consumption variations below 1%. To avoid effluent modification with influent loads, a recycling system was implemented.	Y

Note: BL: Baseline scenario; BR03: 700 mg/L and 0.3 recycling ratio; HR03: 900 mg/L and 0.3 recycling ratio; HR05: 900 mg/L and 0.5 recycling ratio; NA: Not available; N: No; p.e.: Population Equivalent and Y: Yes.

**Table 9**

Selection of substantial contribution EU Taxonomy technical screening criteria for wastewater treatment for the objectives of climate change adaptation, protection of water and marine resources and pollution prevention and control.

EU Taxonomy objectives	Name of the eligible activity	Summarized goal of the criteria	Technical screening criteria (Substantial contribution)	Explanation of the technical criteria for the technology	Aligned activity
Climate change adaptation	Construction, extension and operation of wastewater collection and treatment	Reduction of the physical-climate impacts	The economic activity has implemented physical and non-physical solutions that reduce the most important physical climate risks that are material to the wastewater sector.	Climate risks for wastewater treatment plants, as outlined in Annex A of EU 2021/2139, include sea level rise, saline intrusions, heavy precipitation, floods, soil degradation, and erosion. The technology meets the criterion as it is not impacted by any of these climate risks.	Y
Climate change adaptation	Construction, extension and operation of wastewater collection and treatment	Performance of a Life Cycle Assessment	The climate projections and assessment of impacts are based on best practice and available guidance	An impact assessment was conducted using the ISO 14040 and ISO 14044 standards in the Life Cycle Assessment. The global warming potential for the analyzed technology scenarios ranges from 13.61 to 13.77 kg CO <sub>2eq</sub> /m <sup>3</sup> .	Y
Sustainable use and protection of water and marine resources	Urban wastewater treatment	Fulfillment of discharge requirements of the wastewater treatment system	The wastewater treatment system does not result in a deterioration of the good status of water bodies. The wastewater treatment system fulfils the discharge requirements set up by the competent local authorities.	The Council Directive 91/271/EEC (May 21, 1991) set a maximum COD effluent concentration of 125 mg/L and a 75% reduction target. The technology being analyzed has achieved over 98.73% organic matter removal, with a maximum COD concentration of 11.73 mg/L (BL scenario). Surface water body characterization is not possible yet due to the lack of a specific implementation location.	Y
Pollution prevention and control	Treatment of hazardous waste	Presence of persistent organic pollutants	All waste containing POP substances listed in Annex IV to Regulation (EU) 2019/1021 are controlled and traced as hazardous waste.	2,4-Dichlorophenol is a persistent organic pollutant. However, it has not been already included in the Annex IV of the Regulation (EU) 2019/1021	N

Note: BL: Baseline Scenario; COD: Chemical Oxygen Demand; ISO: International Organization for Standardization; POP: Persistent Organic Pollutants.

(13.61–13.77 kg CO<sub>2eq</sub>/m<sup>3</sup> of treated wastewater and costs ranging from 7.62 €/m<sup>3</sup> to 14,299 €/m<sup>3</sup>, depending on whether only variable costs or all costs were considered). To enhance the competitiveness of technology, it is essential to manage energy consumption for heating and aeration, as well as to increase process automation, which would help reduce labor-related costs.

### CRedit authorship contribution statement

**Sofía Estévez:** Writing – original draft, Visualization, Methodology, Investigation, Formal analysis, Conceptualization. **Domenica Mosca Angelucci:** Writing – review & editing, Supervision, Investigation, Conceptualization. **María Teresa Moreira:** Writing – review & editing, Validation, Supervision. **María Concetta Tomei:** Writing – review & editing, Validation, Supervision, Conceptualization.

### Declaration of competing interest

The authors declare the following financial interests/personal relationships which may be considered as potential competing interests: Sofia Estevez Rivadulla reports financial support was provided by Spanish Ministry of Science, Innovation and Universities. Sofia Estevez Rivadulla reports financial support was provided by State Agency of Research. If there are other authors, they declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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12).

## Abbreviations

AGS	Aerobic Granular Sludge
A2O	Aerobic Anoxic Oxidation
CBA	Cost-Benefit Analysis
BIBP	Immobilized Pig Manure Biochar
BL	Black Lightning
CAS	Conventional Activated Sludge
COD	Chemical Oxygen Demand
CTUe	Comparative toxic units for ecotoxicity
CVM	Contingent Valuation Method
C-TPPB	Continuous Two-Phase Partitioning Bioreactor
CWO	Catalytic Wet Oxidation
1,4-DCB	1,4-Dichlorobenzene
2,4-DCP	2,4-Dichlorophenol
DEA	Data Envelop Analysis
EEA	Eco-efficiency Analysis
FE	Freshwater Eutrophication
FET	Freshwater Ecotoxicity
FRS	Fossil Resource Scarcity
GWP	Global Warming Potential
HCT	Human Carcinogenic Toxicity
HRAS	High Rate Activated Sludge
ILCD	International Reference Life Cycle Data System
IR	Ionizing Radiation
ISO	International Organization for Standardization
LCA	Life Cycle Assessment
LCC	Life Cycle Costing
LCIA	Life Cycle Impact Assessment
LCI	Life Cycle Inventory
LED	Light Emitting Diode
MBR	Membrane Biorreactor
MET	Marine Ecotoxicity
N	No
NA	Not Available
NaEDTA	Ethylenedinitrilotetraacetic acid disodium salt
p.e.	Population Equivalent
PMB	Pig Manure Biochar
POP	Persistent Organic Pollutants
SEEA	System Environmental-Economic Accounting
TAEC	Total Annual Equivalent Cost
TEA	Techno-Economic Assessment
TRL	Technology Readiness Level
UV	Ultraviolet
VSS	Volatile Suspended Solids
Y	Yes

## Appendix A. Supplementary data

Supplementary data to this article can be found online at <https://doi.org/10.1016/j.wri.2026.100346>.

## Data availability

Data will be made available on request.

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