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Detritus - Multidisciplinary Journal for Waste Resources and Residues - is aimed at extending the “waste” concept by opening up the field to other waste-related disciplines (e.g. earth science, applied microbiology, environmental science, architecture, art, law, etc.) welcoming strategic, review and opinion papers. **Detritus is indexed in Emerging Sources Citation Index (ESCI) Web of Science, Scopus, Elsevier and Google Scholar.** Detritus is an official journal of IRACE / International Research Association on Circular Economy (www.irace.eco) and IWWG / International Waste Working Group (www.iwwg.eu).

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Editorial

WASTE MANAGEMENT IS PIVOTAL IN THE CROSSROAD BETWEEN FOOD AND HEALTH, LEGAL, ECONOMIC, FINANCIAL AND ETHICAL INTERESTS ARE MODULATORY

Introduction

Ensuring access to food is one of the main pillars of the global sustainability goals. The challenge of feeding an expanding global population is complicated by the dual demands for both quantity and quality. Consumer expectations are for clean, spotless, uniform, and herbicide-free products. This has led to widespread reliance on agrochemical interventions to provide quantity and desired quality.

This editorial aims to explore the paradox in contemporary food systems: the use of chemicals, particularly herbicides, to satisfy consumer demands on one hand, and the potential consequences for human health on the other. The core message is that waste management plays a crucial role in resolving this paradox. Legal issues, economics/finance, and ethics influence the presence of chemicals in food, but do not determine it. Glyphosate will be used as a case study because of its significant role in modern agriculture and its controversial toxicological and regulatory history.

Glyphosate as a Model Chemical

Since its introduction in the 1970s and the subsequent release of glyphosate-resistant genetically modified (GM) soybean crops in 1996, glyphosate has become the most widely used herbicide worldwide, best known commercially as Roundup. The main advantage of glyphosate over other herbicides is its selective biological target in plants: an enzyme called EPSPS. This enzyme is absent in human cells, which explains glyphosate's low acute toxicity and accounts for its widespread use worldwide.

However, there is a consensus that glyphosate shows toxic properties, and its potential to cause cancer is the most debated issue. Data from *in vivo* studies support comparisons of this carcinogenic potential with levels found in processed meat, diesel exhaust, and nitroso compounds. The available human epidemiological studies complicate this issue because of the lack of individual exposure assessments, thereby limiting causal conclusions. This methodological gap is often cited by glyphosate supporters to dismiss concerns, despite the increasing accumulation of toxicological evidence and public calls for stricter regulation.

Human exposure to glyphosate is widespread, and numerous studies have detected glyphosate in the urine of

up to 90% of tested individuals across various populations. Although these concentrations are below toxic thresholds, available data on glyphosate levels in biological fluids (blood and urine) suggest they are within the same concentration range as endogenous signalling molecules (hormones, neurotransmitters...), raising a valid concern and warranting closer scrutiny.

The toxicological profile of glyphosate has expanded over the past decade, revealing its interactions with microorganisms in the mammalian gut and in insects, amphibians, and other organisms. However, the precise scientific details are still under investigation. Potential effects on neurological and endocrine systems have been identified. I have recently outlined the current toxicological profile of glyphosate in some detail (Fraeyman, 2025). While suggestive evidence exists, a conclusive mechanistic understanding remains elusive, maintaining a policy impasse.

Regulatory Decisions

Glyphosate has undergone several regulatory reviews in Europe – in 2002, 2012, 2017, and most recently in 2019–2023 – with the European Commission renewing its approval until 2033. This decision was based on the European Food Safety Authority's (EFSA) assessment, despite political and public opposition and two unsuccessful committee votes due to disagreement among the European Member States. EFSA concluded that there was insufficient evidence to justify a ban but implemented restrictions, such as prohibiting the use of glyphosate in public spaces and playgrounds. The approval for continued use also involves strict limits on the amount of glyphosate permitted in various food products, known as Maximum Residue Levels (MRLs). These levels are established as safe thresholds to prevent toxic effects from consumption.

All this creates the impression of a strict and well-regulated control system. However, despite claims of safe levels for each type of food (fruit, meat, drinks...), daily consumption results in cumulative exposure to glyphosate, potentially exceeding the allowable daily intake (ADI). Even more concerning is the nearly constant—and involuntary—exposure to these low concentrations, raising essential questions about their long-term effects on human health and sustainability. This is a crucial factor to consider in risk assessment and management.

Economic and/or financial interests

There is no doubt: glyphosate is the most successful herbicide ever produced. The glyphosate market is estimated to be between 600.000 and 800.000 tonnes/year globally. The market value of glyphosate is estimated at 10-15 billion dollars in 2030. Useless to say that this is a firm financial and economic argument in the hands of those who are in favour of retaining and intensifying the use of glyphosate as the herbicide of choice.

Illustrative of the economic and financial significance are the intensive lobbying efforts by companies producing glyphosate across various decision-making bodies. Data from lobbyFacts.EU for 2022 reveals that Bayer spends over 6 million euros at the European level, with a significant portion likely allocated to supporting the approval for the continued use of glyphosate.

A complete and immediate ban on glyphosate would entail a substantial increase in food costs. Consumers' demand for quality food will remain, and other methods of controlling weeds will be needed, among them costly hand labour. Hence, the balance between economic and financial considerations/financial and health considerations, on the other hand, dictates a gradual, well-controlled phasing out of glyphosate over sufficient time to allow agriculture to adopt alternative methods. This should also prompt producers of agrochemicals to seek alternatives to glyphosate.

Ethical Considerations and the Involuntary Nature of Exposure

The modest carcinogenicity of glyphosate is often cited as a sufficient argument not to discontinue its use. However, this argument is flawed for two reasons. Unlike voluntary exposures such as tobacco, alcohol, or red and/or processed meat, exposure to glyphosate is primarily involuntary. People have limited options to avoid dietary and environmental exposure to glyphosate and similar substances, making it essential for regulatory bodies to apply the precautionary principle when uncertainties persist. Unfortunately, this ethical perspective is largely absent from EFSA's public communications and scientific evaluations.

This ethical component is further emphasized when considering glyphosate as part of the exposome – the totality of (chemical) exposures an individual encounters throughout life. The ubiquitous use of glyphosate makes it highly likely that glyphosate is, and probably an important component of, the exposome. The concept of the exposome further strengthens the notion of chemical interactions, although this is scientifically underexplored.

Food Waste, Waste Management, and Chemical Redistribution

Waste – broadly defined as any human-made material requiring disposal or repurposing – has become more complex since the start of industrialisation, especially with

the rise of synthetic chemicals, some of which are found in everyday food. Although some food waste is destroyed during household waste management, practices such as redistributing surplus food and using food as animal feed do not eliminate chemical residues in food. Notably, the Environmental Protection Agency (EPA – USA) identifies “Donate or Upcycle” and “Animal Feed” as valuable methods for reducing the environmental impact of food waste (EPA, 2025). An estimated 30% of our food waste is repurposed as animal feed, much of which is then reintroduced into the human food chain. This is not a call for the destruction of more food waste, but rather an appeal to reduce glyphosate levels in food, thereby preventing continued human exposure.

Taken together, a substantial portion of chemically contaminated food waste is incorporated into circular economy models, such as the reuse of human food waste for animal feed. This practice is problematic unless it is accompanied by rigorous chemical monitoring and comprehensive regulatory reform.

Conclusions

The 2023 reauthorization decision by the European Commission illustrates the broader tensions between regulatory standards, scientific uncertainty, and precautionary principles. The widespread use of glyphosate epitomizes a broader systemic challenge at the intersection of food production, public health, environmental ethics, and economic interests. While regulatory bodies continue to emphasize insufficient evidence for restricting glyphosate, the growing body of toxicological data suggests the need for a more precautionary and ethically attuned approach.

Ultimately, addressing this issue demands an integrated response: advancing our understanding of chemical exposures through scientific innovation, reassessing regulatory frameworks in light of ethical considerations, and reforming waste management systems to prevent the continuous spread of chemical contaminants. The recently launched International Research Association on Circular Economy (IRACE) exemplifies the kind of initiative required (IRACE, 2025). Only through such interdisciplinary efforts can we begin to reconcile the competing demands of feeding the global population while safeguarding human and environmental health.

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MICROWAVE-BASED PYROMETALLURGICAL PROCESS TO PRODUCE FERROSILICON WITH PV CELLS FROM EOL PV PANELS

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ABSTRACT

In this work, silicon recovered from a PV panel recycling facility was utilized as a reactant for the synthesis of ferrosilicon alloys via microwave treatment in the presence of iron oxides and graphite. Microwave-driven processing was further explored using ladle furnace slag from secondary steelmaking as a reactive component. Different amounts of reactants, time and microwave power were studied. Microwave heating allowed to reach temperatures higher than 900°C after few minutes of treatment at a power of 600 W. The materials were characterized by SEM-EDS and XRD. The proposed process allowed to obtain a ferrosilicon alloy with an amount of 21% Si. This study explored the use of three different types of waste, PV silicon, iron oxide and ladle furnace slag to obtain a ferroalloy FeSi which can be afterwards employed in the steelmaking process.

1. INTRODUCTION

In recent years, many significant advances were made regarding the recovery of silicon-based solid waste from the photovoltaic (PV) industry. In fact, the demand for silicon for PV production has increased due to the rise in raw materials cost, whereas the amount of global waste of PV panels is forecasted to reach 80 million tonnes by 2050 (Dubey et al., 2013; Guo et al., 2021; Preet & Smith, 2024): all these factors have stimulated the research in PV recycling.

Many studies demonstrated the feasibility of industrial-scale recovery of materials from end of life PV panels, showing that aluminium, silver, copper, glass, and silicon can be recovered (Cerchier et al., 2021; Huang et al., 2017; Pagnanelli et al., 2017). However, reintegration of the recovered silicon into the photovoltaic value chain still faces some challenges in terms of purity requirements and for now remains in the laboratory scale stage (Ngagoum Ndalloka et al., 2024). It is therefore essential to find new applications to extend the silicon reuse and promote sustainable waste recycling.

The possible routes to reclaim PV silicon waste are many, from processing it in order to meet the requirements of Si anodes for Li-ion batteries (C. Zhang et al., 2021), to using it as a leading element in the metallurgy of Si-based

ferroalloys. Ferrosilicon plays a fundamental role in the de-oxidizing and alloying of steel, their production being a very energy-intensive process.

A study proposed to synthesize ferrosilicon alloys by using waste glass and bakelite recovered from the automotive sector, showing an efficient reduction of iron oxide into Fe₃Si (gupeiite) (Farzana et al., 2014). Another work introduced a novel method for the production of ferrosilicon alloys, recovering two types of critical waste materials: Si PV cells and red mud. They demonstrated that the target composition of Fe-Si alloy could be achieved with Si deviations of 15 wt%, together with the formation of Fe-Si-Ti phases (Blaesing et al., 2024). The chemical composition of ferrosilicon alloys usually ranges between 15 and 90 wt% silicon, depending on the desired application. (Rauf Hurman, 2014).

The present study explores the possibility to employ different types of waste for producing ferrosilicon alloys, using microwave pyrometallurgical process to obtain a target composition respecting the ones typically found in the market; in particular, photovoltaic panels scrap and ladle furnace slag (LFS) as source of Si.

LFS (also known as "white" slag) is a by-product of ladle steelmaking refinement, characterized by a chemical composition rich in CaO, SiO₂, Al₂O₃, and MgO, along with oth-



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er trace elements (Najm et al., 2021). While it has chiefly been disposed of through landfilling, LFS has recently been recognized as a sustainable material for next generation eco-friendly construction industry (Aaros et al., s.d.; Yong et al., 2025; N. Zhang et al., 2025); its mineralogical properties make it in fact a suitable cement binder supplement or replacement (Adolfsson et al., 2011; Disconzi & Bellotto, 2025). These features further highlight its potential for integration into alternative recycling routes, such as the one presented in this study.

The proposed technique enables to perform a carbothermic reduction by using microwave radiation instead of conventional furnace treatments, therefore not only promoting an opened-loop recycling path for PV-Si and LFS, but also testing the efficiency of alternative and more eco-friendly energy sources.

The use of microwaves to perform reduction reactions of different types of ores has recently gained a lot of attention, owing to its good heat selectivity and rapid processing times (Peng & Hwang, 2015). For instance, studies conducted on MW-processed low-grade iron ores demonstrated that reduction was achieved in 10 minutes, reaching temperatures in the order of 800°C (Rayapudi et al., 2020).

In this study a characterization of the products obtained from MW heating was carried out, evaluating the feasibility of achieving a carbothermic reduction of iron ores mixed with Si-based PV cells, recovered from waste PV panels, and ladle furnace slag, another waste produced by the steelmaking industry.

2. MATERIALS AND METHODS

2.1 Materials

Recycled silicon wafer fragments for the experiments performed in this study were supplied by 9-Tech (Venice, Italy), from their pilot plant in Venice. In this facility, raw materials from photovoltaic panels are recovered through a thermomechanical treatment, which enables the separation of glass, aluminium, copper, and PV cell fragments. The latter can be further processed using ultrasound treatment to remove silver and aluminum. After this step, the fragments consist mainly of metallic silicon (>96%) and aluminum (<3%). (Cerchier et al., 2024). The treated PV cells were ground with a ball milling machine operated at

30 Hz for 10 s, until a particle size below 45 µm was obtained.

The iron oxides mix and ladle furnace slag used for conducting the experiments were supplied by a company in Northern Italy.

Synthetic graphite (<20 µm, SigmaAldrich) was added to the powder mix as a reducing reagent as well as heating element, owing to its high absorbance ability towards microwaves.

Before microwave heat treatment, the powders were dried for 24 hours in a BINDER FD 53 heating chamber.

2.2 Experimental

A domestic Panasonic NN-GD351W microwave oven operating at 2.45 GHz frequency was used for pyrometallurgical tests, which were performed in air atmosphere.

Alumina crucibles were used as powder containers for the reaction and a silicon carbide (SiC) susceptor was employed as a heating auxiliary. In fact, dielectric materials like SiC strongly absorb microwave radiation and can therefore passively help the heating of a reaction mixture in the MW field by converting electromagnetic energy into thermal energy (Ishizaki & Nagata, 2007; Singh et al., 2024).

The temperature of the specimens during the experiments was measured by means of a FLIR T420 infrared camera.

The weight in grams (g) of each reactant used in the experiments is reported in Table 1, while the selected process parameters are reported in Table 2.

The input reagents underwent crystallographic analysis through X-ray diffraction (Bruker D8 Advance, Karlsruhe, Germany), conducted in the 2θ range of 15-75°, with a step size of 0.02° and a counting time of 1 s. The X-ray diffraction spectra of all the input reagents are presented in Figure 1.

Sample 1 was obtained by mixing equal fractions of slag and metallic PV-Si, which was distributed at the bottom of the crucible to create a powder bed. The sample was treated at 600 W for 70 s.

Test 2 was performed maintaining the powder proportions of test 1 but decreasing the overall weight to 0.37 g. The experiment was carried out at 600 W for 120 s.

A third experiment was carried out with the same process parameters of test 2 on a sample with equal amounts

TABLE 1: Weight (g) of the reactants used for each experiment.

Sample	Iron oxide [g]	White slag [g]	PV Si [g]	Graphite [g]
1	0.304	0.117	0.106	0.202
2	0.153	0.053	0.055	0.105
3	0.1	-	0.1	0.1

TABLE 2: Microwave process parameters for each test.

Sample	Power [W]	Time [s]	Susceptor	Total weight [g]
1	600	70	No	0.7
2	600	120	Yes	0.37
3	600	120	Yes	0.3

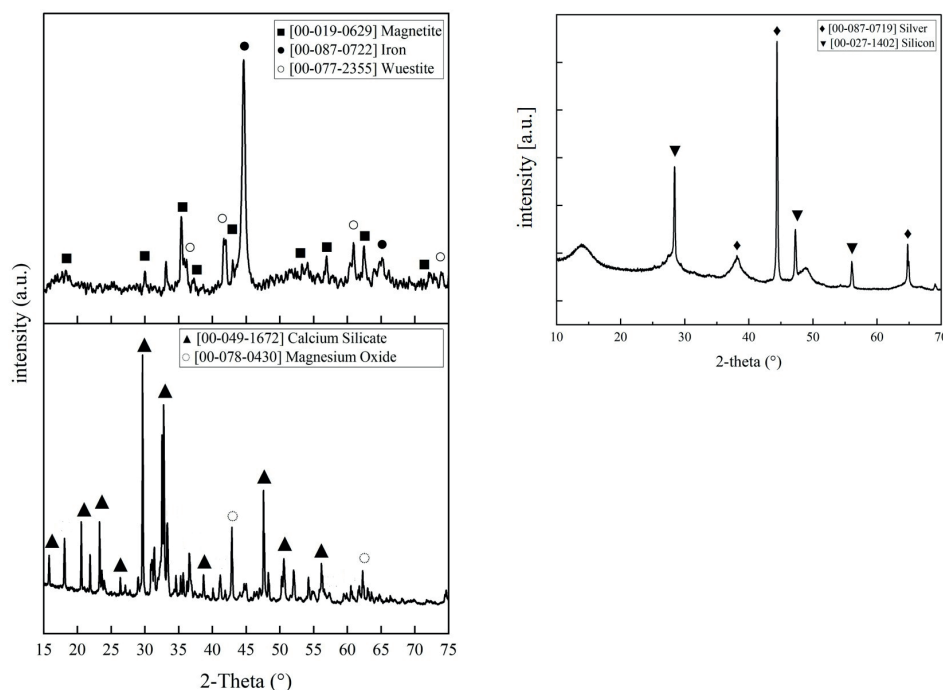


FIGURE 1: a-c) XRD spectra of the input reagents.

of iron oxide, PV-Si and graphite, without adding the white slag powder.

The MW treated metallic samples were mounted into a polymeric resin and polished with standard metallographic preparation as per ASTM E3.

A Zeiss EVO-M10 scanning electron microscope (SEM), equipped with an Oxford Instruments energy-dispersive X-ray spectroscopy (EDS) detector was employed for surface characterization of the specimens.

3. RESULTS AND DISCUSSION

3.1 Results

The specimens obtained after microwave heating were cooled in air and then carefully examined in order to identify the formation of alloys from the initial powder. During microwave exposure of the samples, the reactants are heated with the aid of the susceptor, until they reach a high enough temperature to react with each other and melt. During the reaction, oxidation of the graphite into CO_2 takes place, while iron oxides are reduced into metallic iron. When MW irradiation is interrupted, the molten material starts solidifying and during its cooling off, alloy particles are formed until room temperature is reached.

From SEM characterization images in back scattered electrons mode (BSE) it was observed that the alloy nuggets often nucleated from white slag particle clusters, as inferred from the EDS spectrum 1 in Figure 2, showing peaks belonging to Ca, Si, Al and Mg oxides. Given the higher atomic weight of iron, the alloy colour appears white compared to the slag, which instead appears gray/black.

From low magnification SEM analysis of the particles of sample 1, the majority of the metallic areas seems to

be made of iron and carbon nodules, forming a structure similar to that of nodular cast iron (Figure 3).

However, at higher magnification the presence of small biphasic regions rich in both Fe and Si was detected (Figure 4).

Composition analysis performed with EDS demonstrated that the darker phase is characterized by a higher silicon content, while the lighter is enriched in manganese, as can be observed in the elemental map of Figure 5. The ratio between Si and Fe is equal to about 3:1 (Table 3).

Specimen 2 was subjected to a weight loss of 16% after microwave treatment, probably caused by the formation of CO and CO_2 gases produced by the carbothermic reaction. Thermal camera pictures were taken after 30, 60 and 100 s during the experiment (Figure 6) and display a significant increase in the temperature after 1 minute of exposure to microwaves, reaching a maximum of 930°C at the end of the test.

EDS analysis (Figure 7) shows that the selected test parameters allowed to obtain alloy particles with varying contents of Si, while oxygen and other elements were undetected. Magnification of such areas confirmed the formation of the two different phases: the lighter one containing about 16 wt % Si and 1.13 wt % Mn, respectively, while the darker one 23 wt % Si and 2.27 wt % Mn. The bright white particles were instead found to be impurities of Sn and Cu, probably belonging to some residues from the welded joints in PV electronic circuits.

The third sample was obtained without adding the LFS fraction, to verify how its presence affected the composition of the alloy. SEM analysis showed a morphology very similar to the one observed in cast iron, with carbon flocules dispersed in a Fe matrix (Figure 8). EDS analysis outlined a content of Si equal to 16 wt %.

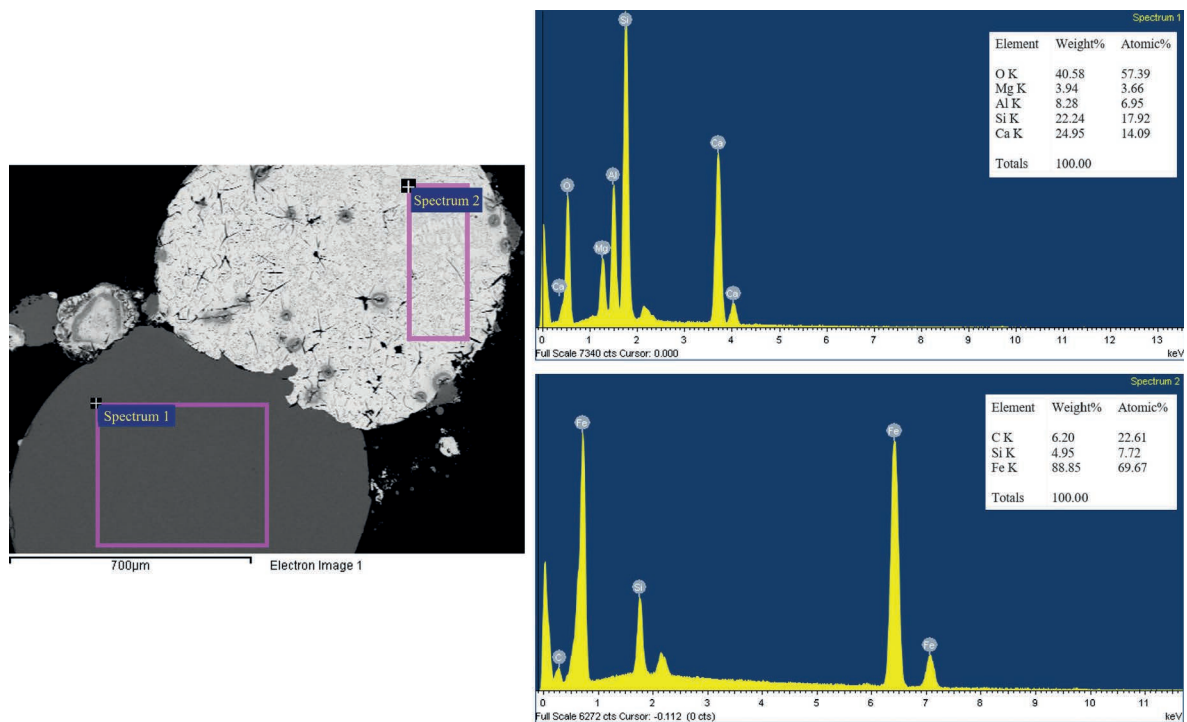


FIGURE 2: SEM-BSE and EDS spectrum of alloy nugget nucleated from LFS granule (sample 1).

3.2 Discussion

Characterization of the MW-synthesized samples demonstrates the feasibility of producing a silicon ferroalloy with MW treatment of silicon PV cells and LFS in presence of iron oxides and graphite.

For sample 1, the addition of a limited amount of Si PV cells powder allowed to obtain the localized formation of a FeSi alloy, unevenly distributed throughout the LFS matrix. The EDS spectrum denoted a content of silicon lower than 10%, which is slightly below the required levels for the pro-

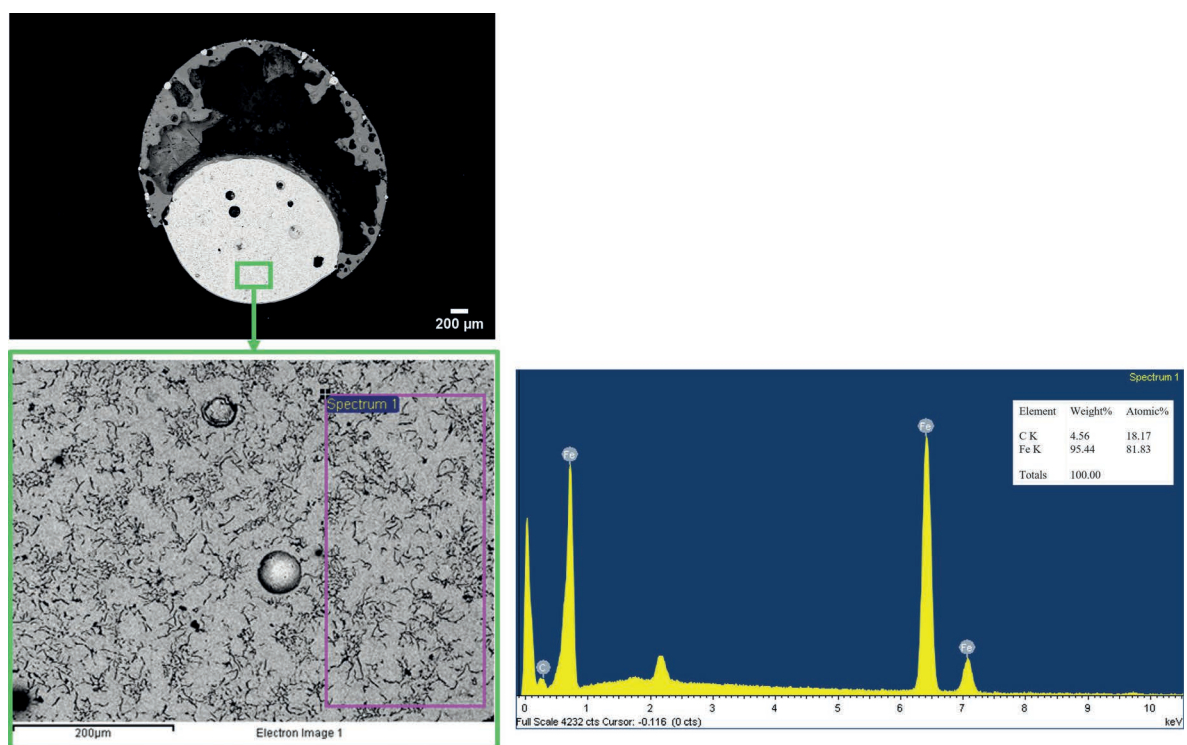


FIGURE 3: SEM-EDS of metallic area in sample 1.

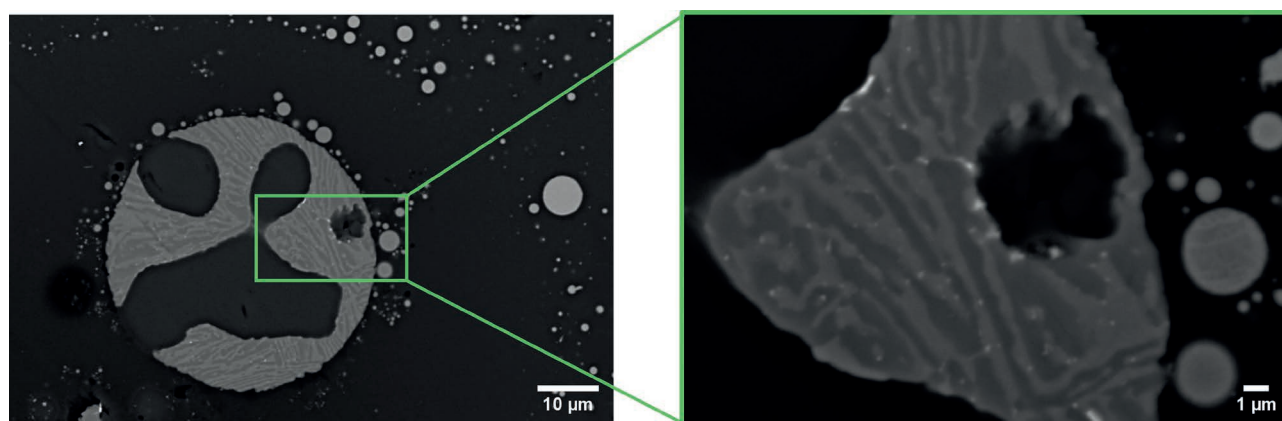


FIGURE 4: High SEM-BSE magnification of biphasic FeSi alloy detected in sample 1.

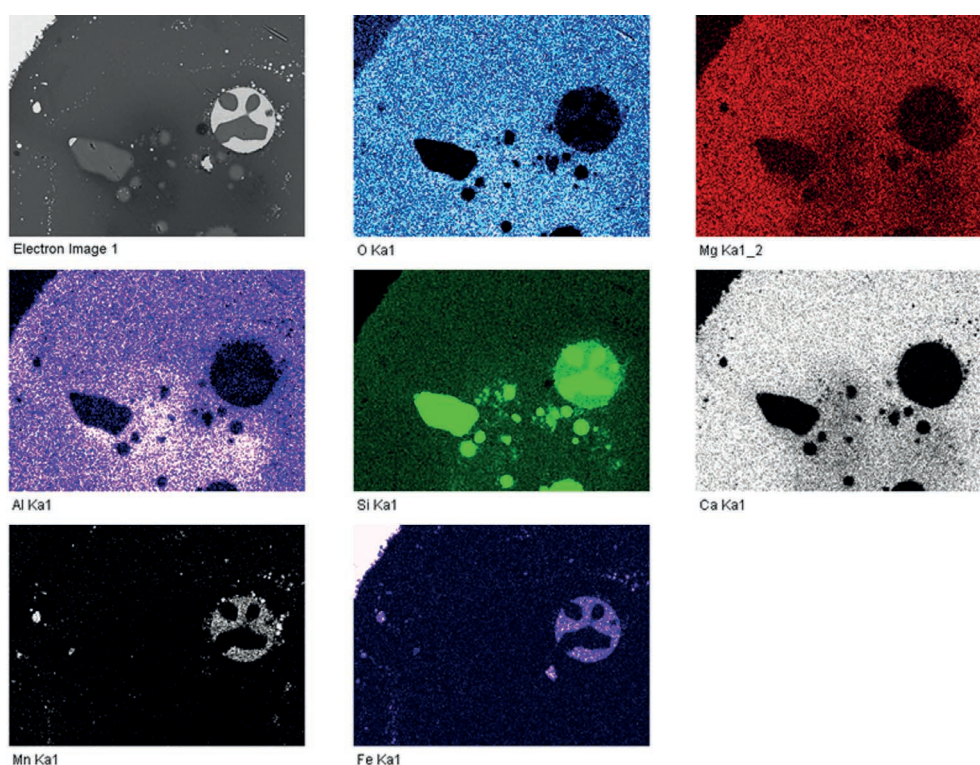


FIGURE 5: SEM-EDS elemental map of FeSi alloy in sample 1.

duction of ferrosilicon (Rauf Hurman, 2014). The majority of metallic particles synthesized during the treatment exhibited a composition rich in iron and carbon, without a discernible presence of silicon. The low content of silicon in the alloy of sample 1 is likely due to low reaction efficiency, possibly resulting from the absence of the SiC susceptor and thus the inability to reach the required temperature for suitable element diffusion.

Experiment 2 led to the formation of a FeSi alloy in which the composition of 15-23 wt% silicon is consistent with the lower limit of the ranges usually required for ferrosilicon alloys.

The lower total weight of the reactants of test 2 (0.37 g) compared to test 1 (0.73 g) probably supported a more effective utilization of the output electromagnetic energy, facilitating the efficient volumetric heating of a smaller

TABLE 3: Chemical composition of Fe-Si-Mn alloy particle formed during test 1.

Elements [wt. %]	Si	Cr	Mn	Fe	Ti
Dark phase	67.98	0.72	10.18	21.11	-
Light phase	61.52	2.14	20.05	14.85	1.44

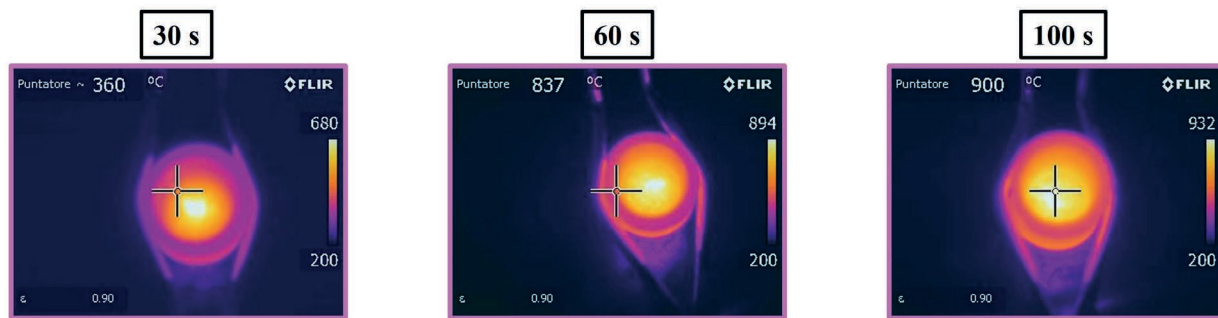


FIGURE 6: Thermal camera images taken during test 2.

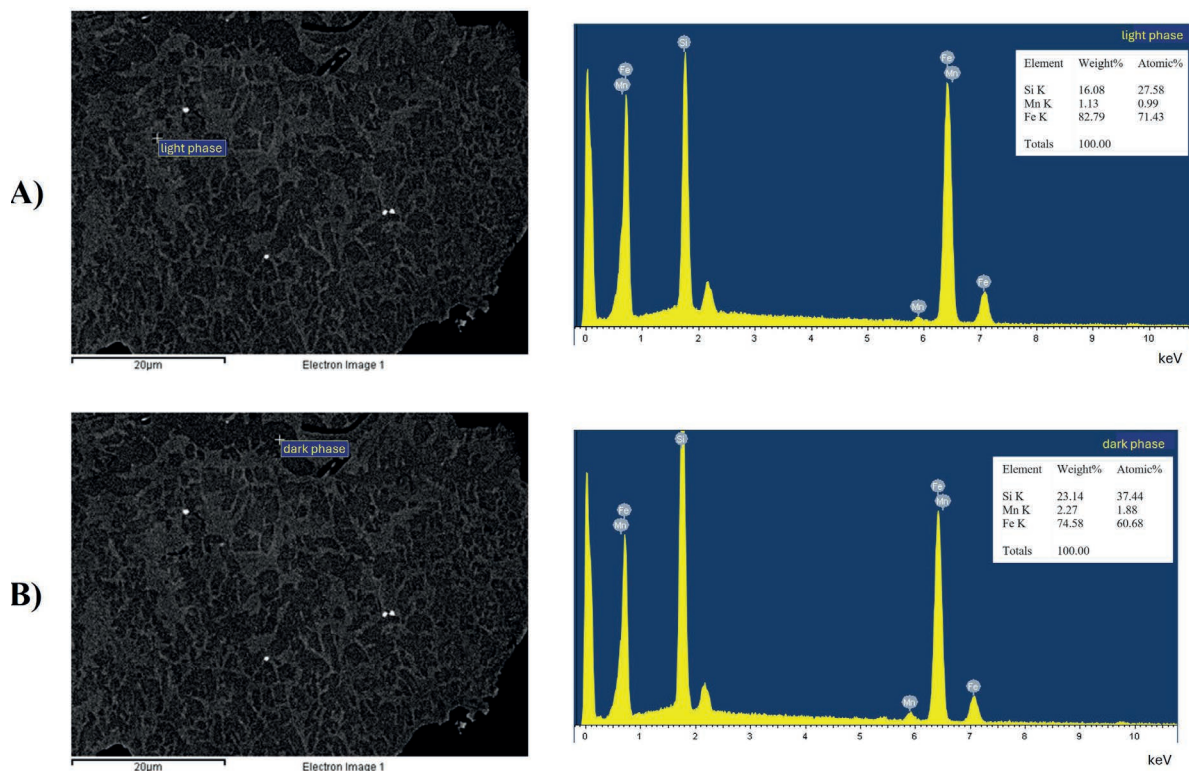


FIGURE 7: EDS spectra of the two phases detected in specimen 2.

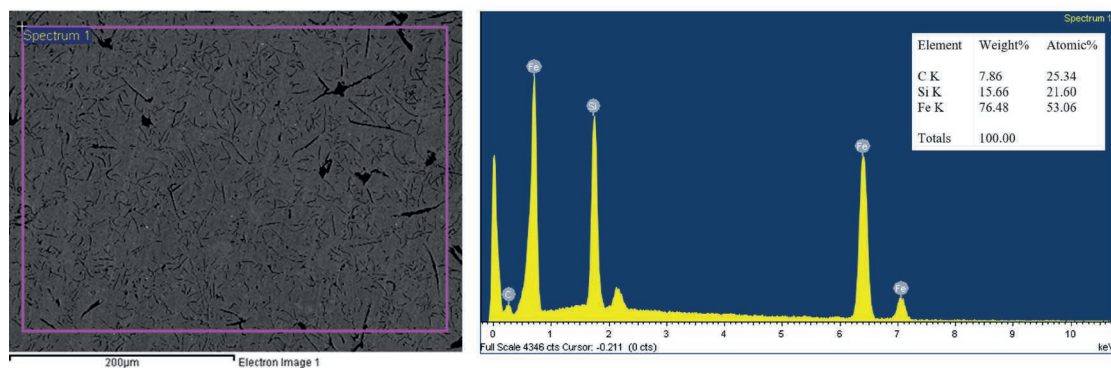


FIGURE 8: EDS spectrum of alloy in sample 3.

amount of powder. An approximate estimate of the energy consumed to heat sample 2 can be obtained by considering that by setting a power of 600 W and heating for 120 s,

the required theoretical energy would be of 72 kJ (energy = power * time). Other more detailed studies on the efficiency of MW heating assert that its energy consumption

can be a hundred times lower than the one of, for example, electrical heating (Zhu et al., 2018).

Moreover, presence of the SiC susceptor and setting of longer microwave exposure time contributed to a more resource-effective carbothermic reaction.

Compared to test 2, test 3 (during which LFS was not added to the reaction) led to the formation of a ferrosilicon alloy with lower silicon content (about 16 wt%). However, the reaction outcome seems to be promising and could be further optimized.

4. CONCLUSIONS

The present study has demonstrated that it is possible to produce a Si ferroalloy using a microwave pyrometallurgical process, which utilizes silicon recovered from PV panels, white slag, and iron oxides as reagents, and graphite as a reducing agent. The performed experiments showed that all the reactant dosages led to the synthesis of a ferrosilicon alloy, differing in the composition due to different reaction efficiencies. SEM-EDS characterization of the specimens highlighted that the alloy reached the target composition by adding equal proportions of PV-Si and LFS. This result was obtained after optimization of the total weight of the specimen and of the MW parameters, leading to the conclusion that longer exposure time for a smaller amount of powder facilitated the carbothermic reaction.

Thermal camera images revealed that the required temperatures for the pyrometallurgical process can be reached after only 100 s of MW exposure.

In the future, further research will be carried out for larger-scale production of ferrosilicon by microwave heating.

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ASSESSMENT OF HEAVY METALS AND PAHs IN FISH WASTE VALORISATION FOR NUTRACEUTICAL PURPOSES

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ABSTRACT

This study presents results on contaminant quantifications for the food safety of initial biomass (fish waste) and its extract product (fish oil) resulting from the activities carried out within the VITADWASTE project. VITADWASTE is a PRIN (Progetti di Rilevante Interesse Nazionale) project funded by Italian Ministry of University Education and Research, which aims to use a multidisciplinary and sustainable approach to transform biological waste into a valuable resource, to develop a safe and effective vitamin D3 nutraceutical product for human use, based on the principles of circular economy and bioeconomy. Fish waste includes by-products such as bones, guts, heads, skin and fins, or species below the minimum conservation size and tissues not suitable for human consumption, which together account for about 20-80% of the total catches and therefore improving the management of such biomass is essential. In the VITADWASTE project, an innovative green extraction method has been developed to recover vitamin D3 from fish waste, to produce a fish oil usable for nutraceutical implementations. Here, the results of Heavy Metals (HMs) and Polycyclic Aromatic Hydrocarbons (PAHs) concentrations are reported in the fish wastes and the fish oil according to the EU regulation. The consistently low concentrations of contaminants, always within regulatory limits, confirm the safety of both the raw material and the extracted product for human use, and support the adoption of SFE as a sustainable and efficient strategy for the valorisation of marine biomass within a circular economy framework.

1. INTRODUCTION

Fish plays an important role in human diet due to its high-quality proteins and beneficial effects. In particular, fatty fish, such as bluefish, contain more micronutrients than other meats, including vitamin D in fish oil and minerals like iron, selenium, zinc, iodine, magnesium and calcium, being a good food source for humans. In recent years, fish production, including that from aquaculture, has seen significant growth, reaching 223 million tons (FAO 2024, Umesh et al., 2025). In addition, the amount of fish waste from fishing has increased. Fish waste includes both com-

mercial species below the minimum conservation reference size and tissues not suitable for human consumption (such as bones, guts, heads, skin and tails), which together account for about 19% and 20-80% of the total catches, respectively (Kruijssen et al., 2020). Meanwhile, the fish by-products from industrial processing can result in waste levels of up to 70% (Dawson et al., 2025).

According to EU strategies, which aim for sustainable economic and environmental growth, and in line with Sustainable Development Goals (SDGs) and circular economy objectives, it is necessary to promote new challenges for the valorisation of fish waste. Up to date, fish waste is



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partly used to produce fishmeal, fertilizers (Radziemska et al., 2019; Lopes et al., 2021), and low-profit fish oil, or as raw material for direct feeding in aquaculture (Meena et al., 2024), while the rest is discarded. Indeed, research is growing in interest in such biomass for their bioactive compound's richness (Caruso et al., 2020; Rigueto et al., 2023).

In this context, VITADWASTE project aims to convert and reuse fish waste into a valuable substrate to obtain extract rich in bioactive compounds, particularly vitamin D3 and omega-3 fatty acids, suitable for pharmaceutical and nutraceutical sectors.

For this reason, this work aligns with SDG 12 (Responsible Consumption and Production), specifically with Objective 12.5: "substantially reduce waste generation by 2030 through prevention, reduction, recycling, and reuse," as well as with SDG 3: "ensuring healthy lives and promoting well-being for all at all ages."

Given that fish have the capacity to bioaccumulate a range of environmental contaminants through their diet and habitat, including polycyclic aromatic hydrocarbons (PAHs), heavy metals (HMs), and other contaminants of emerging concern (CECs), any valorisation process must rigorously assess and ensure the safety of these residues. This is particularly important when the final products are intended for human consumption (De Giovanni et al., 2022) or use in the nutraceutical sector.

For food safety, Regulation (EU) 2023/915 sets limits for contaminant levels in fish and certain food supplements, although fish-derived supplements are not as extensively regulated as others. The threshold limits from Regulation 2023/915 relevant to the samples considered in this study are presented below.

The regulation defines maximum allowable levels of heavy metals like lead (Pb), cadmium (Cd), and mercury (Hg) in fish, expressed in terms of wet weight (ww) of the edible muscle tissue, unless the entire fish is intended for consumption. The limit for Pb is set at 0.3 mg/kg. For cadmium, the general threshold is 0.05 mg/kg, though this increases to 0.1 mg/kg for mackerel (*Scomber* spp.) and to 0.25 mg/kg for anchovy (*Engraulis* spp.) and sardine (*Sardina pilchardus*). Mercury has a general limit of 0.5 mg/kg, but this rises to 1 mg/kg in the case of red mullet (*Mullus barbatus barbatus*).

Regarding polycyclic aromatic hydrocarbons (PAHs), the regulation only provides limits for smoked fish. In such cases, the maximum permitted concentration for benzo(a)pyrene is 2 µg/kg, while the sum of benzo(a)pyrene, benzo(a)anthracene, benzo(b)fluoranthene, and chrysene must not exceed 10 µg/kg.

At present, there are no specific regulatory limits for PAHs in fish oils. However, for other food supplements governed by Regulation 2023/915, where fish-derived products are excluded, the threshold for benzo(a)pyrene is set at 10 µg/kg. Regarding oils and fats, the regulation states: "oils and fats placed on the market for the final consumer or for use as food ingredients, except cocoa butter and coconut oil. This maximum level applies to vegetable oils used as an ingredient in food supplements", and for these, the limit is set at 2 µg/kg. Therefore, the improvement of fish waste management is essential to address environmental con-

cerns and to fully utilize and valorise this biomass, for high value commercial purposes (Coppola et al., 2021).

The purpose of this paper is to assess if the fish oil derived from fish biomass waste, produced with Supercritical Fluid Extraction (SFE) at pilot scale, is suitable for human consumption and appropriate for nutraceutical applications, by evaluating the concentrations of potential contaminants which could be bioaccumulated by fishes and therefore transferred to the fish oil.

2. MATERIALS AND METHODS

2.1 Fish sampling and pretreatment

Five fish species from the Adriatic Sea were chosen for their good source of Vitamin D3 (Ostermeyer et al., 2006). Specifically, three pelagic species such as anchovy (*Engraulis encrasicolus*), sardine (*Sardina pilchardus*), and mackerel (*Scomber scombrus*), and two demersal species such as red mullet (*Mullus barbatus barbatus*) and European hake (*Merluccius merluccius*) were chosen for the study.

Fishing activities were conducted in the northern and central Adriatic Sea using commercial fishing vessels. Two types of fishing gear were employed: pelagic nets, used primarily for catching small pelagic species that inhabit the upper layers of the water column, and trawl nets, used for harvesting demersal species typically found near the seabed.

The collected fish waste was refrigerated on board and then transported to the laboratory, where it was separated by species and immediately homogenized using an automatic mill. The homogenized fish waste was then frozen at -20°C and subsequently freeze-dried.

2.2 Sardine oil production

Sardine oil was extracted using a Luwar supercritical fluid pilot plant (ENEA Research Centre Casaccia, Rome) with a 564 ml extraction vessel. The extraction was performed in duplicate, and each test treated 100 g of lyophilized sardine waste. The extraction pressure was 310±10 bar, the temperature was 40±0.4°C, and the supercritical fluid density was 916.0±4.9 kg/m³. The supercritical CO₂ flow rate was 0.46±0.03 kg/min. The process duration was calculated from the achievement of operating conditions and was 150 minutes for both extractions. The recovery yield was 14.6±0.1%, calculated as the ratio of the amount recovered from the plant to the dry matrix loaded (w/w).

2.3 Contamination analysis

Contaminant analysis were performed in both fish waste (HMs, PAHs) and fish oil (Cd, Hg, Pb).

2.3.1 HMs analysis

For HMs analysis, pools for each species were performed, as two aliquots were collected after the homogenization process, and used for the contaminant analysis of those regulated by EU 2023/915 (Hg, Cd, Pb) and others HMs and metalloids non regulated (Al, Ag, As, Fe, Mn, Ni, Co, Cu, Cr, Se, Zn) which will be available in the supplementary material (S1.1. and S1.2.).

Fish waste mineralization was performed using a MARS-6 Microwave Assisted Acid Digestion, by adding 3 ml of ultrapure nitric acid and 3 ml of hydrogen peroxide to about 0.5 g of homogenized and freeze-dried fish and then the solution was diluted to 20 ml using ultrapure water (18.2 MΩ cm at 25°C).

Sardine fish oil digestion was performed by microwave-assisted acid digestion using a MARS6 microwave and the CEM method "Microwave Digestion of Fish Oil (No Capsule)", by adding to 0.2 g of fish oil 10 ml of ultrapure HNO₃. After the digestion, the solution was diluted with 10 mL of Milli-Q ultrapure water (18.2 MΩ cm at 25°C), yielding a 20 ml clear and colorless solution, which was then analyzed by GF-AAS for Pb and Cd.

Inductively coupled plasma optical emission spectroscopy (ICP-OES) was used for all the HMs determinations in the previously mineralized fish waste, except for mercury. Microwave digestion program and ICP-OES operative parameters are extensively reported in Girolametti et al., (2024).

Hg determinations for both fish waste and oil were carried out using Thermal Desorption Atomic Absorption Spectrometry (TD-AAS) with a Direct Mercury Analyzer (DMA-1). Fish waste Hg quantification were performed in the freeze-dried sample, while fish oil was directly measured without further preparations. Three replicates were analyzed for each sample, by directly measuring 0.1 g of dry weight (dw) for fish waste and 0.02 g of fish oil. The TD-AAS program consisted in three different temperature steps: 1 minute at 200°C, followed by 2 minutes at 650°C, and a final 1-minute hold at 650°C.

The contaminant concentrations in the fish waste are expressed as mg/kg of wet weight (ww) by normalizing their dw concentrations with the water content removed from the sample through the freeze-drying procedure, determined by differential weighing before and after the drying process.

Pb quantification in the fish oil was performed using the standard additions method, while Cd was determined by calibration curve. For both elements, a palladium-based matrix modifier was used to enhance GF-AAS measurement accuracy.

The Limits of Detection (LODs) were evaluated using the calibration curve method and are reported in Table 1 for the regulated heavy metals (HMs), and in Table S1 for the non-regulated ones.

2.3.2 PAH analysis

PAHs extraction from fish waste was performed using the QuEChERS (Quick, Easy, Cheap, Effective, Rugged, and

Safe) technique, which involves a first extraction phase using small amounts of solvent (acetonitrile), separated from the aqueous matrix with magnesium sulfate and sodium chloride, followed by a further purification phase through a 0.2 µm filtration, with a slight modification of the method described by Frapiccini et al., (2018).

16 PAHs were investigated using for calibrations a standard PAH solution (EPA 610 PAHMIX, Supelco, Bellefonte, PA, USA) with dichloromethane:methanol (1:1 v/v) containing naphthalene, acenaphthene, fluorene, phenanthrene, anthracene, fluoranthene, pyrene, chrysene, benzo(a)anthracene, benzo(b)fluoranthene, benzo(k)fluoranthene, benzo(a)pyrene, dibenz(a,h)anthracene, benzo(g,h,i)perylene, indeno(1,2,3cdi)pyrene.

The quantification of PAHs was performed using UH-PLC equipped a FLD Dionex UltiMate 3000 (Termofisher, Italy). Acenaphthylene was not considered because of it does not have fluorescent properties, therefore could not be measured through UHPLC-FLD.

The chromatographic separation of PAHs was performed over a 40-minute run using a reversed-phase gradient elution of acetonitrile (ACN) and water (v/v%). The gradient started at 10% ACN and linearly increased to 100% ACN, which was maintained for the final 5 minutes of the run to ensure effective column cleaning prior to subsequent injections. A fixed injection volume of 1 µL was employed for each analysis. Detailed conditions of UHPLC-FLD analysis are reported in Table 2.

Limit of detection (LOD) of PAHs were evaluated directly from the calibration curve with Chromeleon software using Hubaux-vos at 95% confidence interval.

3. RESULTS AND DISCUSSION

3.1 Heavy Metal(loid)s and PAHs concentrations

3.1.1 In the Fish waste

The contaminants investigated in the five species of the fish waste are HMs and PAHs. Particular attention was given to the three heavy metals regulated by EU 2023/915: Hg, Cd and Pb, which were measured in both the sample typologies. The concentrations of HMs are expressed in mg/kg_{ww}. The results of the non-regulated HMs will be available in the supplementary material (Figure S1 and Figure S2).

In the analysed fish waste samples, the concentrations of Hg, Cd and Pb never exceeded the limits set by EU 2023/915 (Table 3). To facilitate the comparison between measured values and regulatory limits, the ratio between the Regulatory Limit (R.L.) and the Measured Value (M.V.) is also reported in Tables 3 and 5.

TABLE 1: Limits of detection (LOD) for the regulated HMs (Cd, Pb and Hg). Concentrations on mg/kg are evaluated considering the digestion volume and dilution utilized for the quantifications, where necessary.

Analytical Instrument	Matrix	Cd		Pb		Hg
		µg/L	mg/kg	µg/L	mg/kg	mg/kg
ICP-OES	fish waste	0.05	0.002	3.7	0.15	
GF-AAS	fish oil	0.13	0.014	2.1	0.201	
DMA	fish waste and oil					0.003

TABLE 2: Chromatographic and analytical parameters for PAH determination by UHPLC-FLD.

PAH	Retention time (min)	λ_{ex} / λ_{em}	Calibration range (ng/ml)	Calibration algorithm	R ²	LOD (ng/ml)
naphthalene	8.305	265ex/360em	10-495	Linear	0.99997	6.0
acenaphthene	11.439	265ex/360em	10-543	Linear	0.99999	3.6
fluorene	12.005	265ex/360em	2.5- 97.8	Linear	0.99990	2.0
phenanthrene	13.289	265ex/360em	1.3-50	Linear	0.99973	1.6
anthracene	14.398	265ex/410em	4.5-50	Linear	0.99975	1.6
fluoranthene	15.777	290ex/480em	9.7-96.3	Linear	0.99992	2.3
pyrene	16.617	270ex/390em	1.4-51	Linear	0.99995	3.0
benzo(a)anthracene	19.914	265ex/410em	1.0-49	Linear	0.999997	0.1
chrysene	20.481	265ex/410em	0.9-48.2	Linear	0.99993	1.1
benzo(b)fluoranthene	23.148	265ex/410em	1.5-94.2	Linear	0.999996	1.3
benzo(k)fluoranthene	24.281	265ex/410em	0.9-48.7	Linear	0.99998	0.4
benzo(a)pyrene	25.331	265ex/410em	1.4-51	Linear	0.99990	1.0
dibenz(a,h)anthracene	27.331	265ex/410em	2.5-96.2	Linear	0.99986	1.9
benzo(g,h,i)perylene	28.131	290ex/480em	1.8-97.2	Linear	0.99991	2.4
indeno(1,2,3cdi)pyrene	28.827	265ex/410em	3.9-47	Linear	0.99999	1.0

TABLE 3: Comparison of HM concentrations measured in the fish waste samples and the regulatory limits of the EU 2023/915. R.L./M.V. expresses the factor by which the Measured Values (M.V.) falls below the Regulatory Limit (R.L).

Fish Waste	Cd			Hg			Pb	
	Measured Value (mg/kg _{ww})	Regulatory Limit (mg/kg _{ww})	R.L./M.V.	Measured Value (mg/kg _{ww})	Regulatory Limit (mg/kg _{ww})	R.L./M.V.	Measured Value (mg/kg _{ww})	Regulatory Limit (mg/kg _{ww})
Red Mullet	0.0035 ± 0.0002	0.05	14.3	0.045 ± 0.005	1	22.2	< 0.037	0.3
Sardine	0.0138 ± 0.0008	0.25	18.2	0.027 ± 0.001	0.5	18.8	< 0.037	0.3
European Hake	0.0064 ± 0.0024	0.05	7.9	0.048 ± 0.001	0.5	10.4	< 0.037	0.3
Mackerel	0.0034 ± 0.0008	0.1	29.4	0.062 ± 0.001	0.5	8.1	< 0.037	0.3
Anchovy	0.017 ± 0.001	0.25	14.4	0.038 ± 0.002	0.5	13.1	< 0.037	0.3

Pb concentrations were undeterminable due to values below the detection limit, attesting to be below 0.037 mg/kg_{ww} in all the samples (fish waste concentration as mg/kg_{ww} was evaluated by assuming LOD as minimum concentration and using the average digestion mass, volume, dilution and % dry weights for the calculations), with concentrations at least 8.1 times lower than the regulatory limit.

Hg and Cd, however, showed quantifiable but still non-harmful concentrations, as they always resulted below the threshold limit in all the studied species (Table 3).

Cd concentrations ranged between a minimum of 0.0034±0.0008 mg/kg in mackerel and red mullet to a maximum of 0.0174±0.0008 mg/kg in the anchovy, with intermediate values for European hake, 0.0064±0.0024 mg/kg, and sardine, 0.0138±0.0008 mg/kg, with values below the regulatory limit from 7.9 to 29.4 times, depending on the

species. The higher Cd concentrations observed in pelagic species such as anchovy and sardine, compared to demersal species, mirror the higher maximum limits established in Regulation EU 2023/915. This is due to the tendency of Cd to accumulate in the phytoplankton, and thus in the upper photic zone of the water column (Mart & Nürnberg, 1986), which leads to an increase in the bioavailability of Cd to these fishes, because of their diet that is typically plankton-phytoplankton based.

Hg concentrations ranged from minimum values of 0.0265±0.0012 mg/kg in sardine to a maximum value of 0.0618±0.0008 mg/kg in mackerel, with intermediate values for anchovy, red mullet, and European hake. The comparison with regulatory limits (R.L./M.V.) shows concentrations from 8.1 to 22.2 times below the threshold (Table 3).

PAH concentrations were low or absent in the fish waste, in particular for the most relevant PAHs (benzo(a)

TABLE 4: PAH concentrations in the fish waste (r1 and r2 are referred to different fish waste replicate). < LOD, indicates that the analyte concentration is below the detection limit.

PAH compounds	European hake r1	European hake r2	Mackerel r1	Mackerel r2	Sardine r1	Sardine r2	Anchovy r1	Anchovy r2	Red mullet r1	Red mullet r2
naphtalene	< LOD	< LOD	0.18	0.43	0.46	< LOD	0.58	0.56	1.20	1.05
acenaphthene	< LOD	< LOD	< LOD	< LOD	< LOD	< LOD	< LOD	< LOD	2.46	2.36
fluorene	< LOD	< LOD	0.60	0.55	< LOD	< LOD	< LOD	< LOD	1.66	1.93
phenanthrene	< LOD	< LOD	< LOD	0.36	0.28	< LOD	1.26	1.23	< LOD	0.32
anthracene	< LOD	< LOD	< LOD	< LOD	< LOD	< LOD	< LOD	< LOD	< LOD	< LOD
fluoranthene	< LOD	< LOD	< LOD	< LOD	< LOD	< LOD	< LOD	< LOD	< LOD	< LOD
pyrene	< LOD	< LOD	< LOD	< LOD	< LOD	< LOD	< LOD	< LOD	0.17	0.16
benzo(a)anthracene	< LOD	< LOD	< LOD	< LOD	< LOD	< LOD	0.04	0.04	0.07	< LOD
chrysene	< LOD	< LOD	< LOD	< LOD	< LOD	< LOD	< LOD	< LOD	< LOD	< LOD
benzo(b)fluoranthene	< LOD	< LOD	< LOD	< LOD	< LOD	< LOD	< LOD	< LOD	< LOD	< LOD
benzo(k)fluoranthene	< LOD	< LOD	< LOD	< LOD	< LOD	< LOD	< LOD	< LOD	< LOD	< LOD
benzo(a)pyrene	< LOD	< LOD	< LOD	< LOD	< LOD	< LOD	< LOD	< LOD	< LOD	< LOD
dibenz(a,h)anthracene	< LOD	< LOD	< LOD	< LOD	< LOD	< LOD	< LOD	< LOD	< LOD	< LOD
benzo(g,h,i)perylene	< LOD	< LOD	< LOD	< LOD	< LOD	< LOD	< LOD	< LOD	< LOD	< LOD
indeno(1,2,3cdi)pyrene	< LOD	< LOD	< LOD	< LOD	< LOD	< LOD	< LOD	< LOD	< LOD	< LOD

pyrene, benzo(a)anthracene, benzo(b)fluoranthene and chrysene) regulated in the EU 2023/915 (Table 4).

Benzo(a)pyrene, benzo(a)anthracene, benzo(b)fluoranthene and chrysene were not identified in nearly all samples, with quantifiable concentrations only for benzo(a)anthracene in the anchovy (0.04 µg/kg) and in only one replicate (0.07 µg/kg) of the red mullet, indicating safety for the consumption.

The highest PAH concentrations for the fish wastes were found for the lowest molecular weight compounds (LMW-PAHs) such as naphthalene, acenaphthene and fluorene; this was particularly evident for the red mullet, where its demersal behavior and preferred muddy habitat plays a key role for the bioaccumulation of such compounds mainly through the diet, even if this does not represent a matter of concern regarding this contamination for humans. Also, LMW-PAHs, due to their lower log K_{ow}, are more water-soluble and tend to associate with suspended particles that settle in sediments. This makes them more bioavailable to demersal species that live and feed near the sediment-water interface (Frapiccini et al., 2020).

Pyrene also showed measurable concentrations of 0.17 and 0.16 µg/kg only in red mullet, while in the rest of

the investigated species was not detectable. Also, traces of Phenanthrene of 1.26 and 1.23 µg/kg were found in the anchovy, even if this does not represent a matter of concern.

3.1.2 In the extract product (fish oil)

The fish oil production operations were achieved using the green-innovative Supercritical Fluid Extraction (SFE). This technique was chosen because of SFE uses CO₂ as extracting solvent instead of organic – and generally harmful – solvent of the traditional extracting techniques, which gives a first insight for the possibility to the usage for nutraceutical. Also, the investigated species Vitamin D-3 extraction yield of SFE was compared with traditional methods like Ultrasound Assisted Extraction (UAE) and Solid-Liquid Extraction (SLE), resulting in comparable yields among the different techniques, confirming its suitability. A comparison of Vitamin D-3 concentrations obtained under different SFE extraction conditions are described in detail in a separate study (Alessandroni et al., under review).

Therefore, according to the purpose of the VITAD-WASTE project, the different species of fish waste were tested, and as sardine waste resulted to provide a suitable oil for the nutraceutical usage, considering both the high oil extraction yield and Vitamin D-3 concentration, the contamination analysis for the regulated HMs were then conducted.

The concentrations of the contaminants, expressed as average ± standard deviations, are compared with the maximum allowed concentrations established for the food supplements in the EU 2023/915, which establishes limits of 3, 1 and 0.1 mg/kg respectively for Pb, Cd, and Hg (Table 5).

Hg concentrations in the fish oil were found to be 3.11±0.16 µg/kg, which is approximately 32 times lower than the regulatory limit. Similarly, Pb and Cd concentra-

TABLE 5: HMs comparison between the concentrations found in the sardine fish oil and the EU regulation 2023/915 threshold levels. R.L./M.V. expresses the factor by which the Measured Values (M.V.) falls below the Regulatory Limit (R.L.).

HMs	Measured Value µg/kg	Regulatory Limits, EU 2023/915 µg/kg	R.L. / M.V.
Hg	3.11 ± 0.16	100	32
Pb	48.7 ± 6.9	3000	62
Cd	23.4 ± 1.2	1000	43

tions were $48.7 \pm 6.1 \mu\text{g/kg}$ and $23.40 \pm 1.18 \mu\text{g/kg}$, respectively. These values are about 62 times lower than the regulatory threshold for Pb and 43 times lower for Cd, as reported in Table 5. Therefore, based on these results, no health risk was identified for consumers in relation to these contaminants.

4. CONCLUSIONS

In this study, the food safety of both the initial product (fish waste) and the extract product (fish oil) derived from the VITADWASTE project was assessed with respect to contamination by heavy metals (HMs) and polycyclic aromatic hydrocarbons (PAHs), in accordance with Regulation (EU) 2023/915.

Previous investigations within the project tested supercritical fluid extraction (SFE) as a sustainable method for recovering bioactive lipophilic compounds from fish waste. After optimizing the extraction process and comparing different species, sardine waste was identified as the most suitable raw material for producing nutraceutical-grade fish oil, which was subsequently analyzed for contaminants.

From a food safety perspective, no critical levels of contamination were observed. Concentrations of both inorganic (Hg, Cd, Pb) and organic (PAHs) contaminants in the raw biomass and extracted oil were well below the thresholds established by EU legislation. HMs in the fish waste were found to be approximately 8 to 29 times lower than regulatory limits, and no heavy PAHs were detected except for minimal traces of pyrene and benzo(a)fluoranthene, without posing any health risks due to their low concentrations. In the extracted oil, HM concentrations ranged from 32 to 62 times below legal thresholds.

These findings confirm the safety of both the raw material and the extracted product for human use and high-light SFE as a viable and sustainable strategy for marine biomass recovery in line with circular economy principles.

Although this represents an initial validation for the food safety, future work will focus on scaling up the process to obtain larger quantities of oil enriched in vitamin D and polyunsaturated fatty acids, aiming to formulate nutraceutical products for evaluation in clinical studies.

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INSECTERA: GREEN TRANSITION TOWARDS ENVIRONMENTAL SUSTAINABILITY BASED ON A CIRCULAR ECONOMY

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ABSTRACT

Extracting valuable platform chemicals from waste biomass is a crucial part of the path to developing a circular, sustainable materials economy. As a particularly significant example, recent developments at the insect biorefinery – utilization of insect biomass – are herein discussed. The focus is placed on the valorisation of *Hermetia illucens* (commonly known as Black Soldier Fly, BSF) waste created during their industrial production process. In particular, pupae exuviae of BSF are used in studies aimed at obtaining useful raw materials, such as chitin and chitosan, for other industry sectors, namely the health industry.

1. INTRODUCTION

The insect industry is gaining global recognition for its role in sustainable waste management and the development of high value bioproducts. Black Soldier Fly (BSF), mealworms, and crickets are among the most widely farmed species due to their rich protein content, rapid growth, and minimal environmental footprint. Of these, the Black Soldier Fly (BSF) stands out for its remarkable ability to convert organic waste into valuable resources: BSF larvae efficiently break down food waste and agricultural by-products (e.g., fruit and vegetable wastes, kitchen wastes, and brewery's byproducts) (Alciatore et al., 2024), reducing environmental pollution while generating protein, oil, and frass (insect manure) for various applications.

In food and animal feed, insects provide a sustainable alternative to traditional protein sources like soy and fishmeal, while agricultural applications focus on organic fertilizers and pest control. Beyond its well-known use in these sectors, BSF waste is now attracting attention in the cosmetic and pharmaceutical industries. The oils and bioactive compounds derived from BSF biorefinery offer potential as sustainable ingredients for skincare and personal care products (Lai-Foenander et al., 2024). Rich in antimicrobial peptides, fatty acids, and antioxidants, these insect-derived compounds are being explored for their moisturizing, anti-aging, and protective properties.

Portugal has been making significant developments in the insect industry, focusing on sustainable food production and innovative applications. Recently, the Mobilizing Agendas for Business Innovation, framed in the Recovery and Resilience Plan (RRP) (<https://recuperarportugal.gov.pt/>), emerged with the objective of consolidating and expanding synergies between the Portuguese business fabric and the scientific and technological system, contributing to the increase in the competitiveness and resilience of the Portuguese economy, based on R&D, innovation and the diversification and specialization of the productive structure.

It was in this context that, in 2023, the InsectERA Mobilizing Agenda (<https://www.insectera.pt/english/>) was created, bringing together stakeholders from across the value chain who want to explore the potential of insects as a bioindustrial tool and bioindustrial solutions. The InsectERA Agenda covers the entire innovation cycle, from Research & Development to the manufacturing and commercialization of products on the market, based on technologically advanced production methods.

Our task within the Agenda is to valorise insect bio-waste through its technological conversion into industrial raw materials and high-value-added products. In particular, pupae exuviae of insects such as *Hermetia illucens* (Black Soldier Fly, BSF) are used in studies aimed at obtaining useful components, such as chitin and chitosan for the health industry. These BSF derivatives are directly applied in the



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production of new high-value-added products, such as cosmetics for dermatological and haircare applications.

The industrial production of Black Soldier Fly involves several steps, according to its life cycle (Figure 1).

For all stages of the life cycle, except for the pupal stage, the BSF form is utilized and commercialized, generating value. Therefore, throughout the entire BSF cycle, only the pupae are considered waste (Figure 2), as it has no valuable application so far.

Our task is, therefore, to change this paradigm and identify a commercially viable product, thereby completing the BSF-based circular economy. The BSF pupae exuviae are processed to extract their chitin, as explained in the following section.

2. CHITIN EXTRACTION PROCESS

Chitin is one of the most abundant (with annual biological production levels of ca. 10^{11} tonnes – Tharanathan et al., 2003), natural polysaccharides, a linear polymer composed of repeating β -(1,4)-N-acetylglucosamine units (i.e., between 55% and 98% of units by weight, preferably between 75% and 90% of units by weight, Figure 3) that has generated the most interest.

In insects in particular, chitin makes up between 3 and 60 per cent of their exoskeleton.

Extracting chitin from BSF pupae exuviae requires several steps (Figure 4):

- Milling the exuviae;
- Demineralization of exuviae in acid medium;
- Deproteination of exuviae in basic medium;
- Isolation of the chitin;
- Purification of isolated chitin.



FIGURE 2: Life cycle of *Hermetia illucens* (Black Soldier Fly, BSF) illustrating the industrial raw-materials obtainable from each stage. Currently the pupal stage has no applications.

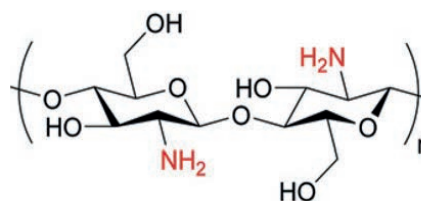


FIGURE 3: Repetitive unit representation of the biopolymer chitin.

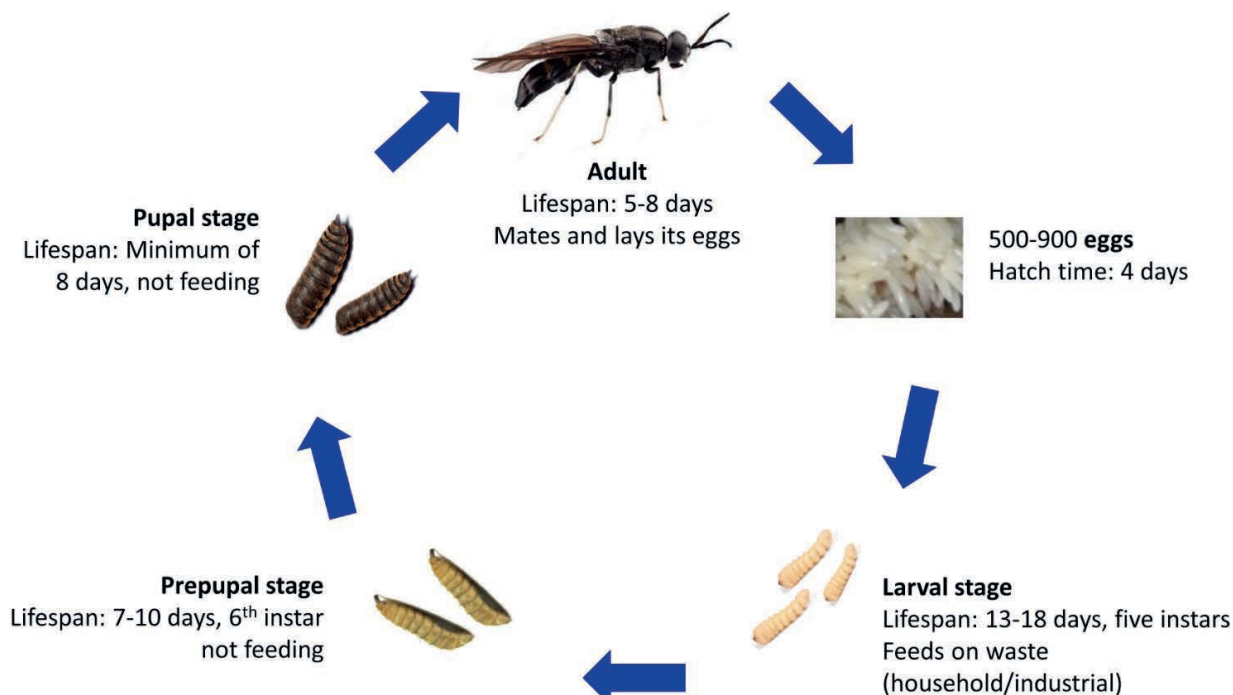


FIGURE 1: Life cycle of *Hermetia illucens* (Black Soldier Fly, BSF).

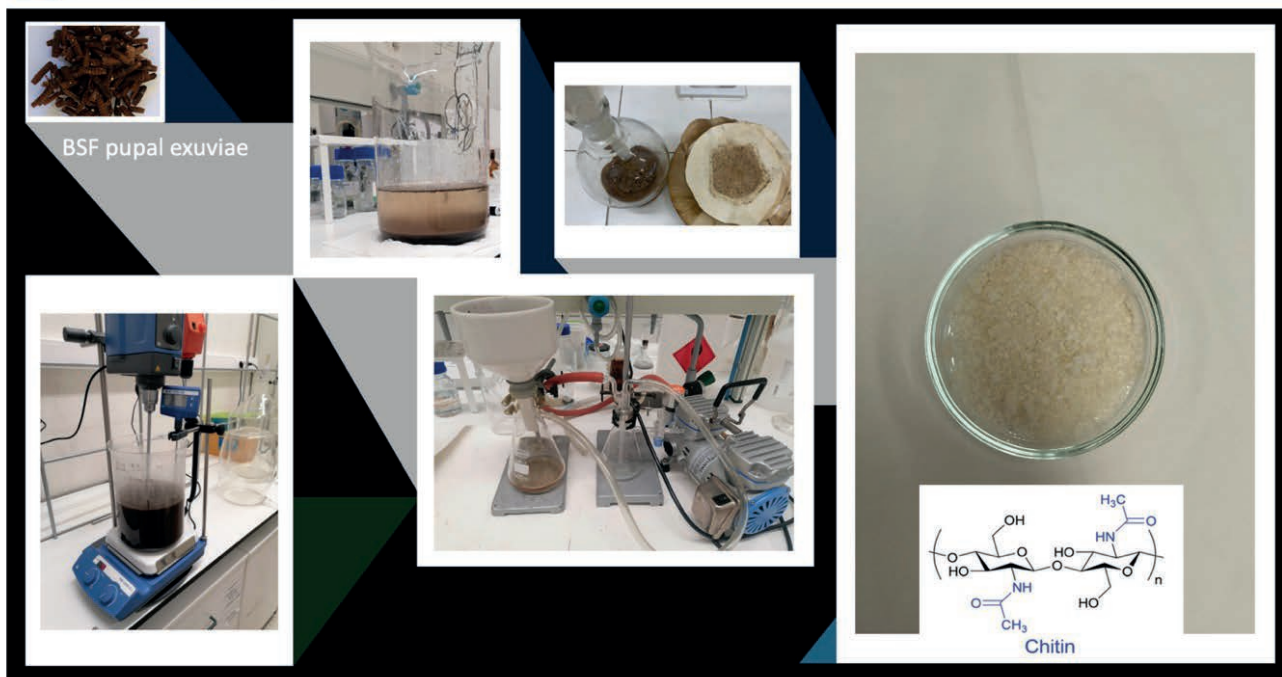


FIGURE 4: Extraction of chitin from BSF pupal exuviae.

The technological and ecological aspects of this lab protocol developed at IST are considered by including dual-purpose reagents that can preserve the characteristics of the purified chitin and can be produced from low-cost biomass.

3. CHITOSAN PRODUCTION PROCESS

Chitin can undergo deacetylation, i.e., the conversion of amide functions into amine functions, giving rise to chitosan (Figure 5), a water-soluble biopolymer that has great chemical versatility as well as antibacterial and antifungal properties.

In fact, chitosan already has a worldwide market that reached a value of USD 2.49 billion in 2020 and a compound annual growth rate (CAGR) of approximately 12% is projected between 2021 and 2026.

It is known that the degree of deacetylation of chitin as well as the average molecular weight and polydispersity of

the chitosan obtained play a crucial role in the properties of chitosan-based biopolymers, and consequently in their applications. For example, the use of a chitosan biopolymer with a high average molecular weight maximises its antimicrobial effect (Matica et al., 2019) and limits trans-epidermal water loss or skin irritation (Feng et al., 2021).

Thus, one of the challenges of this work was to provide a process for producing chitosan biopolymers with a high average molecular weight and a high degree of deacetylation and purity that is, at the same time, environmentally favourable both in terms of energy and resource consumption and in terms of reducing waste generation.

4. CONCLUSIONS

With the increasing global demand for sustainable solutions, combined with government support and technological advancements, the insect industry is poised to

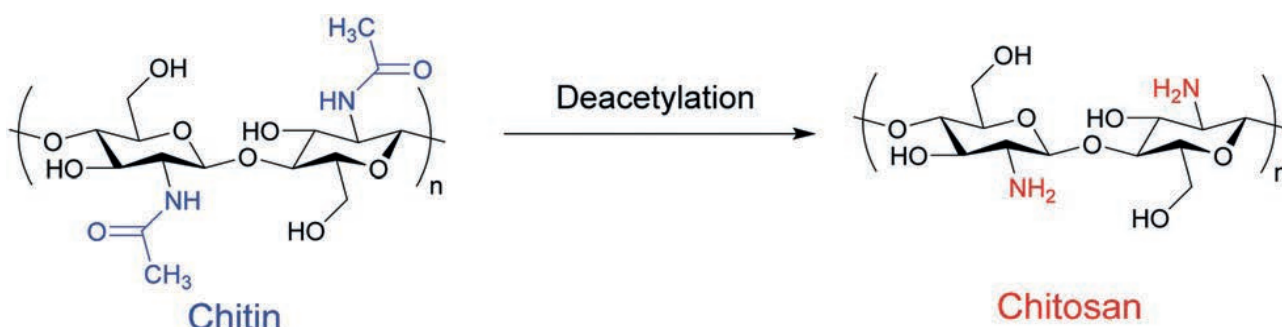


FIGURE 5: Production of chitosan by deacetylation of chitin.

transform multiple sectors, driving circular economy practices and minimizing environmental impact.

The process developed within the InsectERA Mobilizing Agenda effectively utilizes surplus biomass from the agri-food sector - specifically BSF exuviae - allowing it to be reintegrated into the original ecosystem, closing the economic circle. This approach not only prevents waste but also promotes the use of insect-based products.

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INNOVATIVE BIOENERGY SOLUTIONS FOR DEVELOPING NATIONS: DARK FERMENTATION AND ANAEROBIC DIGESTION OF ABATTOIR WASTE

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ABSTRACT

This study investigates the energy recovery potential of abattoir waste by subsequent Dark Fermentation (DF) and Anaerobic Digestion (AD) processes. The 2-reactor system assesses the effect of recycling and parametric optimisation. A simulation model based on the Anaerobic Digestion model 1 (ADM1) was developed and applied for biogas production, which defines carbohydrates, lipids and proteins (CLP) as the main components of anaerobic digestion. The motivation for this work emanates from that a major cause for failure of anaerobic plants is inconsistent feedstock and process design, thus, process modelling provides a cheaper and more reliable evaluation of process components and parameters. Furthermore, a great amount of abattoir waste is generated, coupled with difficulty in disposal thus raising the waste management cost. The study investigated the effect of CLP on biogas yield and assessed the outcome from a co-digestion of manure, blood, and tissue as major abattoir waste streams. Impact on hydrogen yield was in the order of lipids>carbohydrates>proteins whereas for methane it was carbohydrates>lipids>proteins. Manure had the highest impact on methane yield rate, followed by blood, and tissue, whereas hydrogen production was in the order of blood, tissue, and lastly manure, which performed poorly. Recycling improved methane yield by 32%. The study provides optimisation data and linear correlation models for estimating yield based on the three substrates. The study furthermore presents hydrogen and methane potential of various abattoir waste stream. Based on the South African waste stream, there is potential to generate 0,068-156,26 GW of energy from abattoir waste.

1. INTRODUCTION

Biological processes such as anaerobic digestion are receiving global attention due to their positive socioeconomic impact and contributions to environmental remediation. Likewise, many developing countries such as South Africa are tapping into technologies such as anaerobic digestion as a cost-effective and environmentally benign method that can be used in the valorisation of abattoir waste. South Africa is experiencing a steady growth in meat consumption leading to a rise in slaughterhouse (abattoir) facilities and an increased throughput. This has resulted in increased waste generation from the sector. The waste generated in these facilities includes manure, wastewater, offals, blood, rumen content, confiscates (lungs, livers, kidneys), feathers, hooves, animal tissue (Neethling, 2014; Roberts et al., 2009). The South African Red Meat Abattoir Association (2021) reported over 9,544,704 slaughters of cattle, sheep

and pigs in 2021. This accounts for around 1,078,182 tons of waste produced, assuming a waste production rate of 60%, 55%, and 60% for a 500kg cow, 50kg sheep, and a 100kg pig respectively. It was reported that about 46-50% of a cow, 38-40% pig, 28-32% chicken waste, 22% turkey, and 48% sheep/goat ends up as waste after slaughter (Tolera & Alemu, 2020). There was a reported 2 586 334 cattle, 3 547 843 sheep and 3 410 527 pigs slaughtered from the 436 abattoirs in 2021 by the Red Meat Association. There is also around 2 033 000 slaughters of goats in the country (Qekwana, 2012). About 2,3 million chickens were culled in 2018 (DTI, 2019). In 2016, 1500 tonnes of broiler meat was produced from 265 formal abattoirs (CSIR, 2018). It was furthermore stated that about 12,8 litres per bird was produced as wastewater from the poultry abattoirs (CSIR, 2018). Jayatilakan et al. (2012) provided a waste stream analysis for the cattle, pig, sheep and poultry by-products.

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Whole carcass constituted 77,5%, 63% and 62,5% for pig, cattle and sheep body weight respectively. Other wastes include Liquid Blood with 3%, 18%, and 2,4% respectively, and fats with 3%, 4% and 3% respectively. For hides and skin, the order is 6%, 6%, 15% and organs there is 7%, 16%, and 10% of the body weight. Chest and abdomen are 10% for pigs and sheep, and 11% for cattle. Feet constitute 2% for each animal stated. According to the paper, poultry feathers are 7-8% of live weight, blood is 3,2-3,7%, gizzards and proventriculus equal 3,5-4,2%, feet with 3,5-4% and intestines and glands constituting 8,5-9%.

According to the Water Research Commission, over 7.2 million m³ of wastewater is discharged from the South African abattoir industry with a Chemical Oxygen Demand (COD) of 730-9930 mg/l, pH of 5.7-8.4, suspended solids of 189-3330 mg/l, and a Kjeldahl Nitrogen of 0.71-24 mg/l (GDARD, 2009; Müller, 2017; Water Research Commission, 2017). Much of the wastewater is discharged via municipal lines after pretreatment, due to the high cost of treatment for reuse/recycling. The organic load for slaughterhouse waste other than wastewater is reported as 470-960 mg/l O₂ (BOD₅) and 960-1280 mg/l O₂ (COD) (Borowski & Kubacki, 2015; Boughou et al., 2018; Matheri et al., 2017; Palatsi et al., 2011; Singh et al., 2018; Staroń et al., 2017).

The high organic load of the waste has made it difficult for abattoirs to effectively manage it, and it was found out that there is a continuous yearly increase in deviations from adherence to regulations by abattoirs. This is especially worse for small scale abattoirs (Roberts et al., 2009). As a consequence, outbreaks such as listeriosis in 2017, and pathogenic avian influenza in 2023, which resulted in human death and culling of over 7,5 million egg laying chickens was observed (DTI, 2019; Tchatchouang et al., 2020; Thomas et al., 2020). The common disposal methods in South Africa include waste disposal facilities (WDF), burial on farms or private land, and less common methods such as incineration, composting, rendering, and anaerobic digestion (Western Cape Government, 2016). There is a need to assess opportunities for valorisation of the waste stream to maximise the value of abattoir waste, which may effectively improve the economics of the facilities.

The recent developments in waste management as countries heed the need for environmental protection and climate change have led to new regulations regarding waste handling and disposal. In South Africa, non-infectious abattoir waste falls under the classification of class B landfills (low risk hazardous waste landfills), of which many municipalities have only a few or none, thus raising the difficulty in disposal logistics (Gogela et al., 2017). This has had dire implications for the abattoir industry, which has begun experiencing some of the highest increases in the cost of disposal due to stringent disposal regulations and long-distance transportation costs. As a trade-off to the waste management costs, a study by Gogela et al. (2017) drew a financial feasibility for a medium to large scale biogas plant producing facility (>50 kilowatt-electric (kWe)) at an abattoir site in South Africa, whereby an abattoir can benefit directly from the energy produced. South Africa is furthermore aiming to cut organics to landfill and is leading toward a landfill ban, and municipalities such as

Western Cape are already implementing such restrictions since 2019. As such, it will be beneficial for waste producers to find alternative waste treatment technologies to cut the rising costs of handling and disposal.

This study assesses the energy recovery potential of abattoir waste via dark fermentation and anaerobic digestion. There is little information about the valorisation of abattoir waste for hydrogen production, and especially optimisation information and potential for upscaling in South Africa. This is due to the high protein and fat content of the waste stream. Nevertheless, whereas it makes for a good substrate for AD, the slow hydrolysis rates and process inhibition makes them a less desirable feedstock (Palatsi et al., 2011). Thus, the aim was to investigate the availability and energy potential of the South African abattoir waste stream. The study presents methane and hydrogen yield potential of the typical abattoir waste stream components, and their corresponding energy based on their abundance. The composition of the waste stream is obtained from literature, whereas energy yield was achieved using the Aspen Plus simulation model. There is limited literature providing detailed biochemical waste stream characterisation data, especially on carbohydrates, proteins and lipids. However, articles such as Palatsi et al (2011), and Hejnfelt and Angelidaki (2009) provide more comprehensive characterisation data which was useful for simulating the anaerobic process. The 2-stage process has hydrogen production via dark fermentation, followed by methane production in the second stage. As per our understanding, dark fermentation has the highest potential for hydrogen production and feasibility for upscaling among biological pathways, and a technological integration of hydrogen and methane via AD provides the highest feasibility towards commercialisation due to the advancement in the application of the conventional anaerobic digestion (Ahmed et al., 2021; Dell'Orto & Trois, 2022). Combination with other technologies such as photofermentation, gasification, microbial electrolysis cells and microbial fuel cells has been studied in various literature (Osman et al., 2020; Sekoai et al., 2018; Sittijunda et al., 2022). The underlying motivation for integration, however, is that less than 20% of biodegradability is achieved during dark fermentation (Jain et al., 2024), and furthermore, multistage processes were seen to improve the yield, reduce the overall fermentation period, improve the energy balance of the process and provide a better handling of the substrates. Dawei (2008) achieved a 21% improvement to hydrogen yield in a 2-stage arrangement, and an 88% increase by recycling and sparging methane from the second reactor. The author also found out that a multistage system is more tolerable to inconsistent feedstock or varying feed composition. Other authors also found considerable gains conversely by sparging hydrogen gas in the methane reactor and gaining 42% in energy yield while enhancing biogas quality by 20% (Ghorbanian, 2014). The findings of this work are an important knowledge base for anaerobic digestion of abattoir waste, basing on the fact that a major reason for failure of such plants is the feedstock and substrate compositional inconsistencies. Therefore, understanding how the composition affects yield can be useful information for upscaling and maintaining a digester, par-

ticularly in the case of a double stage process. Developing countries can benefit from this innovative solution to ease the landfilling capacity and exploring opportunities for energy recovery from costly waste streams such as abattoir wastes. The simulation developed is not limited to abattoir waste only but can be applied to any biodegradable waste resource.

2. MATERIALS AND METHODS

2.1 Model building and development

The anaerobic process was developed and modeled using Aspen Plus software, as a modification to previous models by (Rajendran et al., 2014; Serrano & Knud, 2011). The model and reaction inputs were based on the Anaerobic Digestion Model 1 (ADM1 model) as described in (I. Angelidaki et al., 1993, 1999). The basic model development, including the set of reactions and reaction kinetics was obtained from the mentioned literature, and applied to a 2-stage process for the production of hydrogen and biogas in separate reactors. Greater details of the methodology are therefore found in the literature stated (Rajendran et al., 2014; Serrano & Knud, 2011). Based on the ADM1 model, the input components were defined as water, lipids (triolein, tripalmitin), proteins (soluble and insoluble), carbohydrates (cellulose, hemicellulose) and inert compounds. These form the basis for anaerobically degrading materials. The Non-Random Two Liquids (NRTL) method was also maintained as in previous studies due to appraisal for accurate calculation of mole fractions, and activity coefficients and suitability for gas and liquid phases and since it was already validated against experimental and plant data for anaerobic digestion modelling. Due to the limiting nature of the hydrolysis phase, the set of reactions were modelled separately in a stoichiometric reactor (Rstoic), which required the extent of reaction and reaction stoichiometry as inputs. Modelling hydrolysis separately allows for testing the effects of pre-treatment on conversion efficiencies. In the current model, eleven hydrolysis reactions were added, gathered from (I. Angelidaki et al., 1993; I. Angelidaki et al., 1999; Rajendran et al., 2014; Serrano & Knud, 2011) (Table A1). The justification and validation for the reactions is provided in the stated literature. The remaining major metabolic pathways, which are acetogenesis, acidogenesis, methanogenesis, amino acid degradation reactions and hydrogen pathways (Dark Fermentation) were simulated using Continuously Stirred Tank Reactors (CSTR). CSTR units are kinetic reactors and as such reaction kinetic information was provided, based on the first order reaction kinetics (I. Angelidaki et al., 1993, 1999; Rajendran et al., 2014). The first-order kinetic framework allowed for simplicity and compatibility with Aspen Plus. Furthermore, due to the limited substrate-specific data, a major gap in the current literature, identical or similar rate constants were used in some cases as described in the cited literature. Amino acid degradation reactions were added separately to allow for clearer tracking of nitrogen transformations, but microbial biomass was not explicitly included in the reactions to avoid over-parameterisation, in line with the mentioned literature. The hydrolysis reaction set stated in this work may not be exhaustive, and more re-

actions can be added to enhance the model's capability to specific substrates. Hydrolysis reactions and subsequent pathways depend on the specific substrate composition, captured in the form stated above. Therefore, the hydrolysis reactions stated are not simultaneously active and depend on the particular composition of the input substrate and adding or removing hydrolysis reactions may not impact the model's accuracy. Two CSTRs were used to simulate hydrogen pathways in the first reactor and methanogenesis in the second reactor. 42 reactions were used to represent the major metabolic pathways (Table A2). A mixer was used for the initial mixing of the input substrates (to create homogeneity) prior to the hydrolysis reactor, and another for mixing the recirculate (which was simulated at 30%) with the liquid product from the dark fermentation reactor. Recirculation was only applied to the methanogenesis reactor. A heat exchanger (pre-heater) was added before each unit to ensure that the input material was at the stipulated temperature. A phase separator separated the produced gas from the aqueous phases. All units were modeled at atmospheric pressure. The sequential flow of material started with the initial mixing of input materials in a mixer, and the homogeneous mixture was sent to a stoichiometric reactor (hydrolysis), followed by reaction rate calculations, before passing to kinetic reactors CSTR(1) (dark fermentation) and toward CSTR(2) (methane production). In the case of recirculation, the digestate from CSTR(2) was sent to a splitter, and the recirculate mixed with the liquid product from CSTR(1). Aspen performed the mass and energy transfer convergence calculations.

To capture the non-linearity of the process, a more dynamic approach was followed to model the kinetics rather than fixed-rate constants. The power law kinetics incorporated process inhibition and environmental dependency terms. Inhibition terms included substrate concentration, acetates, butyrate, Long Chain Fatty Acids (LCFA), pH, ammonia, hydrogen partial pressure, and temperature (modelled using Arrhenius equation). At each simulation step, the power law kinetic reactions were calculated, depending on the input species and conditions, which introduced non-linearity into the system, as the reaction rate changed in response to the varying concentrations and conditions. These factors were applied to the kinetic parameters for acidogenic, acetogenic, amino acid degradation, methanogenic and dark fermentation reactions. The inhibition factors were calculated and applied to the reaction rate (K) before running the CSTRs, where the major metabolic pathways were computed.

Complex organic matter (input components) underwent hydrolysis according to the extent of reaction in Table A1, and the products in the liquid phase were directed to the CSTRs where reactions in Table A2 were implemented. Materials from hydrolysis are sent to specific Calculator blocks (Aspen Flowsheeting tool), which performed reaction rate calculations and accounted for inhibition. These reaction rates were then exported to the CSTRs. Reaction rates were calculated for acidogenesis, acetogenesis, methanogenesis, dark fermentation, glycerol, valeric acid, butyric acid, propionic acid, linoleic acid, amino acid, palmitic acid, and oleic acid according to the products from

hydrolysis reactions. The calculator blocks imported substrate flow rates, VFAs, ammonia flow, and temperature, from the hydrolysis product stream. The Arrhenius equation was used to calculate the effect of temperature, and pH was calculated based on the chemical equilibrium constants for the VFAs according to the work of (I. Angelidaki et al., 1999).

Despite the efforts to simulate the non-linear behavior of the anaerobic digestion process, by incorporating dynamic reaction rate modelling, several limitations persist, which must be noted when expanding the results beyond the current scope. The simulation work was done based on the oversimplification of the ADM1 model, to make it suitable for the ASPEN platform. Whereas, it is suitable for the Aspen framework, using fixed reaction kinetic data introduces a limitation to the modeling of complex microbial community interactions. A more linear output may be observed, as a result, particularly when operating at low concentrations, and isothermal conditions, where the effects of inhibition are not pronounced. Furthermore, the temperature effects were incorporated using the Arrhenius equation to influence the reaction rate constant, which is an oversimplification of the enzymatic, microbial shifts that may occur from temperature changes. Lastly, thermodynamic models such as Henry's law used for gas-liquid interactions in Aspen are not designed for biological systems. As such, to address the solubility limitations, a conservative approach was used for carbon dioxide accounting, and all carbon dioxide in both the gaseous and liquid streams was counted for to overestimate the gas in biogas.

2.2 Substrate characterisation and model validation

The characterisation information used as input for the simulation is presented in Table A3. The information was gathered from literature. The data was used for both the simulation runs and validation against the experimental data. Carbohydrates, lipids and proteins were validated against the experimental results of Gu et al, 2024, which studied the effect of the aforementioned on hydrogen and methane production. Hydrogen and methane yields were compared with the experimental results based on the starting composition and operating conditions. Hejnfelt and Angelidaki, (2009) and Budiyo et al (2011), and Al Rubaye (2019) provided sufficient compositional data for their slaughterhouse and manure substrates to be mimicked under simulation conditions, with clear results for comparison.

2.3 Energy potential for biogas

Energy recovery potential (E) for biogas was calculated as follows:

$$\text{Theoretical Energy Potential} = [\text{Vmethane (m}^3/\text{day)} * \text{HHVmethane}] + [\text{Hydrogen (m}^3/\text{day)} * \text{HHVhydrogen}] \quad (1)$$

$$\text{Practical/usable Energy Output} = [\text{Vmethane (m}^3/\text{day)} * \text{LHVmethane}] + [\text{Hydrogen (m}^3/\text{day)} * \text{LHVhydrogen}] \quad (2)$$

$$\text{HHVmethane} = 39,8 \text{ MJ/m}^3$$

$$\text{LHVmethane} = 35,8 \text{ MJ/m}^3$$

$$\text{HHVhydrogen} = 12,7 \text{ MJ/m}^3$$

$$\text{LHVhydrogen} = 10,8 \text{ MJ/m}^3$$

3. RESULTS AND DISCUSSION

3.1 Model Validation

A model validation was conducted from various experimental results. In a study assessing methane potential from pig slaughterhouse waste, Hejnfelt and Angelidaki (2009) conducted thermophilic tests on substrates with a composition of 22% carbohydrates, 11% proteins and 55% lipids at a pH of 7,5 in a 26-day retention time. A methane yield of 369 l/kg feed was obtained. The same composition in the model yielded 374 l/kg CH₄ which was a 1,35% difference. In an experimental result presented by Al-Rubaye et al, (2019) for a cattle manure feed of 0,33 l/day with a retention time of 15 days, a biogas composition of 49,89% methane was reported, and this compared well with the simulation which obtained 48,76% methane and 51,10% carbon dioxide. The feed composition was 0,083 glycine, 0,204 cellulose, 0,086 hemicellulose, 0,257 glucose, 0,015 triolein, and 0,355 inerts. An experimental study by Budiyo et al. (2011) on the potential for cattle manure with a characterisation of 70% carbohydrates, 8% proteins, 2% fat and 20% ash, achieved a biogas composition of 48,89% carbon dioxide and 47,87% methane. A slight difference was found with the simulation which read 52,50% carbon dioxide and 47,30% of methane. Hydrogen simulation was tested against Gu et al. (2024) who conducted batch experimental work under mesophilic conditions with a feedstock of carbohydrates, proteins, lipids, and cellulose rich substrates. In the study, dark fermentation was conducted for 52 hrs and the digestate anaerobically digested to a further 40 days. The results showed that carbohydrates rich substrates yielded the highest cumulative hydrogen amount, owing to the faster degradation and vulnerability to microbial attack compared to proteins and lipids. Gu et al. (2024) further associated this with the rather straightforward structure of carbohydrates, a high moisture content, and high levels of cellulose and hemicellulose exposed to pretreatment. Carbohydrates hydrogen yield was 47,90% and 38,70% higher than that of the protein and lipids, respectively. For methane production, the study obtained about the same yield with only a slight advantage for lipid rich (60% lipids), followed by carbohydrates rich (60% carbohydrates) and protein rich (60% protein) substrates. In the simulation, hydrogen production was highest in protein rich, followed by carbohydrates and lipids. Methane production was in the order of carbohydrates rich, protein rich and lipids rich, respectively. An observation of the results from the publication points to the possibility of process inhibition of proteins and lipids during the dark fermentation test which caused such low yields. Furthermore, these substrates are known to degrade over an extended period, with long lag phases. In fact, Hejnfelt and Angelidaki (2009) realised a lag phase of 20 days when anaerobically digesting pork fat.

A test was done to observe the model's response to temperature. To this cause, cattle manure substrate, characterised above (Al-Rubaye et al., 2019), was used under mesophilic (35°C), thermophilic (55°C) and hyperthermophilic (75°C) conditions at a feed rate of 1 l/ day and total solids of 6,62 g/l with a 5- and 20-day retention time for DF and AD respectively. With both DF and AD operated

under same conditions, the highest yield was obtained for hyperthermophilic, followed by thermophilic and mesophilic. Hyperthermophilic yield for hydrogen, methane and carbon dioxide, respectively was found to be 0,918 m³/day, 0,170 m³/day, and 0,243 m³/day for DF, and <0,001 m³/day, 0,759 m³/day and 0,844 m³/day for AD. Thermophilic yield were 7,52%, 7,74% and 5,79% lower for hydrogen, methane and carbon dioxide, whereas for mesophilic runs the yield dropped by 11,50%, 12,20% and 11,80% respectively. The average composition for DF was 69.00 +/- 0.14% hydrogen, and 12.00 +/- 0.037% methane, whereas for AD the average was 47,45 +/- 0.085% methane and no hydrogen. The results were typical with increased kinetic performance at higher temperature. Whereas the results present a more direct increase in kinetic performance, and thus higher bacterial growth; microbial activity and COD removal optimality are complex and rely on many conditions combined, which is why AD results are inconsistent and difficult to standardise. Therefore, studies such as Manogram et al. (2023) observed an 8-fold increase in yield from psychrophilic to mesophilic co-digestion of chicken manure and fruit bunch, but a 3-fold reduction when the operating conditions were set to thermophilic. Hejnfelt and Angelidaki (2009) achieved methane yields at 8 times lower for thermophilic as compared to mesophilic runs during the digestion of slaughterhouse waste, at equal feeding rates in a CSTR. In fact, an attempt to increase the load resulted in reactor failure for the thermophilic test. The composition for methane was 72,50 +/- 1,50%. In the same study, feedstock with 20% and 5% mixed pork waste yielded the same methane yield and composition under thermophilic tests, whereas under mesophilic tests, at 5% pork waste, the yield increased by 138 l/kg.

High protein and lipid substrates are more prone to inhibition due to ammonia concentration which increases with temperature and organic load (Ghimire et al., 2015). Angelidaki and Ahring (1999) thus recommended operating high nitrogen bearing substrates at a lower pH and high dilution. This improves the C/N ratio and maintains ammonia in its liquid phase as ammonium instead of the gaseous form, which is toxic to methanogens. In Dawei, (2008), it was highlighted that at extreme thermophilic conditions, hydrogen yield can reach the maximum theoretical yield of 4 mol of hydrogen per mole of glucose, and the bacteria shows better tolerance for high hydrogen partial pressures. In the study, better handling was achieved through enriching bacteria culture. The same result was reached by Okonkwo (2020) where mixed cultures improved resilience of the process to short- and long-term temperature fluctuations, improving resilience to thermal failures. As mentioned before, pH inhibition was accounted for in the model, however, as a limitation, the correlation used yielded the standard highest yield at pH of 6,5. Increasing and decreasing the pH produced a similar effect in terms of pH inhibition figures to the reaction kinetics. As such, using the same manure feed, the same results were observed for pH values of 6,0, and 7,0 as well as 5,5 and 7,5. The results showed a reduction of 3,4% methane yield when pH is changed by 0,5 and 15,4% at a pH change of 10 units. Thus, in this study, all reactions were performed at pH 6,5 to remove the effect.

3.2 Carbohydrates, Proteins and Lipids

As mentioned before, carbohydrates, lipids, and proteins are core components for anaerobic digestion, which can be used for accurate modeling of the process. Hence this section is dedicated to understanding the combined effect of these components on hydrogen and methane yields. Optimising these substrates can provide reliable information on the effect of co-digestion substrates on process outcome. To allow for proper running of the model, all three components were included in all the runs, and the main component accounted for 80% total solids, whereas the other two constituted 10% each. DF was run at a thermophilic temperature, and the second stage was mesophilic. High temperatures favour hydrogen producing bacteria and heat shock is often applied to restrict methanogens (Dell'Orto, 2017).

A sensitivity test was conducted on Aspen Plus at a feed rate of 0,1-1 l/d for each of the substrates. Figures 1-3 show process outcome as total volume produced from both stages. The plots show the effect of codigesting carbohydrates and lipids at a constant protein rate of 0,5 l/day. Increasing or decreasing the protein content, shifts the graphs upwards or downwards uniformly with the least maximum achievable yield of hydrogen at 936,374 l/d and the highest maximum at 1087,63 l/day corresponding to 0 l/d and 1 l/d of proteins respectively. Keeping lipids constant showed a much higher effect on the achievable hydrogen yield with only 398,992 l/day produced at 0 l/day lipids. The interaction of proteins and lipids yielded a maximum of 832,524 l/day at 0 l/day of carbohydrates. This shows that proteins had the least effect of the three. For methane production, codigestion of carbohydrates and lipids only, resulted in the maximum methane yield of 433,165 l/day, whereas carbohydrates and proteins only, achieved a yield of 451,034 l/day. In the absence of carbohydrates, the highest methane yield was 675,529 l/day.

The discussion above is emphasised in the perturbation plots which presents the effect of a point change of one component using a line graph, while keeping the other two constant. There is a linear relationship between the components load and yield of hydrogen, methane and carbon dioxide. Lipids had the highest effect on hydrogen yield followed by carbohydrates and proteins respectively. As mentioned, this is unlike the findings of Gu et al, (2024), where carbohydrates achieved the highest yield, and lipids appeared to be inhibited. However, similar results were obtained for methane yield, where proteins and lipids showed a high and comparable response, and carbohydrates had the least effect. A reason for the disparity in hydrogen production could be that in each run Gu et al (2024) was feeding 40% of carbohydrates for protein and lipids rich substrates, and 60% carbohydrates when studying its effect as the main component, compared to 20% when studying the other two. Whereas in this study, the main constituent was 80% and the others 10%. The faster biodegradability of carbohydrates allowed it to be utilised in the first stage, for a retention time of 2 days compared to the slower degrading lipids and proteins. Whereas, the lipids and proteins have a higher yield potential, the biodegradability of carbohydrates is the highest

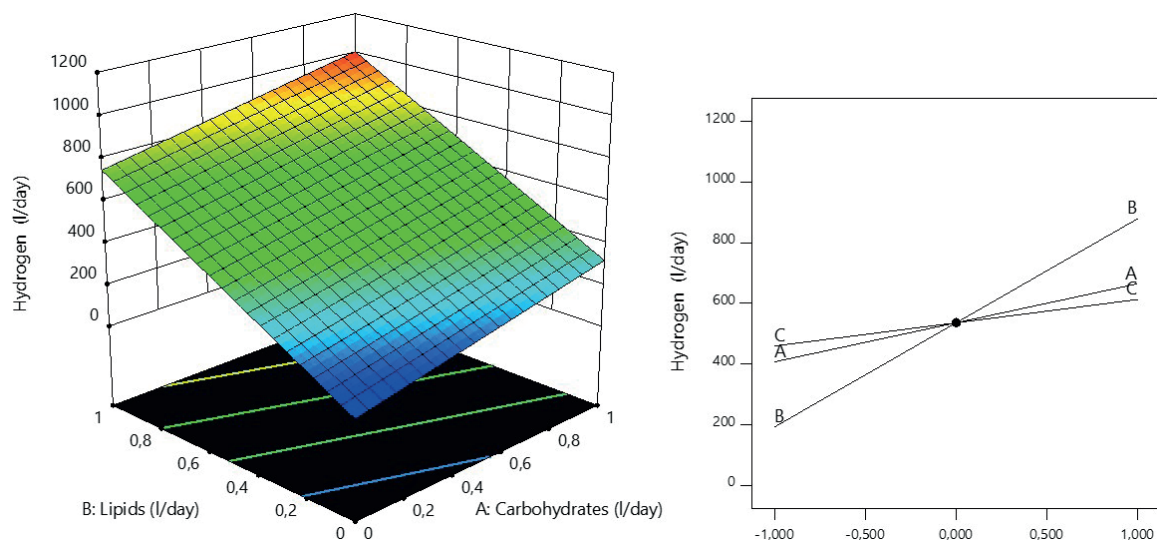


FIGURE 1: The effect of carbohydrates, lipids and proteins on hydrogen yield. A = Carbohydrates; B = Lipids, C = Proteins.

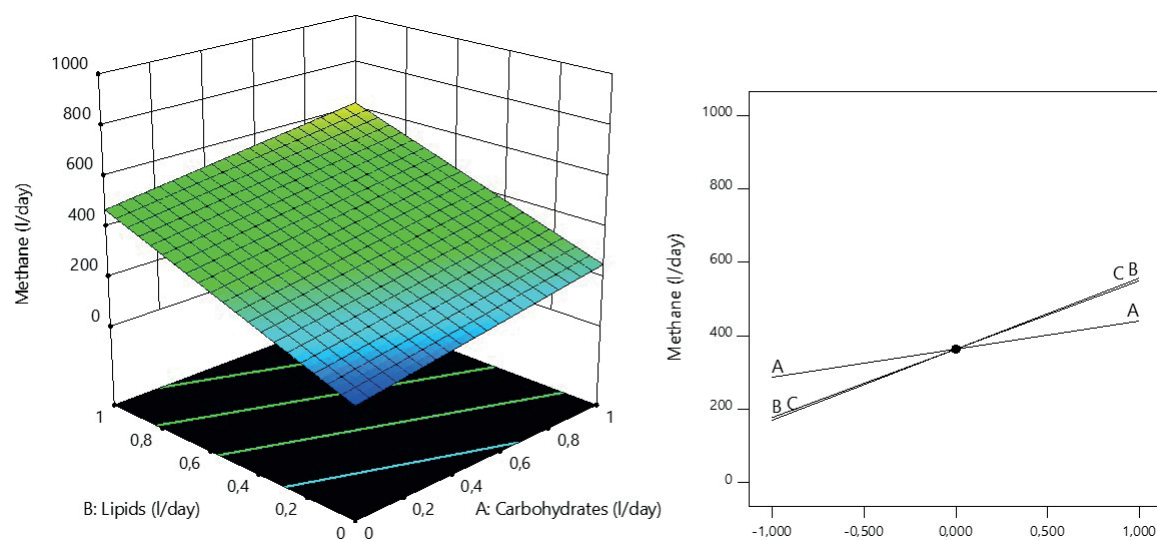


FIGURE 2: The effect of carbohydrates, lipids and proteins on methane yield. A = Carbohydrates; B = Lipids, C = Proteins.

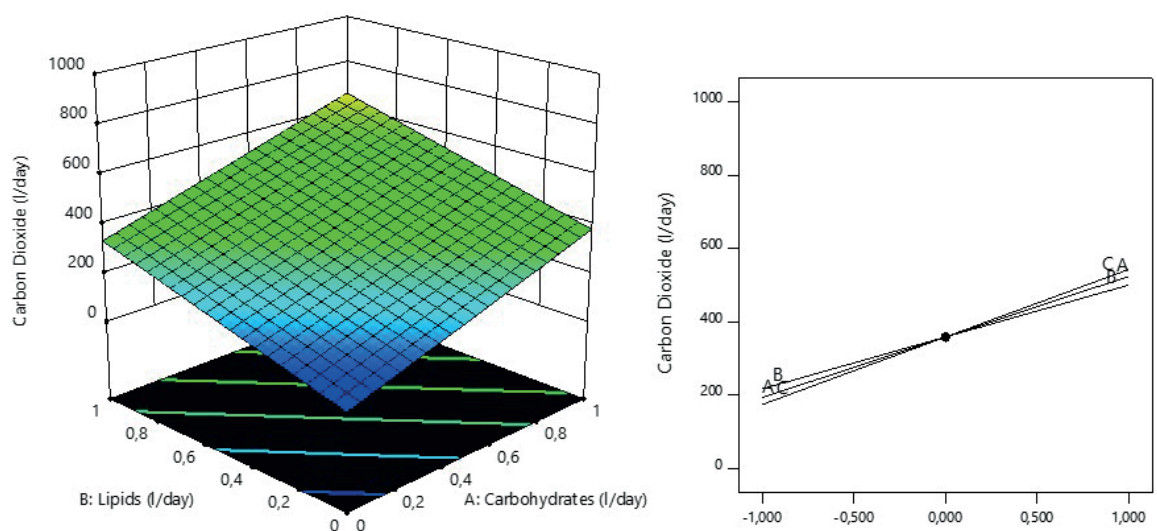


FIGURE 3: The effect of carbohydrates, lipids and proteins on carbon dioxide yield. A = Carbohydrates; B = Lipids, C = Proteins.

and this is significant in experimental tests (Alhraishawi & Aslan, 2022). The biogas potential of the three substrates is 990-1452 ml/g for lipids, 480-630 mg/l for proteins, and 373-921 for carbohydrates (Alhraishawi & Aslan, 2022). The theoretical yield of methane is in the order 0,99 m³ CH₄/kg, 0,63 m³ CH₄/kg, and 0,42 m³ CH₄/kg for lipids, proteins and carbohydrates, respectively (Alves et al., 2009).

To further this, process inhibition due to ammonia and volatile fatty acids accumulation, biomass blockages, foaming and floatation from nitrogen rich substrates such as proteins and lipids is very common in lipid and protein digestion (Alhraishawi & Aslan, 2022; Breure et al., 1986; Ortnet et al., 2014). Hejnfelt and Angelidaki (2009) observed

foaming on digestion of lipids rich substrates especially in CSTRs, and this was attributed to the degradation of Low Chain Fatty Acids (LCFA), which decreased the bio-accessibility of the LCFA and other particles. In the studied range, for this work, ammonia production was at 3,1 +/- 0,1% of the biogas for proteins and lipids. Ammonia production was in the order of protein>lipids>carbohydrates. Hydrogen sulphide production was also in the same order (Table 1). Codigestion at 1 l/day for each substrate reduced the amount of ammonia by 23%, and hydrogen sulphide by 5,8%. The digestate observed an increase in acetate by 55%, whereas ethanol, propionate and butyrate were reduced in volume (Table 2).

TABLE 1: Biogas and energy recovery potential for various abattoir substrates Feed = 1 l/day.

Substrate	Biogas (l/day)					Theoretical Energy	Practical Energy
	CH ₄	H ₂	CO ₂	Ammonia	Hydrogen sulphide	kWh/day	kWh/day
Protein (P) (no recycle)	1523,34	2243,96	2986,98	215,15	71,92	24,76	21,89
Lipids (L) (no recycle)	1522,95	2249,87	2989,05	215,12	71,92	24,78	21,9
Carbohydrates (C) (no recycle)	1548,07	2253,26	3001,51	214,23	71,92	25,07	22,16
P: L: C At 1 l/day each	1804,03	1777,46	2948,07	166,14	67,68	26,22	23,28
Cattle rumen	933,13	882,43	975,25	192,70	58,11	13,44	11,93
Goat rumen	1161,86	1405,95	1519,04	180,77	52,41	17,81	15,78
Cattle Blood	955,19	836,81	964,74	207,97	63,36	13,52	12,02
Cattle muscle	975,85	829,24	982,98	216,40	66,59	13,72	12,2
Cattle Manure	826,57	838,39	964,10	165,73	30,57	12,1	10,74
Confiscates (liver, lung, kidneys)	860,10	864,29	859,94	148,30	44,66	12,56	11,15
Pig tissue + fat	984,77	1188,76	971,52	111,16	33,74	15,09	13,37
Pig stomach	832,75	934,08	840,13	123,32	36,65	12,51	11,09
Chicken Feathers	1106,38	817,29	1119,39	286,76	88,32	15,12	13,46
Poultry wastewater	845,26	920,44	847,89	128,43	37,78	12,60	11,17

TABLE 2: Digestate stream for various abattoir substrates Feed = 1 l/day.

Substrate	Acetate	Propionate	Butyrate	Ethanol	Keratin	Protein	Cyano-acetate
l/day							
Protein (P) (no recycle)	0,4018	0,1425	1,099	1,314	0,1098	0,1093	0,1117
Lipids (L) (no recycle)	0,4020	0,1425	1,098	1,314	0,1098	0,1093	0,1118
Carbohydrates (C) (no recycle)	0,4182	0,1425	1,052	1,314	0,1098	0,1093	0,1157
P:L:C At 1 l/day each	0,6205	0,0847	0,2682	1,243	-	0,1053	0,1592
Cattle rumen	0,2287	0,0824	0,0537	0,0649	0,1358	0,0872	0,0556
Cattle Blood	0,2197	0,0800	0,0242	0,0389	0,1321	0,0955	0,0541
Cattle muscle	0,2199	0,0788	0,0156	0,0389	0,1268	0,1006	0,0542
Confiscates (liver, lung, kidneys)	0,2279	0,0819	3,360E-05	0,0389	0,1205	0,0667	0,0578
Pig tissue + fat	0,2930	0,1158	0,0097	0,0389	0,1218	0,0500	0,1218
Pig stomach	0,2236	0,0893	0,03424	0,0389	0,1180	0,0543	0,1180
Goat rumen	0,3166	0,1200	0,2784	0,3509	0,1867	0,0772	0,0837
Chicken feathers	0,2276	0,078947	0,0254	0,03892	0,1476	0,1340	0,0527
Poultry wastewater	0,2240	0,088596	0,0185	0,0389	0,1261	0,0558	0,0601
Cattle manure	0,2143	0,076712	0,0473	0,1725	0,1098	0,0450	0,0541

Past research has proven that such inhibition can be overturned and adsorption by materials such as biochar is being studied. Biochar can also be instrumental in reducing the carbon content of the process. Whereas Angelidaki and Ahring (1999) suggested co-digesting protein and lipids rich substrates at high dilution and mesophilic temperatures, Alves et al, (2009) noted that appropriate equipment and feeding scheme can improve the feasibility of working with this feedstock. The study proposed a specialised reactor system with a primary biomass retention through floatation and a secondary biomass retention through settling. Neves et al, (2009) suggested progressive addition of lipids rich substrates while staying below the threshold values for Low Chain Fatty Acids (LCFA). Breure et al (1986), recommended spatial separation during the hydrolysis of high protein concentrations after discovering that glucose adapted mixed bacteria turned to struggle degrading proteins. Separation of hydrolysis and fermentation was applied in this work. Lastly, carbon dioxide generation was found to follow a more linear trend to a point change in substrates load and there is minimum difference in cause and effect of the three components.

Following the study of the interaction of the components to the yield, three optimisation criteria were set. First with the aim to maximise hydrogen yield, second one maximising methane, and the third one minimising carbon dioxide. It was found out that at an equal feed rate of 1 l/day, the highest hydrogen yield of 1084 l/day was produced, with 875 l/day of methane and 937 l/day of carbon dioxide. This also served as the highest achievable methane production in the studied range. Carbohydrates were found to generate a very high amount of carbon dioxide particularly in the dark fermentation stage, with lipids generating the least. As such minimising carbohydrates and increasing lipids was the best way to obtain low yields of carbon dioxide while maintaining high yields of hydrogen and methane. The least carbon dioxide was measured at 0 l/day carbohydrates, 1l/day lipids and 0,520 l/day proteins where 318,74 l/day carbon dioxide was produced with 753,73 l/day hydrogen, 464,371 l/day methane. Based on the simulation, a linear model was developed with an adjusted and predicted $R^2 > 0,99$ and sequential and lack of fit p-values $< 0,0001$. Equations 3-5 shows the correlation for estimating the biogas yield at varying feed rates of the three components.

$$\text{Hydrogen l/day} = 257,349*A + 686,277*B + 153,356*C - 12,303 \quad (3)$$

$$\text{Methane l/day} = 153,120*A + 372,375*B + 386,567*C - 92,699 \quad (4)$$

$$\text{Carbon dioxide l/day} = 330,038*A + 282,587*B + 367,772*C - 130,901 \quad (5)$$

where:

A = Carbohydrates (l/day)

B = Lipids (l/day)

C = Proteins (l/day)

3.3 Co-digestion of manure, animal tissue and blood

Manure, animal tissue and blood forms some of the highest wastage at abattoir facilities. As such, these of-

fers the potential for viability in conversion to biogas and a great benefit for energy production. Such a waste stream is available in the long term and can be useful in sizing an anaerobic plant, with a guarantee for a consistent feed composition, a gap that exist in the biogas industry and a major cause of failure for biodigesters. Animal blood and tissue are rich in proteins and low in lipids and carbohydrates (Table A3), whereas manure has a slightly higher carbohydrate. In an experimental setup, manure offers a microbial community and a buffering capacity which can be useful for maintaining pH. The following section details the results from the co-digestion of manure, animal tissue and blood.

3.3.1 Substrate impact on biogas production

The impact of substrate composition was investigated over the range of 0-1 l/day of each of animal manure, blood, and tissue. The study compares the effect of introducing recycling in the second reactor with a 30% recycling rate. As such recycling affected methane and carbon dioxide yields only. This was because the first reactor which in a typical 2-stage arrangement acts as an acidification reactor, is expected to be smaller and take much lesser loads. The first reactor was simulated at thermophilic temperature, whereas the second reactor was operated under mesophilic conditions. The reason for this is because there is favorable hydrogen production kinetics, and the first reactor may be incorporated as a pretreatment stage due to the hazardous nature of the abattoir waste stream, which is often required to be pasteurised prior to use. Both were operated at 1 atm. Figures 4-6 show the effect of Manure and blood at constant tissue of 0,5 l/day without recycling (upper image) and with recycling (lower image). Increasing or decreasing the amount of tissue shifted the plot up or down respectively. The highest possible output for hydrogen at 0 l/day of tissue was 85 l/day. The highest yield dropped down to 70 L/day when animal blood was set to zero and up to 153 L/day when excluding animal manure. The results shows that animal manure had a negative effect on hydrogen yield, and blood has the highest impact. The perturbation plot reflects this observation showing the linear slope of blood having the highest response to a unit change, followed by tissue and animal manure.

At a feed of 1 l/day for each substrate, the maximum achieved methane was 643,089 l/day. The highest possible output of methane at 0 l/day tissue was 550 l/day, and the highest possible at 0/day of blood was 500 l/day. Manure showed the highest impact on methane production with only 260 l/day achievable at 0 l/day manure. This is shown in clearer detail in the perturbation plots where A has a much steeper slope compared to B and C. Animal tissue had the least influence on methane yield. Following the three criteria for optimisation, the highest yield of methane was observed at an equal feed of 1 l/day of each substrate, producing 649,04 l/day methane, 148,74 l/day hydrogen, and 898,53 l/day of carbon dioxide. The effort to maximise hydrogen while maintaining a high methane production was observed at the same feed rate. An attempt at minimising carbon dioxide, resulted in the reduction in

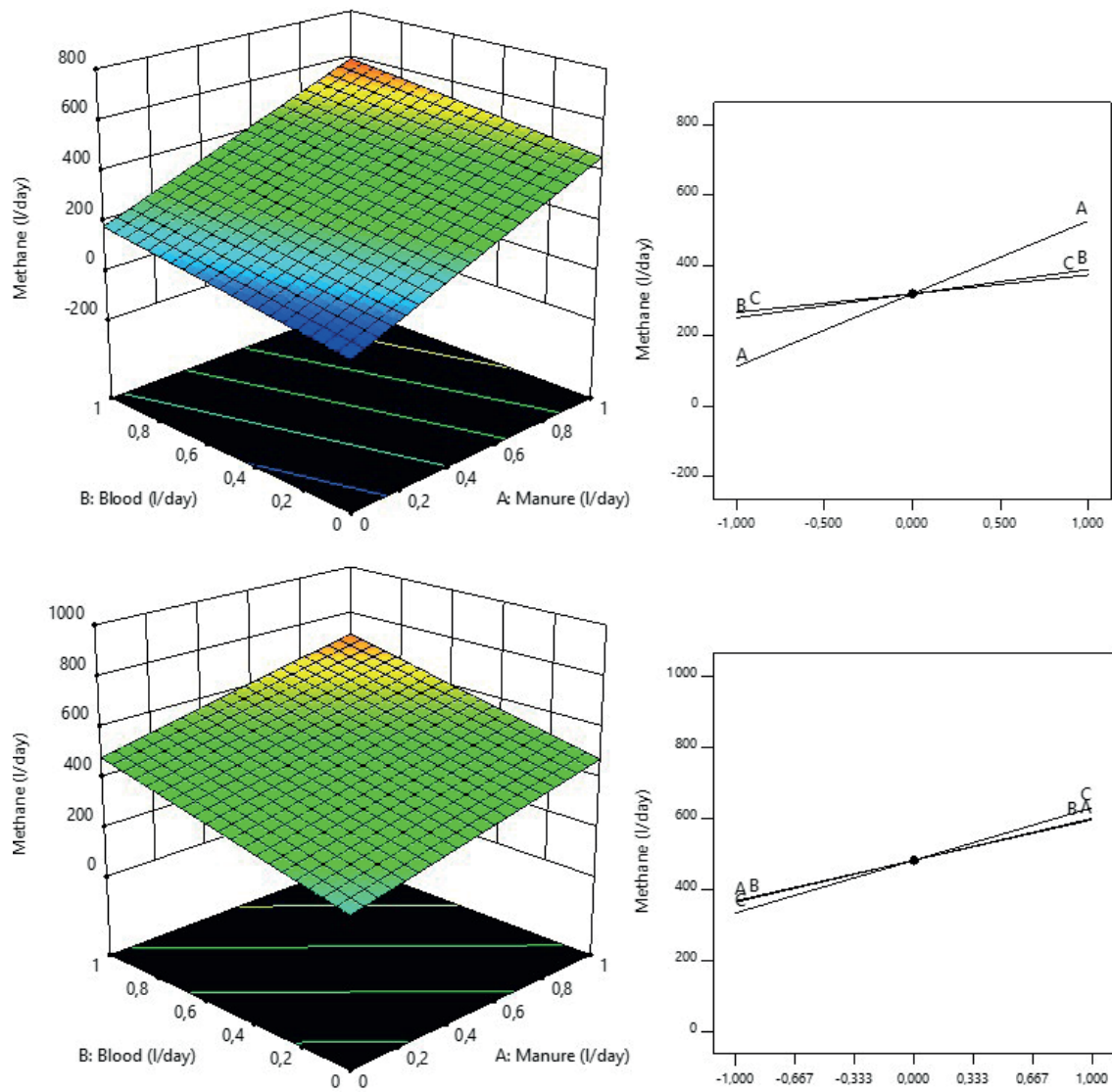


FIGURE 4: The effect of manure, blood and tissue on methane yield. upper image = without recycling, lower image = with 30% recycling. A = Manure, B = Blood, C = Tissue.

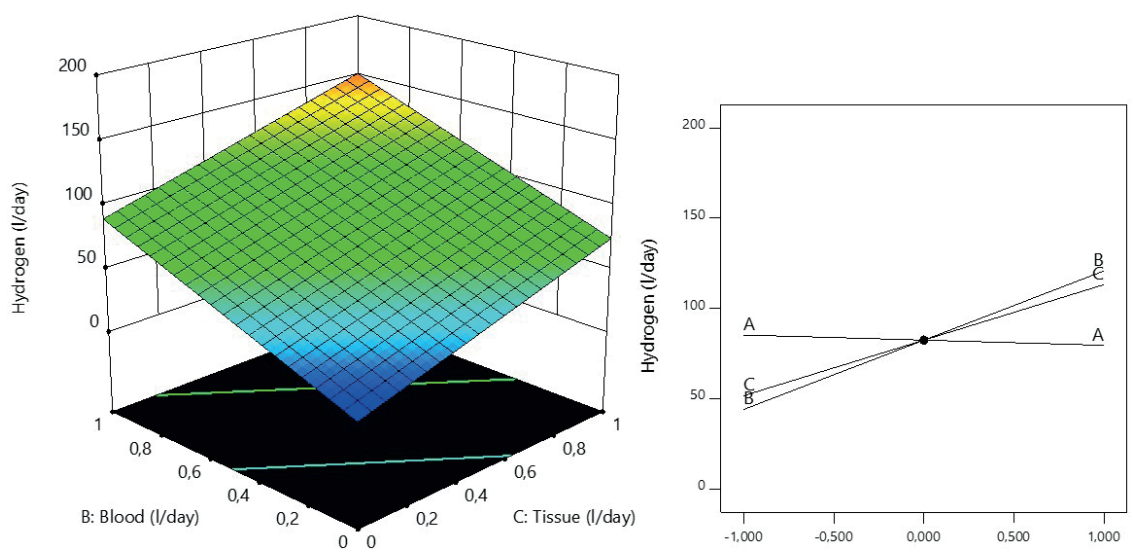


FIGURE 5: The effect of manure, blood and tissue on hydrogen yield. A = Manure, B = Blood, C = Tissue.

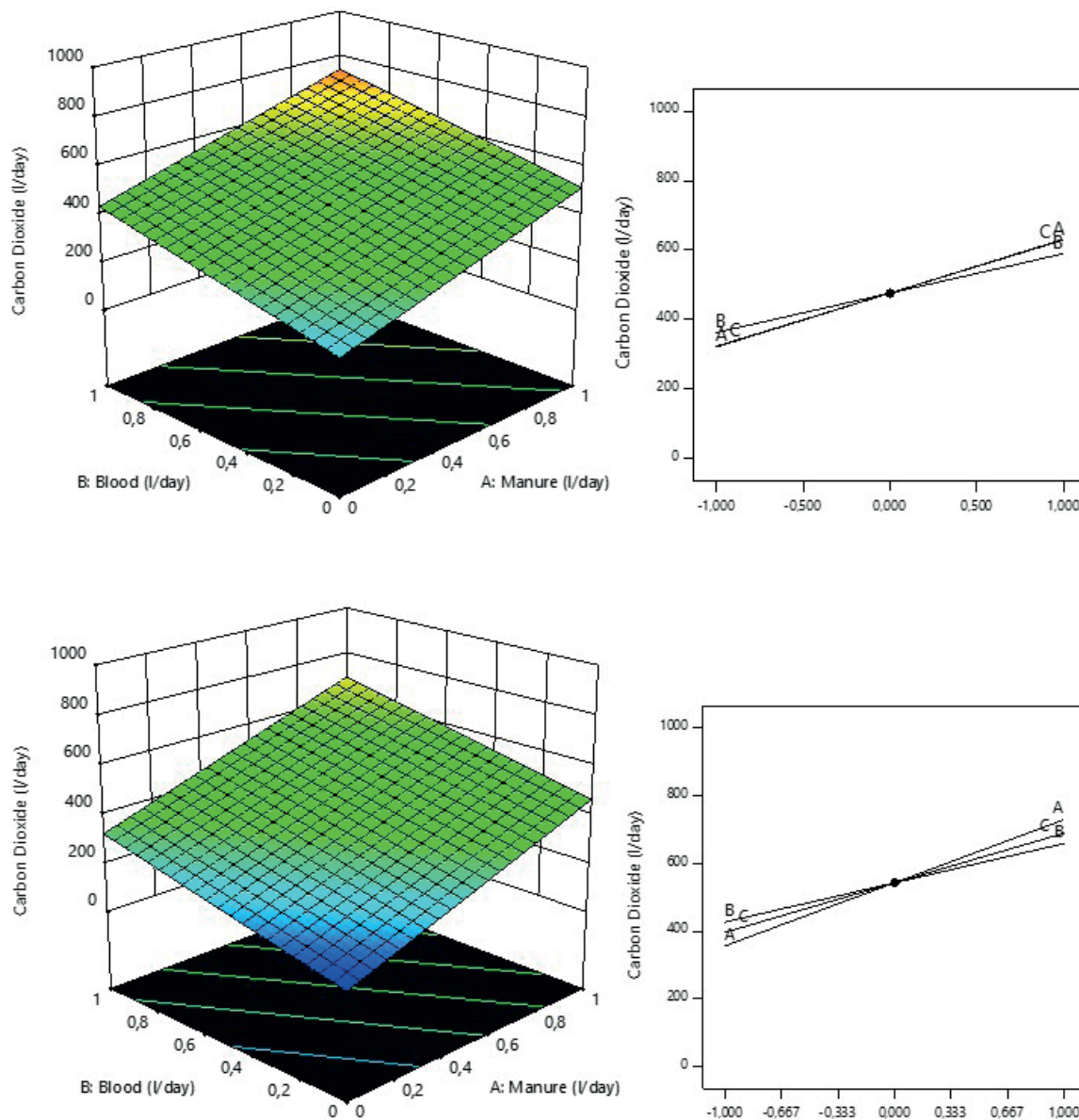


FIGURE 6: The effect of manure, blood and tissue on carbon dioxide yield. upper image = without recycling, lower image = with 30% recycling. A = Manure, B = Blood, C = Tissue.

the yield of methane and hydrogen. The optimum feed for the criteria was 0,73 l/day manure, 1 l/day blood, and 0,088 l/day tissue, which yielded 438,48 l/day methane, 94,25 l/day hydrogen, and 531,55 l/day carbon dioxide. Based on the results presented, it is clearly visible that manure may not be the best co-digestion substrate for hydrogen production, but it is good for methane production as reflected in equation 6-8 below and various experiments (Gaogane, 2021). As such in a 2-stage process, it is best to introduce the substrate in the second stage.

$$\text{Hydrogen l/day} = 76,756*B + 61,496*C - 5,701*A + 16,190 \quad (6)$$

$$\text{Methane l/day} = 414,221*A + 137,294*B + 106,092*C - 8,568 \quad (7)$$

$$\text{Carbon dioxide l/day} = 306,433*A + 227,074*B + 310,181*C + 54,842 \quad (8)$$

where:

A = Manure (l/day)

B = Blood (l/day)

C = Tissue (l/day)

There were observable changes in substrate impact on yield when recirculation was introduced with recycling a small portion of the digestate to the second reactor. The results for recycling showed the maximum yield of methane at 1 l/day of each substrate as 860,83 l/day. At 0 l/day tissue, the maximum yield was 564 l/day, which went up to 625 l/day in the absence of animal blood. The effect of manure was much closer to that of animal blood, as the highest methane yield was predicted at 632 L/day at 0 l/day of manure. This is presented in the perturbation plot with the highest effect on yield observed for animal tissue. The carbon dioxide production rate was mostly influenced

by the feed rate of animal manure, followed by animal blood and tissue. At zero tissue, carbon dioxide produced was 699 l/day, and at no blood addition, the maximum produced was 760 l/day, whereas in the removal of animal manure the yield was only 619 l/day. Under optimisation with basis for methane yield, the maximum amount of methane was found at a feed rate of 0,993 manure, 0,995 blood, and 0,987 tissue, with a production of 854,25 l/day methane, and 984,48 l/day carbon dioxide. This was a 32% increment in the maximum achievable methane value at 30% recycling compared to non-recycling conditions. The best effort to minimise carbon dioxide while maintaining the highest possible methane production was defined at 0 manure, 0,999 blood, and 0,480 tissue. Biogas yield had a significantly lowered methane yield of 478,46 L/day and 466,27 L/day carbon dioxide. The corresponding coefficients for estimating biogas yield in the case of recycling are presented below:

$$\text{Methane l/day} = 228,187*A + 235,487*B + 296,114*C + 101,045 \quad (9)$$

$$\text{Carbon dioxide l/day} = 372,965*A + 232,095*B + 293,313*C + 93,596 \quad (10)$$

where:

A = Manure (l/day)

B = Blood (l/day)

C = Tissue (l/day)

3.4 Biogas composition of abattoir waste and potential for energy recovery

Table 1 shows that for a 1 l/day feed of cattle rumen, 933 l/day of methane and 882 l/day of hydrogen can be produced. Goat rumen can produce a higher amount with methane and hydrogen production of 1161 l/day and 1405 l/day respectively. A study on the mono-substrate feed of cattle rumen fluid achieved 3,28 ml/ml rumen fluid at STP using a rumen fluid extracted hydrogen enhancing bacteria culture namely *Staphylococcus* (Maman et al., 2024). The study achieved the highest yield at a pH of 6,5. An advantage of rumen fluid is that it is internal predigested and does not require any pretreatment, and it is also abundant since a single cow generates 90-100 L of rumen per cow, which is often disposed (Maman et al., 2024). Because of the predigested form, the rumen fluid can be used as both an inoculum and substrate.

Cattle blood at the same feed rate can produce 955 l/day of methane and 836 l/day of hydrogen. Hejnfelt and Angelidaki, (2009) achieved 562 l/kg waste from a batch digestion of blood at 5% concentration. Increasing the concentration, reduced the yield significantly on the study. In the study mentioned, batch experiments on mixed pork waste achieved theoretical yields, reaching over 600 l/kg VS methane yield. There was no difference in the yield at mesophilic and thermophilic, showcasing reliability as feedstock. Increasing the feedstock to 50%, the theoretical yield was also reached, but only at mesophilic temperatures. Reaching the theoretical yield means pretreatment is not required to enhance the performance, making pork waste a favourable feedstock. Under CSTR conditions, methane yield was 45% of the theoretical yield at both 5%

and 20% mixed pork concentrations, but mesophilic temperatures elevated the yield to 74% of the theoretical yield at 5% concentration. Pork fat achieved a methane yield of 562 l/kg waste and meat and bones at 10% concentrations achieved a methane yield of 580 l/kg VS. In the current study, pig tissue + fat observed a methane yield of 984,77 l/day, and cattle muscle was lower with 975,85 l/day. Hejnfelt and Angelidaki, (2009) achieved a yield of 550 l/kg VS meat pieces at 20% concentration. A higher methane yield was obtained for cattle muscle compared to internal organs (liver, kidney) but the converse was observed for hydrogen production. The result was in agreement with (Cieciura-Włoch & Borowski, 2019). The difference was that Weronika and Borowski (2019) achieved significantly lower hydrogen yields in both batch experiments. Ammonia concentration was significantly higher in the biogas of cattle muscle, blood and chicken feathers of which according to Hejnfelt and Angelidaki, (2009), digesting them under mesophilic temperatures is a better choice to reduce the total ammonia-N in the free form.

Sawyer et al. (2019) estimated cattle manure production of 136 161 tons per year in South Africa, and potential for recovery of methane and hydrogen gas was found to be 826 l/day and 838 l/day, respectively. Chicken feathers produced a high methane yield of 1106 l/day and 817 l/day of hydrogen. Due to the high organic load of wastewater, a high amount of biogas can be recovered with yields of up to 845 l/day methane, and 920 l/day hydrogen. These figures show a great potential in energy gains from anaerobic digestion of abattoir waste stream in a 2-stage process. The main composition of digestate was acetate, propionate, ethanol, butyrate, cyano-acetate and unreacted proteins. Similar results were observed for digested manure, in comparison to the experiment by Hussien et al. (2024) where the digestate was composed mainly of acetate, followed by propionate and butyrate. A review by Ghimire et al. (2015) and Xiong et al, (2024) showed that these are major digestate constituents regardless of substrates, temperature and pH in various experiments. Using the percentage weight of waste per animal, the total slaughter per year, and energy yield per litre per day, the total energy production in GWh was calculated and presented in Table 3. Cattle rumen and blood showed great potential for energy production with over 2075 GWh/d and 3750 GWh/d energy, respectively, generated from double stage anaerobic digestion. Poultry wastewater based on the figure stated above is over 19,2 million litres and has potential to produce 240 GWh/d.

4. CONCLUSIONS

A simulation model was developed to estimate the outcome of a 2-stage digestion of abattoir waste. The simulation was based on the ADM1 model which uses carbohydrates, lipids and proteins as the main components of anaerobic digestion. The effect of codigestion of cattle manure, blood and tissue was studied on hydrogen and methane production. The study found out that in a codigestion of blood, manure, and tissue, blood had the highest impact on hydrogen yield, whereas manure was the

TABLE 3: Energy potential from abattoir waste stream in South Africa.

Substrate	CH ₄ l/day	H ₂ l/day	Total Energy (MJ/day)	Total Energy (GWh/day)
Cattle rumen	933,13	882,43	7,46E+09	2071,25
Goat rumen	1161,86	1405,95	3,63E+08	100,80
Cattle Blood	955,19	836,81	1,35E+10	3750,64
Cattle muscle	975,85	829,24	1,90E+09	528,66
Cattle Manure	826,57	838,39	5,89E+06	1,64
Confiscates (liver, lung, kidneys)	860,10	864,29	1,12E+10	3099,43
Pig tissue + fat	984,77	1188,76	6,63E+08	184,14
Pig stomach	832,75	934,08	3,66E+08	101,75
Chicken Feathers	1106,38	817,29	1,52E+07	4,22
Poultry wastewater	845,26	920,44	3,40E+12	944116,58

most producer of methane. It was found out that there is so much potential in energy production from the abattoir waste stream for developing countries such as South Africa, which has seen growth in meat demand over the years. A key observation is that the waste stream has potential to generate between 0,068-156,26 GW of energy. It was pointed out that substrates such as animal manure and rumen tissue can play an important role as both inoculum and substrates for methane and hydrogen production respectfully, and they can be digested effectively without pretreatment. As such results like these can form a great benchmark for the abattoir industry for the uptake of 2-stage anaerobic digestion for waste management, energy production, and utilisation.

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PRELIMINARY EXPERIMENTAL STUDY ON GREEN DIESEL PRODUCTION WITH REDUCED IMPACT OF SOLVENT

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ABSTRACT

This study investigates the catalytic deoxygenation (DO) of rapeseed oil for the production of green diesel, focusing on minimizing environmental impact through solvent reduction in accordance with green chemistry and green engineering principles. A series of batch-scale experiments was performed using a novel Ni-Mo-Al catalyst (MOM), synthesized via a new solvent-free method (under patenting procedure) and applied in combination with a process strategy to reduce solvent usage in the DO reaction. Reactions were conducted under varying n-hexane:oil mass ratios (10:1 to 4:1) and oil loadings (2-5 g) to assess the compromise between solvent use and process performance. Key metrics—triglyceride conversion, diesel-range hydrocarbon yield, and product composition—were evaluated by gas chromatography. The optimal condition (4:1 n-hexane:oil mass ratio, 3 g oil loading) resulted in over 99% triglyceride conversion and a 74.2 wt% diesel yield, with more than 90% of the hydrocarbon fraction falling within the C₁₅-C₁₈ range. Product selectivity across all tests favored decarboxylation and decarbonylation over hydrodeoxygenation. Notably, the MOM catalyst showed high activity despite solvent reduction in the DO environment, providing a preliminary yet promising demonstration of how solvent-free catalyst design and solvent minimization can jointly advance more sustainable green diesel production.

1. INTRODUCTION

The Earth's climate system is governed by complex interactions that have been increasingly disrupted by anthropogenic emissions of greenhouse gases (GHGs) (Baede et al., 2022). While climate variability has occurred naturally over geological timescales, human activities—particularly the combustion of fossil fuels, land-use changes, and industrial emissions—have accelerated global warming at an unprecedented rate through elevated GHG concentrations (IPCC, 2023; World Energy Council, 2024). In this context, a rapid and sustained transition toward renewable energy sources and low-carbon technologies are essential to mitigate the most severe climate impacts (IEA, 2024a, IEA, 2024b).

For over a century, fossil fuels have dominated the global energy landscape, with coal, oil, and natural gas providing the majority of primary energy (Energy Institute, 2024). Among these, diesel fuel—derived from crude oil—plays a pivotal role in transportation, powering compression ig-

niton (CI) engines used in light- and heavy-duty vehicles, maritime vessels, railway systems, and stationary industrial machinery (Energy Institute, 2024; Eurostat, 2024a; Eurostat, 2024b). As of 2023, fossil fuels still accounted for approximately 80% of global energy consumption (Energy Institute, 2024), with diesel engines responsible for about 70% of petroleum usage in Europe, equivalent to roughly 245 million tonnes of oil equivalent (Reitz et al., 2019, Eurostat, 2025).

Green diesel—also known as renewable diesel—is a mixture of diesel-compatible n-alkanes derived from fatty biomass, and it has emerged as a particularly promising substitute for fossil-derived diesel (Lucantonio et al., 2023). Unlike conventional biodiesel (fatty acid methyl esters, FAME), which must be blended with fossil diesel due to compositional differences, green diesel consists of straight-chain alkanes that are chemically indistinguishable from those in petroleum diesel (Moser, 2009; Mahdi et al., 2021). This structural compatibility allows green diesel to integrate seamlessly into existing fuel infrastructure



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while potentially reducing lifecycle GHG emissions (Mahdi et al., 2021; Douvartzides et al., 2019; Mahmoudi et al., 2017).

Green diesel is produced through a catalytic deoxygenation (DO) process (Figure 1), in which triglycerides are converted into hydrocarbons via hydrogen-assisted cracking and saturation, yielding products that can be directly used in CI engines.

In the DO process, triglycerides (TGs) are catalytically converted with H_2 to produce linear n-alkanes and various byproducts (Lucantonio et al., 2023; Mahdi et al., 2021; Douvartzides et al., 2019; Di Vito Nolfi et al., 2021). The reaction pathway typically begins with the hydrogenation of TG double bonds, followed by hydrogenolysis, which cleaves ester bonds to form propane (C_3H_8) and saturated free fatty acids (FFAs, $R-COOH$). These FFAs are then converted into alkanes through three competing reaction mechanisms: hydrodeoxygenation (HDO), decarbonylation (DCO), and decarboxylation (DCO_2), illustrated in Equations (1–3) (Lucantonio et al., 2025; Di Vito Nolfi et al., 2021; Nugraha et al., 2024):

- HDO:

$$R-COOH + 3H_2 \rightarrow R-CH_3 + 2H_2O \quad (1)$$
- DCO:

$$R-COOH + H_2 \rightarrow R'-H + CO + H_2O \quad (2)$$
- DCO_2 :

$$R-COOH \rightarrow R'-H + CO_2 \quad (3)$$

Among these, HDO is the most desirable pathway in terms of atom economy, as it retains the full carbon chain of the FFA and produces only water as a byproduct. In contrast, DCO and DCO_2 each lead to a one-carbon loss per molecule, forming carbon monoxide (CO) and carbon dioxide (CO_2), respectively (Lucantonio et al., 2025; Lin et al., 2023a; Lin et al., 2023b; Srifa et al., 2014). When long-chain

fatty acids (C_{16} – C_{24}) undergo deoxygenation, the resulting n-alkanes (C_{15} – C_{18}) closely mimic fossil diesel in terms of cetane number and energy density, ensuring compatibility with conventional diesel engines. Nevertheless, side reactions such as cracking and isomerization may also occur, leading to the formation of shorter-chain or branched iso-alkanes, which can influence cold-flow behavior and combustion properties (Choudhary & Phillips, 2011; Choo et al., 2020).

The DO process typically operates at temperatures between 250–450°C and hydrogen pressures ranging from 1 to 300 bar in the presence of a solid catalyst that promotes C–O bond cleavage and hydrocarbon formation (Naqvi et al., 2023).

Both noble metals (e.g., Pd, Pt, Rh, Ru) and non-noble metals (e.g., Ni, Mo, Zn, Co) have been investigated as catalysts. Noble metals generally offer superior catalytic activity and selectivity, but their high cost limits their scalability. Consequently, industrial DO processes often favor Ni- and Mo-based catalysts, commonly supported on materials such as alumina, silica, titania, zeolites, or activated carbon (Lucantonio et al., 2023; Lucantonio et al. 2025; Upadhyay & Das, 2022; Siraj & Ceylan, 2025).

It is important to emphasize that, from a life cycle assessment (LCA) perspective (IEA, 2024c), green diesel can be considered truly sustainable only if the triglyceride feedstock, the hydrogen source and the energy needed to carry out the system are renewable. In general, green diesel qualifies as a biofuel when it is derived from biological sources—such as vegetable oils, waste cooking oils, animal fats, or algae oils—that effectively recycle atmospheric CO_2 through photosynthesis (Cherwoo & Gupta, 2023; Raman et al., 2025; Asase et al., 2024). Likewise, hydrogen sustainability is a critical factor: only electrolytic hydrogen ($e-H_2$), preferably generated via renewable electricity (e.g., wind or photovoltaic power), ensures a low-carbon footprint (Con-

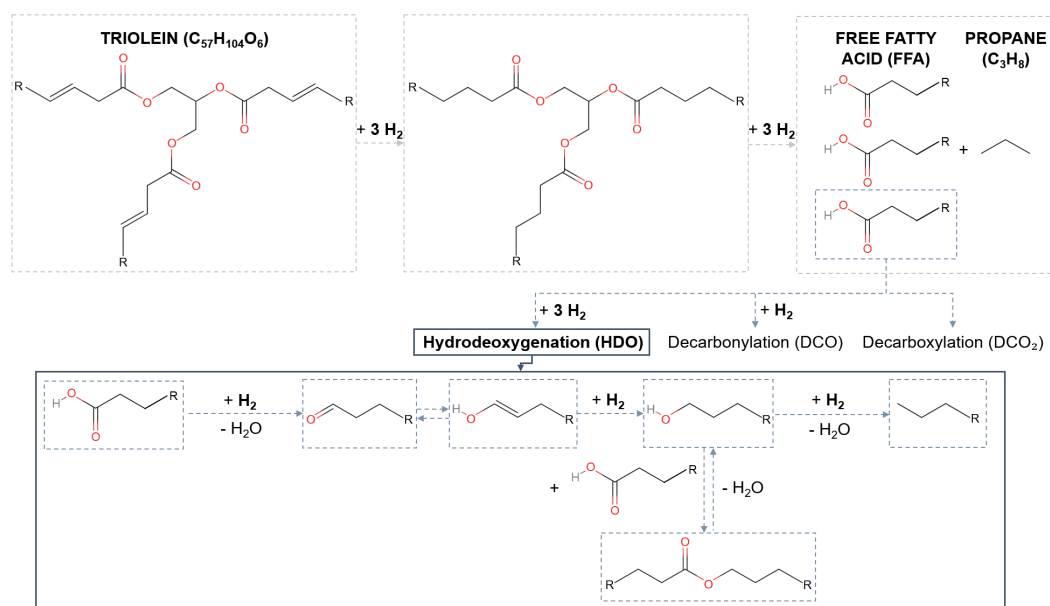


FIGURE 1: Deoxygenation process for green diesel production considering triolein as feedstock, with a focus on the hydrodeoxygenation (HDO) process.

cawe, 2024; Dell'Aversano et al., 2024; Ababneh & Hameed, 2022). Therefore, for green diesel to have a meaningful impact on GHG reduction, it must be produced as an e-biofuel—that is, a fuel made entirely from renewable feedstock and green hydrogen.

From the perspective of LCA (IEA, 2024c), in the light of the 12 principles of Green Chemistry and 12 principles of Green Engineering, the sustainability of green diesel production does not depend only on the feedstock selection; the features of the production process (DO) might be quite influential too, e.g., solvent usage.

In DO reactions, solvents not only serve as the reaction medium but also contribute to thermal stability, enhance mass transfer, and promote effective contact between reactants and catalysts. Solvent choice can markedly affect reaction kinetics, fuel quality, and process efficiency (Goh et al., 2024). Non-polar solvents like n-hexane are typically employed to reduce the viscosity of the feedstock and facilitate homogeneous mixing (Goh et al., 2024). However, the widespread use of organic solvents raises ecological and safety concerns due to their volatility, flammability, and the generation of post-reaction waste (Buldyrev et al., 2015; Kordouli et al., 2024). From a sustainability standpoint, minimizing solvent consumption per unit of fuel produced is a priority. Lower solvent usage translates to reduced operating costs, minimized emissions from solvent recovery, and less environmental risks during handling and disposal.

This study addresses the solvent-related sustainability issue by conducting a series of batch-scale DO experiments aimed at reducing the solvent-to-feedstock ratio (n-hexane:rapeseed oil mass ratio) while maintaining acceptable reaction performance. A Ni-Mo-Al catalyst synthesized through a solvent-free method was used, contributing further to the green credentials of the process. To date, most studies on green diesel synthesis have focused either on solvent-assisted deoxygenation using conventional catalysts or on novel catalyst synthesis without addressing solvent minimization (Lucantonio et al., 2023; Goh et al., 2024; Lin et al., 2023a; Nugraha et al., 2024; Mahmoudi et al., 2017). Other works have examined the influence of solvent type and quantity on the deoxygenation process (Qu et al., 2021; Maglinao et al., 2024; Zhang et al., 2024; Saleheen et al., 2019; Behtash et al., 2014). Recent advances in catalyst design for biofuel upgrading have also explored innovative preparation and activation routes, such as microwave-assisted synthesis, immobilized or supported ionic liquids, and task-specific functionalization. These strategies aim to enhance catalytic efficiency, stability, and recyclability by tailoring the microenvironment of active sites and improving mass transfer properties. A comprehensive overview of these emerging systems and their potential for biofuel upgrading is provided by recent reviews (e.g., Osman et al., 2023), which highlight their capacity to achieve high space–time yields and durable performance. However, the combined evaluation of a catalyst synthesized via a solvent-free route, together with systematically reduced solvent usage during the DO reaction, has received very limited attention. The present study specifically addresses this gap by exploring the synergy between solvent-saving

approach in catalyst design and in the deoxygenation process.

A series of catalytic DO tests was performed under controlled laboratory conditions to assess the feasibility of solvent minimization without compromising product quality or deoxygenation efficiency.

2. MATERIALS AND METHODS

2.1 TG feedstock and DO catalyst

Commercial rapeseed oil was used as the triglyceride (TG) feedstock for all experiments. Its detailed composition is available in previous studies (Lucantonio et al., 2025).

The catalyst employed—referred to as Mixed Oxides by Milling (MOM)—is a powdery solid based on oxides of nickel (Ni), molybdenum (Mo), and aluminum (Al). It was synthesized through a two-step solvent-free procedure: (i) calcination of precursor salts to form individual oxides, followed by (ii) mechanical mixing of the resulting oxides via milling.

This synthesis method and MOM formulation are original and therefore currently under a patenting procedure, with related documentation reporting additional catalyst features (Di Giuliano et al., 2023; Patent No. 102023000023556).

The solvent-free synthesis was designed to achieve a nominal of equivalent NiO between 10–30 wt%, equivalent MoO₃ between 30–80 wt%, equivalent Al₂O₃ between 10–40 wt%. These results confirmed that the synthesis successfully attained the target stoichiometry. The textural properties of the material were characterized by N₂-physisorption using BET (Brunauer-Emmett-Teller) and BJH (Barret-Joiner-Halenda) analyses: the MOM catalyst exhibited a specific surface area of 35 m²/g, a pore volume of 0.06 cm³/g, and an average pore diameter of 4 nm.

Particle size distribution (PSD), determined by laser diffraction (Malvern Mastersizer 2000), showed a bimodal profile with peaks between 0.3–3 µm (maximum ≈ 0.8 µm) and 3–600 µm (maximum ≈ 10 µm). Cumulative PSD percentiles were: ≈ 0.6 µm (10th percentile), ≈ 5.9 µm (50th percentile), and ≈ 56.1 µm (90th percentile). The Sauter mean diameter was ≈ 2 µm, consistent with the ball-milling process.

2.2 Experimental setup and procedure

All catalytic deoxygenation (DO) were conducted using a PARR® 4598 Mini Batch Reactor equipped with a 100 mL stainless steel vessel suitable for high-pressure operations. This setup allowed precise control of temperature, hydrogen pressure, and stirring rate—key parameters for the DO process.

Reaction conditions were selected based on methodologies reported in the literature (Lucantonio et al., 2025; Di Vito Nolfi et al., 2021) and are summarized in Table 1.

Before each run, the catalyst was pre-reduced in situ at 320°C for 4 hours under an initial hydrogen pressure of 60 bar (measured at room temperature).

DO reactions were conducted using n-hexane as the solvent, with n-hexane:oil mass ratios ($R_{\text{S/O}}$) ranging from 10:1 to 4:1. The initial oil mass (m_{oil}) varied from 2 g to 5 g,

TABLE 1: Experimental reaction conditions (RT*: Room Temperature).

Reaction parameters	Value
Temperature (T) [°C]	320
Catalyst/oil mass ratio (γ) [%]	10
Starting P(H ₂) (RT*) [bar]	40
Impeller rotation [rpm]	600
Reaction time [h]	6

with the goal of increasing oil load per batch and thereby enhancing productivity.

At the end of each experiment, the catalyst was separated by filtration, and the solvent (n-hexane) was removed by rotary evaporation, leaving the final liquid product whose mass (m_{product}) was recorded.

The composition of the liquid product was determined via gas chromatography (GC), as described in Lucantonio et al., 2025. Peak areas ($A[\%]_p$) were quantified for the following product (p) categories:

- C_i: n-alkanes with i carbon atoms (e.g., C₁₅-C₁₈)
- FAME: unconverted TG, secondarily transformed into FAME during analytical preparation
- UHC: unsaturated hydrocarbons
- BHC: branched hydrocarbons
- Others: unidentified components

2.3 DO performance metrics

To evaluate the DO process performance, the following metrics were calculated from experimentally measured quantities (Lucantonio et al., 2025; Kaewchada et al., 2021; Ruangudomsakul et al., 2021; Sonthalia & Kumar, 2019; Nagraha et al., 2024):

Product yield (Y_{product} , Equation 4), i.e., percentage share of the initial mass of oil (m_{oil}) that is found as a liquid product (m_{product}) at the end of each test:

$$Y_{\text{product}} = \frac{m_{\text{product}}}{m_{\text{oil}}} \cdot 100 \quad (4)$$

Triglyceride conversion (x , Equation 5), i.e., the percentage share of initial oil mass converted into hydrocarbons (C_p, UHC, BHC):

$$x = \left(1 - \frac{m_{\text{product}} \cdot \left(\frac{A[\%]_{\text{FAME}} + A[\%]_{\text{Others}}}{100} \right)}{m_{\text{oil}}} \right) \cdot 100 \quad (5)$$

Diesel-range hydrocarbon yield (Y_{diesel} , Equation 6), i.e., the percentage share of initial oil mass converted into diesel hydrocarbons C₁₅-C₁₈:

$$Y_{\text{diesel}} = \frac{m_{\text{product}}}{m_{\text{oil}}} \cdot \sum_{i=15}^{18} A[\%]_{C_i} \quad (6)$$

HDO vs. DCO_x selectivity ($S_{\text{HDO/DCO}_x}$, Equation 7), i.e., the preference towards HDO or DCO_x paths (values importantly higher than 1 indicate a preference towards HDO; values lower than 1 indicate a preference towards C removing paths DCO and DCO₂):

$$S_{\text{HDO/DCO}_x} = \frac{A[\%]_{C_{18}} + A[\%]_{C_{16}}}{A[\%]_{C_{17}} + A[\%]_{C_{15}}} \quad (7)$$

In addition, the mass-green-metric “Mass Productivity” defined by Roger A. Sheldon (Sheldon, R. A., 2018) was

adapted to this case of study (MP, Equation 8): MP estimates the fraction of the total mass loaded in the reactor that became desired product (i.e., diesel) on the basis of:

$$MP = \frac{A[\%]_{C_{15-C_{18}}} \cdot m_{\text{product}}}{m_{\text{oil}} + m_{\text{solvent}} + m_{\text{catalyst}} + m_{\text{H}_2}} \quad (8)$$

where m_{solvent} , m_{catalyst} , and m_{H_2} are the initial masses of hexane, MOM, and hydrogen, respectively.

All metrics were derived from single experimental runs. Although replicated tests were not performed in the present work, the same PARR® 4598 batch reactor and GC-FID analytical setup described in Lucantonio et al. (2025) were used. In that study, three replicated experiments yielded in a standard deviation of approximately ± 2 wt% (absolute) for diesel yield, corresponding to an overall reproducibility of about $\pm 3\%$ RSD. Considering the identical equipment and analytical procedures, this value is adopted as a representative estimate of uncertainty. The estimated uncertainty ($\pm 3\%$ RSD) primarily reflects the combined reproducibility of the reactor system and GC-FID quantification, and therefore applies to all mass- and composition-based performance indicators (Y_{product} , x , and Y_{diesel}). In contrast, selectivity and peak-area percentages are expected to fall within a narrower analytical uncertainty of ≈ 1 -2%.

2.4 Estimations of hydrogen availability and reactor filling

The 10:1 R_{sl} with 2 g of oil was adopted from prior studies (Lucantonio et al., 2025) as a reference condition. Given the aim to minimize solvent usage and increase oil load, hydrogen availability was assessed as a critical process parameter.

The volume of the reactor occupied by liquids (V_L) was estimated by Equation 9 (assuming the ideal behavior of the mixing), where m_i and ρ_i denote the initial mass and density (at room temperature) of rapeseed oil and n-hexane:

$$V_L [l] = \left(\frac{m_{\text{n-hexane}}}{\rho_{\text{n-hexane}}} + \frac{m_{\text{oil}}}{\rho_{\text{oil}}} \right) \quad (9)$$

Hydrogen availability was then estimated using the ideal gas law and normalized against the minimum stoichiometric requirement for full deoxygenation, assuming triolein as a representative TG. Equation 10 defines the hydrogen coverage ratio (HCR):

$$HCR = \frac{P \cdot (V_{\text{tot}} - V_L)}{R \cdot T} \div (15 \cdot n_{\text{triolein}}) \quad (10)$$

where V_L is the volume of rapeseed oil and hexane, P and T are the initial loading pressure and temperature of H₂, R is the ideal gas constant, V_{tot} is the total reactor volume, and $(15 \times \text{moles of triolein})$ is the estimated minimal H₂ demand for total DO (see figure 1). While triolein may slightly overestimate hydrogen demand due to its higher unsaturation, the HCR remains a useful indicator of system capacity.

3. RESULTS AND DISCUSSION

Table 2 presents the composition of the products obtained from the DO reactions. Table 3 summarizes the main performance metrics: product yield (Y_{product}), triglyceride conversion (x), diesel yield (Y_{diesel}), the HDO vs. DCO_x

TABLE 2: Product composition of DO tests as GC-FID peak-area percentages. Estimated analytical uncertainty $\pm 1\text{--}2\%$ based on instrument reproducibility (Lucantonio et al., 2025).

Entry	R_{SIO}	$m_{oil}[g]$	$C_8\text{--}C_{14}$	$C_{15}\text{--}C_{18}$	$C_{19}\text{--}C_{30}$	BHC	UHC	FAME
1	10:1	2	2.0	88.9	2.9	5.8	0.4	0.0
2	7:1	2	0.0	95.5	2.1	1.6	0.0	0.8
3	7:1	3	3.2	62.7	2.4	1.0	2.2	23.0
4	4:1	3	0.8	94.2	3.0	1.6	0.0	0.0
5	4:1	5	0.0	34.8	0.6	0.0	3.1	60.4

TABLE 3: Performance evaluation metrics of DO tests, including mass productivity (MP) as an indicator of material efficiency (g_{diesel}/g_{input}). Estimated experimental uncertainty $\pm 3\%$ RSD for mass- and composition-based quantities ($Y_{product}$, x , Y_{diesel}) based on Lucantonio et al. (2025).

Entry	R_{SIO}	$m_{oil}[g]$	$Y_{product}[\text{wt}\%]$	$x[\text{wt}\%]$	$Y_{diesel}[\text{wt}\%]$	$MP[g_{diesel}/g_{input}]$
1	10:1	2	75.9	100.0	67.5	0,064
2	7:1	2	83.8	99.3	80.0	0,099
3	7:1	3	81.2	76.8	50.9	0,063
4	4:1	3	78.8	99.6	74.2	0,146
5	4:1	5	82.4	49.4	28.7	0,056

selectivity ratio ($S_{HDO/DCOx}$), and the mass productivity (MP) at varying n-hexane:oil mass ratios (R_{SIO}) and initial oil load.

The data reported in Tables 2 and 3 are illustrated in Figures 2 and 3. Figure 2 compares triglyceride conversion (x) and diesel yield (Y_{diesel}) for the three representative conditions: reference (10:1, 2 g), intermediate (7:1, 3 g), and constrained (4:1, 5 g). Figure 3 further separates the effects of the R_{SIO} and oil loading. Figure 4 shows the variation of the hydrogen coverage ratio (HCR, 4a) and the reactor liquid volume fraction (V_L , 4b).

3.1 Effect of n-hexane:oil mass ratio and oil load

The reference test-based on Lucantonio et al. (2025) using other catalysts-consisted of a 10:1 R_{SIO} and 2 g of rapeseed oil. Under these conditions, the MOM catalyst achieved excellent outcomes, with 100% triglyceride conversion and a diesel yield of 67.5 wt% (Table 3, Entry 1), and nearly 89% of the liquid product falling within the diesel-range ($C_{15}\text{--}C_{18}$) (Table 2, Entry 1). This served as a benchmark for comparative analysis.

From this starting point, progressively more constrained tests were designed to evaluate the impact of limited-solvent operation and increased oil load. A particularly

demanding configuration-4:1 R_{SIO} with 5 g of oil-resulted in significant performance losses (Figure 2): conversion dropped to 49.4% and diesel yield to 28.7 wt% (Table 3, Entry 5). Product composition confirmed incomplete reaction, with 60.4% of the product identified as FAMEs, and diesel-range hydrocarbons reduced to just 34.8% (Table 2, Entry 5). These losses can be associated with the lower HCR (≈ 0.8 , Figure 4a) combined with reduced solvent

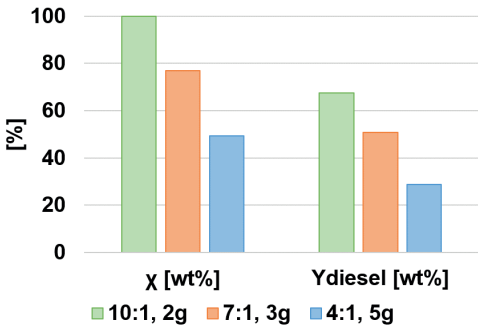


FIGURE 3: (a) Influence of n-hexane:oil mass ratio at fixed oil mass; (b) Influence of oil mass at fixed n-hexane:oil mass ratio. Estimated uncertainty $\pm 3\%$ RSD (Lucantonio et al., 2025).

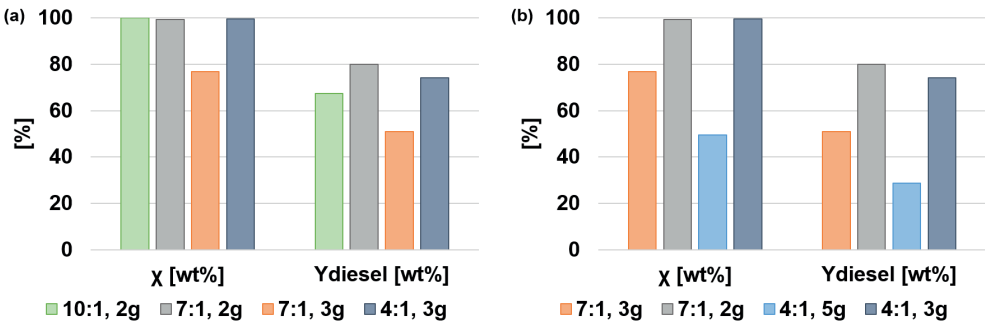


FIGURE 2: Comparison of triglyceride conversion (x) and diesel yield (Y_{diesel}) for the reference (10:1, 2 g), intermediate (7:1, 3 g), and constrained (4:1, 5 g) conditions. Estimated uncertainty $\pm 3\%$ RSD (Lucantonio et al., 2025).

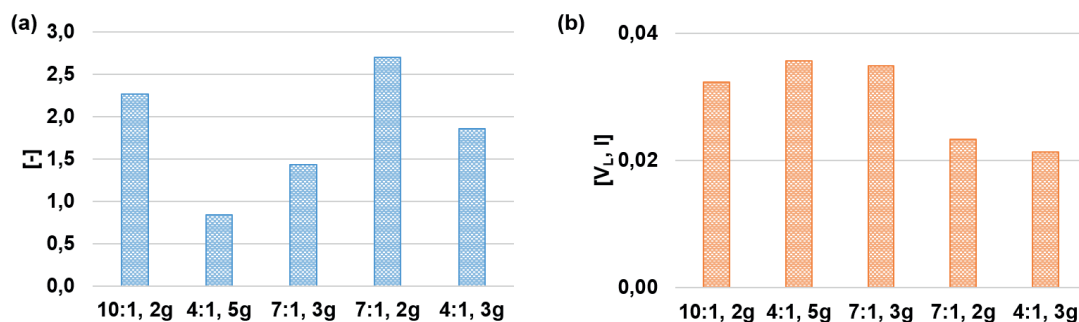


FIGURE 4: (a) estimated hydrogen coverage ratio and (b) reactor volume filled by liquids (VL) across all tested conditions.

volume. Such conditions may have limited both hydrogen availability ($HCR < 1$) and gas-liquid mass transfer efficiency, as reported in previous studies (Maglinao et al., 2024; Žula et al., 2022; Wu et al., 2021; Saleheen et al., 2019).

To improve productivity, an intermediate test was conducted using a 7:1 $R_{S/O}$ and 3 g of oil. This configuration partially restored performance, achieving 76.8% conversion and a diesel yield of 50.9% (Table 3, Entry 3), likely due to the higher HCR (≈ 1.4 , Figure 4a), which reflects greater hydrogen supply. Nevertheless, conversion and yield in this case (7:1, 3 g) did not reach the benchmark values obtained for 10:1, 2 g (Figure 2), possibly due to the lower HCR (< 1.5 , Figure 4a).

Figure 3 further clarifies the separate effects of $R_{S/O}$ and oil loading. Figure 3a compares cases with equal oil loading (2 g or 3 g), while Figure 3b compares cases with equal $R_{S/O}$ (7:1 or 4:1). When the oil mass is fixed at 2 g and the $R_{S/O}$ is varied between 10:1 and 7:1, outcomes remain largely unchanged, with a slightly higher diesel yield at $R_{S/O} = 7:1$. When the oil mass is fixed at 3 g, a noticeable improvement in both conversion and diesel yield is observed when $R_{S/O}$ decreases from 7:1 to 4:1. This can be explained by the corresponding change in HCR (Figure 4a): for 3 g of oil at $R_{S/O} = 7:1$, the HCR is only slightly above one, indicating just sufficient hydrogen availability. However, when the solvent amount is reduced (4:1), the liquid volume (V_L) decreases (Figure 4b) and the HCR increases (to about 1.8, Figure 4a). This suggests that, within the studied range, solvent reduction can be beneficial—provided hydrogen availability remains adequate. In these experiments, an empirical threshold of $HCR \geq 1.5$ (Figure 4) appears necessary to ensure satisfactory performance. Figure 3b shows that increasing the oil mass while holding a constant $R_{S/O}$ leads to a significant drop in performance. Both conversion and diesel yield decline sharply when moving from 3 g to 5 g of oil at a 4:1 $R_{S/O}$, and from 2 g to 3 g at 7:1 $R_{S/O}$. These results indicate that excessive oil loading—without a corresponding increase in hydrogen—worsens DO performance. Excessive oil loading and the associated decline in conversion and yield correspond to HCR values (Figure 4a) below the empirical threshold of 1.5 identified earlier in Figure 3a. This confirms that hydrogen availability is a key factor for successful DO under the examined cases.

These observations also correlate with the differences in V_L values (Figure 4b): the two tests with the poorest performance correspond to the highest V_L values and oil

loadings. Thus, while solvent reduction is feasible and even advantageous within reasonable limits, maintaining an appropriate balance between oil mass and hydrogen supply is essential to preserve reaction efficiency.

Overall, the combined comparison of Figures 2-4 suggests an interdependence between oil loading and hexane loading, both of which influence bulk hydrogen availability and hydrogen transfer through the liquid phase. Since hydrogen was preloaded in the reactor rather than continuously fed during the DO process, the actual liquid level and hydrogen partial pressure directly affect hydrogen provision to the catalyst surface. Empirically, ensuring an adequate HCR appears necessary, even though it may not be sufficient to ensure complete deoxygenation.

In summary, the 4:1 $R_{S/O}$ with 3 g of oil emerged as the compromise between process performance and solvent consumption, achieving high conversion ($> 99\%$) and diesel yield (74.2 wt %) while substantially reducing solvent usage. This condition likely benefits from maintaining HCR well above one (empirical threshold ≈ 1.5); in constant-volume systems, this corresponds to avoiding excessive oil loading that would otherwise limit hydrogen availability.

To complement the traditional DO performance indicators, the mass productivity (MP) was calculated as in Equation 8. MP values (Table 3) varied between 0.056 and 0.146 $g_{\text{diesel}}/g_{\text{input}}$. Reducing the solvent-to-oil ratio from 10:1 to 7:1 (2 g oil) increased MP by over 50%, confirming that less solvent enhances atom economy. The 4:1, 3 g test exhibited the highest productivity (0.146 $g_{\text{diesel}}/g_{\text{input}}$), reflecting the most effective balance between solvent reduction and hydrogen supply. Conversely, further oil increase to 5 g under the same ratio led to a sharp MP decline, consistent with the drop in conversion and yield. These results emphasize that optimal productivity arises from coupling solvent reduction with sufficient hydrogen availability rather than from maximizing oil load alone.

3.2 Catalyst performance

To further evaluate the MOM catalyst's performance, product selectivity was examined for all tests made.

As shown in Figure 5a, the optimal test (4:1, 3 g) yielded over 90% diesel-range hydrocarbons (C_{15} - C_{18}), closely matching the outcome of the 7:1, 2 g experiment. In contrast, increasing the oil load to 5 g at the same $R_{S/O}$ ratio reduced this fraction to below 40%, confirming the catalyst's sensitivity to hydrogen limitation.

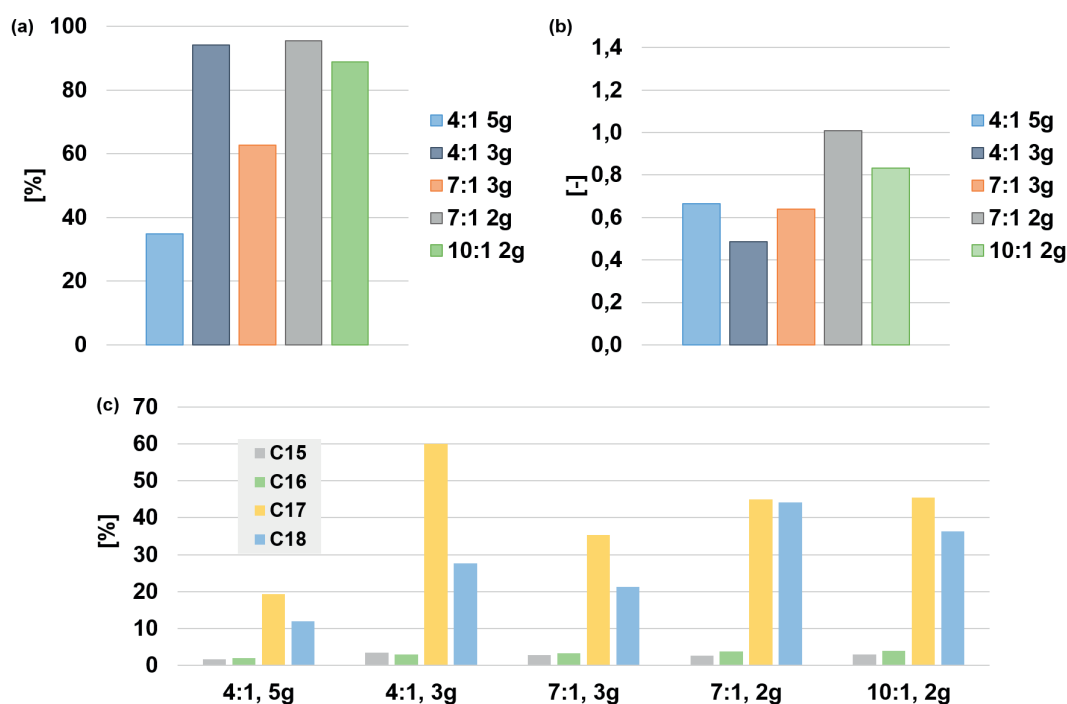


FIGURE 5: (a) A[%] of hydrocarbons in the C₁₅-C₁₈ range; (b) S_{HDO/DCO_x} selectivity ratio; (c) detailed composition of diesel-range hydrocarbons. Analytical uncertainty within ± 1 -2 % based on GC-FID reproducibility.

Selectivity data (Figure 5b) showed that the HDO/DCO_x ratio remained below 1 in all cases, indicating that decarbonylation (DCO) and decarboxylation (DCO₂) were the dominant reaction pathways. Nevertheless, higher solvent loadings and lower oil masses enhanced selectivity toward hydrodeoxygenation (HDO), suggesting a shift toward more favorable reaction routes under hydrogen-rich environments. This behavior is consistent with the stoichiometric requirements of the HDO, DCO, and DCO_x, as HDO requires a higher H₂-to-feed ratio per mole of converted fatty acid. This trend can be attributed to the combined effects of limited hydrogen solubility and restricted gas-liquid mass transfer under more constrained conditions (lower R_{S/O} and higher oil loading). As the available solvent volume decreases, the effective hydrogen concentration at the catalyst surface also declines, reducing the probability of hydrogenation steps required for HDO. Consequently, the reaction preferentially follows the less hydrogen-demanding DCO/DCO₂ pathways.

Further evidence of this behavior is provided by the relative distribution of C₁₇ and C₁₈ hydrocarbons (Figure 5c). The formation of C₁₇ species implies the loss of one carbon atom from the parent fatty chain—typical of DCO/DCO₂ routes—and their prevalence over C₁₈ is a clear indicator of carbon loss. In the 4:1, 3 g case, C₁₇ accounted for approximately 60% of diesel-range hydrocarbons, while C₁₈ contributed only 28%, confirming dominance of decarbonation mechanisms.

Although diesel-range alkanes were the primary objective, minor amounts of light (C₈-C₁₄) and heavy (C₁₉-C₃₀) n-alkanes were also detected (Table 2). While these fractions are less valuable for direct use in CI engines, they could be further refined or valorized into other fuel streams,

thereby enhancing overall process circularity and product utilization.

4. CONCLUSIONS

This work presents an experimental assessment of a more sustainable approach to green diesel production through solvent minimization during the catalytic deoxygenation of triglycerides. A novel Ni-Mo-Al catalyst, synthesized via a solvent-free procedure, was tested under varying R_{S/O} ratios and oil loadings to evaluate its performance in increasingly constrained process conditions.

The results demonstrated that the reference case—R_{S/O} = 10:1, 2 g of rapeseed oil—achieved complete triglyceride conversion and a diesel yield of 67.5 wt%, confirming the high activity of the MOM catalyst. As the solvent-to-oil ratio was decreased and the oil load increased, reaction outcome was significantly affected. In particular, the test conducted at a 4:1 ratio with 5 g of oil exhibited a marked decline in both conversion and diesel-range yield, along with a substantial amount of unconverted feedstock. These findings confirm the critical influence of solvent volume and process intensification on hydrogen availability and catalyst accessibility. However, an intermediate condition—4:1 R_{S/O} with 3 g of oil—proved to be an effective trade-off. Here, the catalyst maintained over 99% conversion and a diesel-range hydrocarbon yield of 74.2 wt%, with more than 90% of hydrocarbons falling within the C₁₅-C₁₈ range. These results demonstrate that, in a 100 mL batch reactor, solvent usage can be reduced to a 4:1 ratio while retaining acceptable yield and selectivity.

Throughout the experiments, product selectivity analysis revealed a predominance of DCO₂ and DCO pathways

over HDO, especially under hydrogen-limited environments. Nevertheless, partial HDO activity was retained, under higher solvent loadings or lower oil masses. These outcomes highlight the need for careful process balancing to ensure both fuel quality and sustainability goals.

Overall, the MOM catalyst showed strong performance and robustness under reduced solvent conditions, confirming its effectiveness for green diesel production in small-scale batch systems.

From a broader perspective, these findings represent a first step toward developing greener process designs for green diesel. Integrating solvent-free catalyst synthesis with solvent-minimized operation could ultimately reduce separation costs and enhance overall process sustainability, as gathered in evaluations in the mass-green-metric “mass productivity”. Further validation under continuous-flow and pilot-scale systems will be required to translate these promising results toward industrial application.

ABBREVIATIONS

(CI) - Compression Ignition Engine
(DCO) - Decarbonylation
(DCO₂) - Decarboxylation
(DO) - Deoxygenation
(FAME) - Fatty Acid Methyl Esters
(FFA) - Free Fatty Acid
(GHG) - Greenhouse Gases
(HDO) - Hydrodeoxygenation
(LCA) - Life Cycle Assessment
(MOM) - Mixed Oxides Mill
(TGs) - Triglycerides

LIST OF SYMBOLS

(%V_L) - Percentage reactor filling by liquids (Equation 8)
(A[%]_p) - percent of area of component p in Agilent GC-FID chromatogram with p = C_i for the n-alkane with i carbon atoms; FAME for unconverted oil; UHC for unsaturated hydrocarbons; BHC for branched hydrocarbons; Others for unidentified peaks
(m_{oil}) - Initial oil mass
(m_{product}) - Mass of recovered product
(n_{triolein}) - Moles of triolein
(MP) - Mass productivity
(PSD) - Particle size distribution
(S_{HDO/DCOx}) - Catalyst Selectivity (Equation 7)
(V_{tot}) - Total reactor volume
(V_L) - Reactor volume filling by liquids
(x) - Conversion of TGs (Equation 5)
(Y_{diesel}) - Diesel Yield (Equation 6)
(Y_{product}) - Product Yield (Equation 4)

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CIRCULARITY BY DIGITAL FABRICATION FROM THEORY TO PRACTICE: THE CASE OF THE WOOD ARCHITECTURE AND DESIGN INDUSTRY

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ABSTRACT

This paper examines the application of digital technologies as circularity drivers in the wood design industry analyzing project scalability, material sustainability, and the challenges of transitioning from theoretical research to practical implementation. To this end, a scoping review methodology was employed in a time frame between 2015 and 2023 to identify key concepts, research gaps, and emerging trends, through the web platform Scopus. Since using keywords such as “furniture manufacturing,” “CNC,” “3D printing,” “sustainable material,” “sustainability,” and “recycle wood,” initially retrieved results across diverse fields, the focus was narrowed to architecture and design, but then yielded limited findings, highlighting the lack of sector-specific studies. To address this gap, the review was supplemented with an analysis of relevant case studies. Findings indicate that most documented applications of digital technologies for circular wood design originate in Europe – particularly Denmark – where institutional support and policy initiatives have fostered experimental, university-led projects. Despite their innovation, these cases remain largely unvalidated in academic literature, exposing a disconnect between empirical experimentation and theoretical consolidation. The increase in publications from 2023 suggests a shift toward formal validation of earlier experiments, potentially signaling the emergence of a sustained research trend in this domain.

1. INTRODUCTION

Wood as a foundational raw material, especially from forest-rich areas, is crucial for global construction and manufacturing. Nevertheless, wood-based industry – and architecture and design especially – faces severe environmental and ecological concerns such as deforestation, extensive material waste, and heavy dependence on virgin materials; even as wood is celebrated for its sustainability (Bergsagel & Heisel, 2023; Wagner et al., 2020). These challenges are recognized in international frameworks, including the European Union Circular Economy Action Plan and the United Nations Sustainable Development Goals, which emphasize responsible production, resource efficiency, and climate action.

Addressing escalating concerns regarding sustainability and digitalization, digital technologies like digital prototyping, robotic fabrication, and CNC machining are being recognized as essential tools for optimizing material use and improving production efficiency (Alvarez et al., 2019; Velikov et al., 2016). The potential of these innovations lies in their capacity to minimize environmental impact by en-

abling accurate material utilization, while offering greater creative flexibility in design. However, beyond the premises, there is still a significant lack of knowledge regarding the effectiveness of application of digital technologies and methods for circularity in wood architecture and design.

To identify key concepts, research gaps, and emerging trends in the integration of digital fabrication technologies in the wood industry, this study applies a scoping review methodology, following the framework of Arksey & O'Malley (2005), to systematically map existing research and experimental practices as publications. The scoping review was conducted using the Scopus database with keywords including “digital fabrication,” “CNC,” “3D printing,” “wood industry,” “life cycle assessment,” and “sustainable materials.” However, the findings of this literature review show that limited existing research on this topic exists: the results of this study outline a significant gap between theoretical advancements and practical application in the industry. Despite innovation in research and development in the field of digital fabrication, many companies seem to continue to depend on traditional methods, limiting their ability to fully leverage digital innovations (Wagner et al., 2020).



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Indeed, the second part of the study is conducted to bridge this gap by examining case studies from leading academic and industry institutions. Studies from 2015 to 2023 were selected, focusing on European architectural publications and case studies. The analysis considered materials, processes, technologies, and sustainability outcomes. In this way, it was questioned how emerging digital technologies can be effectively integrated into wood production processes to reduce waste, enhance resource efficiency, and promote sustainability.

The paper is structured in 4 paragraphs: the Introduction, "Materials and Methods", "Results and discussion", "Conclusions". In "Materials and Methods", a scoping review is conducted on digital fabrication in the wood industry, combining qualitative literature analysis and quantitative bibliometric data. The scope is refined to architectural publications and case studies are analyzed within a time frame 2015–2023 and limited to European institutions, assessing materials, processes, technologies, and sustainability outcomes. In "Results and discussion", gap between theoretical research and practical applications are identified, highlighting challenges in material sourcing, cost efficiency, and scalability. Experimental case studies showing innovative uses of reclaimed timber, bio-based materials, and digital fabrication techniques are examined and their potential and limitations are described. In "Conclusions", it is underlined the transformative role of robotic fabrication, CNC machining, and additive manufacturing in optimizing material use and reducing waste. Key barriers include high costs, slow industry adoption, and scalability issues. Future research outlooks are identified in cost-effective implementation strategies, interdisciplinary collaboration, and industry integration to enhance sustainability in wood fabrication.

2. MATERIALS AND METHODS

This research adopts a scoping review methodology to map the existing literature on the integration of digital fabrication technologies in the wood design industry, focusing on sustainable practices and material efficiency. Scoping reviews are valuable for identifying the scope and extent of research activities, especially in areas that are complex or have not been comprehensively reviewed before (Munn et al., 2018).

We employed both qualitative and quantitative methods to ensure a comprehensive analysis. The qualitative approach involved an in-depth review of existing literature, case studies, and industry reports to understand key themes, trends, and challenges in digital fabrication for sustainable wood design. The quantitative approach consisted of analyzing bibliometric data from the Scopus database, including the number of relevant publications per year, geographical distribution of studies, and keyword frequency to identify research patterns in this field.

To narrow down the focus, we filtered the results to include only publications within the field of architecture, design and building engineering. The rationale for making these filters is that architecture and the built environment are among the largest users of wood and wood products,

where material use efficiency, waste minimization, and scalability rank as top priorities. As compared to other fields, architecture operates at close presence to sustainability issues, circular design, and mass implementation. By constraining the scope in this way, the review keeps its conclusions industry-specific and pertinent to real issues in sustainable wood construction and building.

Our primary objective was to map the available literature on the use of CNC, 3D printing, and robotic fabrication techniques applied to wood, emphasizing sustainable practices and material reuse.

We searched the Scopus and Web of Science databases, covering publications from 2000 to 2023 and limited to English-language articles. The search strategy was organized into three main categories: fabrication technology (CNC, 3D printing, robotics), circularity and material reuse (recycling, upcycling, biocomposites), and sector-specific applications (furniture, architectural elements, design objects).

After initial retrieval, duplicate records were removed, and publications were screened based on relevance to experimental fabrication, application scale, and focus on sustainability. Data extraction included bibliometric information, type of fabrication technology, material used, product scale, and circularity considerations. This process allowed a structured comparison between quantitative bibliometric data and qualitative case studies.

Due to the limited findings, we adapted our methodology to include an analysis of case studies. We expanded our research to include case studies published between 2015 and 2023, primarily from European regions, with Denmark being prominently represented. We sourced these case studies from additional sources, including project reports and institutional databases such as the Royal Danish Academy and the University of Stuttgart's Institute for Computational Design, rather than from the scoping review. The evaluation criteria for the case studies included project scale, material usage, sustainability outcomes, and practical challenges during implementation.

By examining diverse applications involving reclaimed timber, bio-based materials, and advanced digital fabrication techniques, we aimed to provide a comprehensive overview of how these innovations are currently applied in the wood industry. Comparing methodologies and outcomes across these projects allowed us to identify key challenges and opportunities for further technological integration.

3. RESULT AND DISCUSSION

While our initial approach focused on a scoping review to assess the broadness of research on digital fabrication in the wood industry, we observed a significant gap between theoretical studies and practical applications (Table 1, Figure 1). From the systematic search, a total of 66 documents were retrieved across different query combinations. After removing duplicates and excluding papers outside the scope of architecture, design, and sustainability, such as those focusing on chemical, medical or unrelated engineering applications, the number of relevant publications

TABLE 1: Limits of detection (LOD) for the regulated HMs (Cd, Pb and Hg). Concentrations on mg/kg are evaluated considering the digestion volume and dilution utilized for the quantifications, where necessary.

Article title	Authors	Source	Year	Citations
Investigation of material properties of composite materials based on wood waste for FDM Additive manufacturing	Beniak, J.; Klepochavà, K.; Holdy, M.; Soos, L.	Waste Forum (3), pp. 156-165	2023	0
Development and carachterization of sustainable PLA/Olive wood waste composites for rehabilitation applications using Fused Filament Fabrication (FFF)	Fico, D.; Rizzo, D.; De Carolis, V.; ... Palumbo, E.; Corcione, C. E.	Journal of Building Engineering, 56, 104673	2022	42
Creating novel furniture through topology optimization and advanced fabrication	Ma, J.; Li, Z.; Zhao, Z.IL.; Xie, Y. M.	Rapid Prototyping Journal, 27(9), pp. 1749-1758	2021	27

was reduced to 7. From this smaller pool, Additive Technologies and Their Applications in Furniture Design and Manufacturing (Jarža et al., 2020) emerged as the most relevant quantitative contribution. This paper provides numerical insights into material utilization, efficiency, and recycling potential in 3D-printed furniture, demonstrating the possibilities of additive manufacturing for circular design. However, its relevance is limited for the purposes of this research, as it focuses exclusively on furniture rather than architectural or spatial applications, and it does not address broader sustainability strategies, modularity, or living-lab contexts. Despite these limitations, the study offered a useful quantitative reference point, supporting the integration of advanced fabrication methods into discussions on circularity in wood design.

The identified gap between theoretical studies and practical architectural applications is clearly supported by this evidence from the literature.

The scarcity of literature in the architecture and sustainability domains led us to complement our review with a detailed case study analysis. This shift allowed us to examine real-world applications of digital fabrication technologies, offering deeper insights into their feasibility, challenges, and impact on sustainable wood design.

To select and analyze case studies, we established a set of evaluation criteria, focusing on:

- Materials used (reclaimed timber, bio-based composites, prefabricated elements);
- Processes applied (robotic fabrication, CNC machining, additive manufacturing);
- Technologies and machines involved (3D printing, para-

metric modeling, automated assembly);

- Sustainability impact (waste reduction, life cycle assessment, material efficiency).

The selected case studies, spanning from 2015 to 2023, are sourced from European institutions, with Denmark being particularly active in this field. It is notable that most of the case studies are from 2023, reflecting the recent acceleration in the application of digital construction technologies in timber architecture. The analysis of these cases highlights key challenges in material sourcing, cost efficiency, and structural integrity, while also showcasing innovative approaches to extending the lifespan of wood materials through advanced digital fabrication techniques.

For instance, The Wood ReFramed Pavilion (Figure 2), developed by researchers at the Royal Danish Academy, exemplifies the integration of reclaimed timber into modern construction. This project demonstrates how digital fabrication methods can enhance the reuse of wood while maintaining structural integrity and design flexibility (Browne et al., 2023). Despite its success, challenges such as material availability, ensuring structural safety, and scalability remain significant barriers to broader adoption (Bergsagel & Heisel, 2023).

The livMatS Biomimetic Shell (Figure 3) project, led by the University of Stuttgart, further highlights the potential of digital technologies in timber architecture. This 200 m² structure utilizes bio-based materials and computational fabrication techniques to create a sustainable architectural solution (Wagner et al., 2020). Employing both additive and subtractive manufacturing processes, along with a 4D-printed adaptive shading system, the project illustrates how digital tools can enhance precision and sustainability in complex designs (Alvarez et al., 2019).

Other notable examples include from the 2022 Women in Architecture exhibition, which focuses on reducing material waste by using reclaimed timber off-cuts and crooked oak logs. This project underscores the efficiency of digital modeling in design and assembly processes (Publications, 2016). Similarly, the Wisdome Stockholm project showcases the use of laminated veneer lumber (LVL) in constructing a large-scale timber grid-shell, demonstrating how prefabrication techniques contribute to sustainable, high-performance timber structures (Osnaburgh, 2023).

These case studies offer valuable insights into the current state of digital fabrication technologies in the wood industry. The Wood ReFramed Pavilion highlights the promise of using reclaimed timber but also reveals challenges

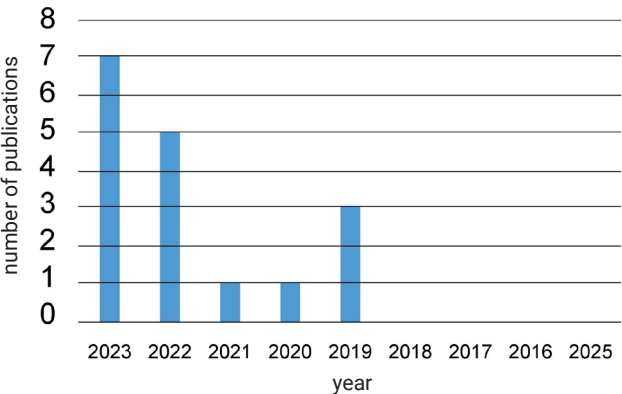


FIGURE 1: Average publication from 2015 to 2023.

related to sourcing materials and ensuring structural integrity (Browne et al., 2023; Bergsagel & Heisel, 2023). Addressing these issues is crucial for developing scalable solutions and establishing industry-wide standards that facilitate broader adoption.

The livMatS Biomimetic Shell project illustrates the effectiveness of integrating bio-based materials with computational design to achieve sustainability goals through reduced waste and increased precision (Wagner et al., 2020). Nevertheless, the project's complexity and associated costs present obstacles to widespread implementation, especially in large-scale industrial settings.

The Marina Spa Prototype (Figure 4) and Wisdome Stockholm projects demonstrate how digital fabrication can optimize material usage and minimize waste, emphasizing the significance of modular design and prefabrication in advancing sustainable timber construction (Os-naburgh, 2023).

Together, these case studies provide concrete evidence supporting the discussion of gaps in the literature, particularly regarding scalability, cost, and integration of sustainability strategies (Table 2).

As the case study analysis shows that among the various digital manufacturing techniques, robotic manufacturing, CNC machining and additive manufacturing (3D printing) emerge as the most promising technologies for sustainability. These processes stand out due to their ability to optimize material use, minimize waste and increase design accuracy. For example, CNC machining allows for high precision cutting that significantly reduces undercutting, while robotic manufacturing increases automation and material efficiency, particularly in wood manufacturing. Additive manufacturing, although still in its early stages in wood design, has the potential to reduce reliance on raw materials by enabling circular production models.

From these case studies, 3 key lessons might be summarized:

- Result 1: Material selection plays a critical role in sustainability: Projects that integrate recycled wood, bio-composites, or prefabricated techniques show significant reductions in environmental impact.
- Result 2: Industry adoption remains a challenge: While digital fabrication offers clear benefits, the high initial investment in technology and expertise limits widespread implementation in traditional wood fabrication.
- Result 3: Collaboration between academia and industry is essential: Many projects remain at a laboratory scale with limited real-world applications due to the lack of structured industrial partnerships and practical testing environments.

This gap stems from several factors:

- Slow adoption of advanced digital tools in industry: while research is exploring advanced techniques, many wood manufacturers still rely on conventional methods due to cost concerns and unfamiliarity with the technology.
- Limited interdisciplinary integration: digital fabrication often involves architecture, engineering, and compu-



FIGURE 2: Wood ReFramed Pavilion. Photo by Sara Lohse, 2023.



FIGURE 3: livMatS Biomimetic Shell. Photo by Roland Halbe, 2023.

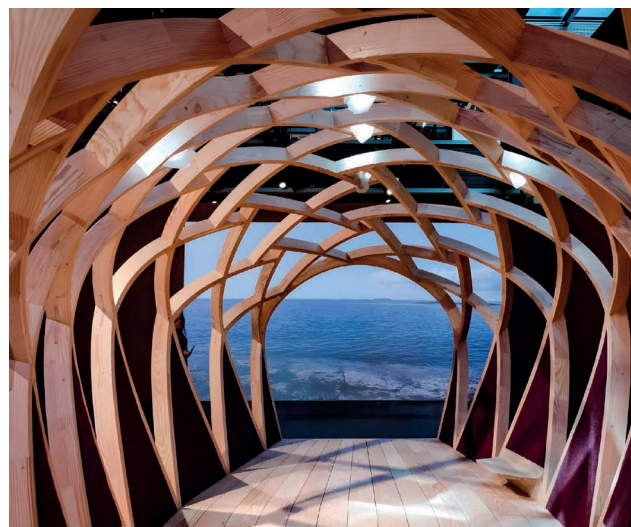


FIGURE 4: Marina Spa Prototype. Credits: Helen & Hard, 2022.

TABLE 2: Summary table of analyzed case studies.

Project Name	Institution	Objective	Scale	Materials	Workflow	Technologies	Timeline
IivMatS Biometric Shell	ICD & ISYS, University of Stuttgart	Recycling/Upcycling	Building	Waste wood, spruce boards, cellulose powder, bio-based thermoplastic polyketone	Integration of fabrication processes	Additive & subtractive fabrication, 4D printing, automated assembly	2023
Sourced Wood	IAAC	Reuse/Upcycle	Component	Waste wood	Collecting surplus material from construction sites	Robotic fabrication, optical & CT scanning, computational design	2018
Marina Spa Prototype	Danish Architecture Center	Upcycling/Reuse	Small Arch	Reclaimed timber flooring off-cuts, crooked oak logs	Integrated digital delivery	Robotic machining, scanning, computational design, laser projection	2022
Wood ReFramed	Royal Danish Academy	Reuse/Upcycle	Building segment (prototype)	Reclaimed timber	Exploring novel structural applications	Robotic machining, scanning, computational design, heuristic algorithms	2023
eGrow Willow	Karlsruhe Institute of Technology	Recycle/Upcycle	Building elements	Willow, Earth	Integrating closed-loop systems	Lightweight fabrication, scanning, computational design	2023
Weaving the Shimoni Cave	Royal Danish Academy	Recycle/Upcycle	Object		Hand-woven structure guided by computational design	LiDAR scanning, Tri-Mesh Remeshing	2022
Osnaburgh Pavilions	Xylotek	Reuse	Small Arch Pavilion	Oak (glue-laminated), locally sourced timber	Collaborative design processes	Minimal machinery, hand tools for on-site fabrication	2023
RawLam	CITA	Recycle/Upcycle	Small Arch (pavilion)	Timber	Flow from forest to timber product	Robotic machining, scanning, computational design	2022
Wisdom Stockholm	Design-to-Production, Zürich; Bliding Oscarson, Stockholm	Reuse	Small Arch	Laminated Veneer Lumber (LVL)	From competition to completed dome	Digital prefabrication, parametric modeling, CNC, design for manufacture & assembly	2022

tational design, but collaboration between these disciplines and industry professionals is sparse.

- Scalability issues: most academic research is conducted at the prototype or small-scale level, making it difficult to implement findings in large-scale industrial settings without further testing and refinement.

The intersection of the bibliometric screening and the case studies lies in that robotics and automation, responsible for most publications in the quantitative search, are also the center of experimental works such as Wood ReFramed and Marina Spa where robotic milling and automated assembly enabled reuse of piecewise timber with high precision. Prefabrication using CNC also appears in literature and practice, as with Wisdom Stockholm. Additive manufacturing, on the other hand, appears in indexed literature, particularly on the scale of furniture and prototyping but not yet on architecture-scale projects in the cases investigated. Scalability issues, logistics, and contractor cooperation, however, are sufficiently documented in case studies but hardly appear in the bibliometric body. In combination, both threads highlight the importance of CNC and robotics in achieving circularity and note that experimental investigations provide pragmatic lessons to be gleaned from the scientific literature, and that expressing results in standard terms remains a prevalent shortcoming across methodologies. These gaps are therefore supported by evidence from

both the scoping review and the case studies, confirming that additive manufacturing is mostly at the furniture/prototype level, while robotics and CNC show promise in both literature and practice.

In the final analysis, despite the advancements demonstrated in case studies, the gap between theoretical research and practical application remains evident. While digital technologies hold significant potential for enhancing sustainability in the wood industry, further research is necessary to address challenges related to scalability, cost-efficiency, and industry acceptance. Overcoming these hurdles is essential to fully harness the benefits of digital fabrication in promoting a more sustainable and efficient wood industry.

4. CONCLUSIONS

This study offers a detailed examination of digital fabrication as a strategic driver for advancing circular economy practices in the wood industry. By integrating qualitative analysis with international case studies analysis, it demonstrates how principles such as reuse, repair, modularity, and upcycling can be effectively applied to valorize wood waste. Technologies like CNC machining, robotics, and 3D printing emerge as key enablers, though their adoption varies: robotics and CNC systems show strong alignment between theory and practice, while additive manu-

facturing remains confined to furniture and prototyping applications.

The combined bibliometric and case study approach reveals practical challenges—scalability, logistics, and interdisciplinary coordination—that are underrepresented in academic discourse. These insights help delineate which digital strategies are currently feasible and where clearer industry standards and support mechanisms are needed.

Through its scoping review and targeted case analysis, the research provides a foundational mapping of existing practices and gaps, setting the stage for future framework development. Subsequent studies should aim to bridge the divide between conceptual research and real-world implementation by proposing cost-effective solutions, sector-specific guidelines, and collaborative models linking academia and industry.

As digital fabrication technologies continue to evolve, their integration into mainstream production will be essential for achieving scalable, sustainable, and resource-efficient wood design systems. This work lays the groundwork for further inquiry into their role in circular economy transitions and material waste reduction.

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BETWEEN SCARCITY AND CIRCULARITY OF RESOURCES: TECHNOLOGICAL INNOVATION FOR GRANITE SCRAP IN SARDINIA

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ABSTRACT

Throughout the past two centuries, human activities have had a profound impact on Earth, leading to the progressive consumption of natural resources, increasing in waste, pollution and alteration of ecosystems. This has induced a well-known climate change phenomenon with significant consequences for humankind and ecosystems. The growth of cities and improved living conditions will lead to increased consumption of resources, many of which are already expected to run out by the middle of the next decade. It is essential to monitor resource consumption using relevant indicators to drive economic growth toward resources decoupling. Starting from an analysis of the consumption of the four major categories of natural resources, biomass, fossil fuels, non-metallic minerals and metal ores on an international scale, attention is gradually turned to consumption at the Italian level, focusing on the consumption of non-metallic minerals for use in construction. Thus, the focus was on the granite mining district of Buddusò, in Sardinia, the largest producer in Italy. Granite quarries, which have been drastically reduced in recent decades, have generated huge amounts of processing waste, causing profound changes to landscapes and ecosystems. Based on the analysis of disused quarry sites, the types of rock abandoned have been studied in detail, investigating the chemical-physical, mechanical and technological characteristics and experimenting with innovative products and alternative uses for these wastes. The findings indicate that these residues can be utilised as a secondary raw material within the ceramics industry, also as a protective layer for ventilated facades.

1. INTRODUCTION

1.1 The Anthropocene's effects

The steam engine triggered the first industrial revolution in the second half of the 17th century, with disruptive effects on textile and metallurgical production.

Around 1870, the advent of electricity, internal combustion engines, and oil as energy source allowed for the second industrial revolution. A century later, information technology and industrial automation fuelled the third industrial revolution, opening the digital age, which is still expanding. Internet of Things (IoT), sophisticated robotics, big data, and artificial intelligence are pushing a radical transition, which has been officially recognized as the fourth industrial revolution at 2011 Hannover Fair.

Exploiting natural resources and, consequently, modifying the natural landscape emerge as a common character of this centuries-old evolution. Technological advancements have increased the opportunities for economic

growth, but they have in parallel engendered strong and enduring impacts due to anthropogenic activities.

Scholars have devoted increasing attention in recent years to innovative technologies and their contribution to increasing the anthropogenic alterations of Earth balance (Saqib and Qin, 2024), mostly due to the only maximization of profit and productivity.

The word Anthropocene - which was coined and made popular by biologist Eugene Storer and chemist Paul Crutzen in 2000 - has become current to characterize the most recent period in Earth's history, when human activity began to exert a substantial impact on the planet's climate and ecosystems (Crutzen and Stoermer, 2000).

This "new geological age" - although not officially recognized as such - is fuelled by two main dynamics: the huge amounts of greenhouse gases (GHG) due to energy production from fossil sources, with the related global warming this causes, and the more and more intense resource



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exploitation, which reduces natural stock and increases waste (Harbel et al., 2020) -. Indeed, the global consumption of raw materials has risen four-fold since 1970 (IRP, 2019) and further expansion is set by 60% by 2060 (IRP, 2019), also due to the urban population increasing (OECD, 2018). What we are recording from the 2020s onward is that Technosphere, namely the totality of human-made products, is bigger than biosphere and this profoundly alters the planet's ecosystems, causing 60% more global heating, 40% air pollution impacts, and 90% water stress, which leads to water cycle perturbation and ocean acidification, increase in extreme meteorological phenomena, disappearance of forests and biodiversity loss.

Furthermore, this scenario is accompanied by significant landscape degradation, diminution of natural resources, local economic failures, increased conflicts, and forced migrations (WEF, 2025), that even intensify climate change and contribute to environmental disasters (WEF, 2023). Additionally, the exploitation of physical resources has become easier and cheaper, especially when mining is carried out by acquiring inexpensive land in developing countries. The only way to enhance our capacity in utilizing natural resources while reducing our environmental footprint is that we must leverage emerging technologies in curbing air pollution, revitalizing degraded soils, conserving natural habitats, and enhancing carbon sequestration.

This means that we must inspire more intensely from the sufficiency approach (Jungell-Michelsson and Heikkurinen, 2022) and take concrete actions accordingly, to preserve and enhance our planet in a more resource-efficient perspective (COM, 2011).

Thanks to this approach, the value of environmental capital is projected to rise by a minimum of 28%, as reported by the COM final "EU Soil Strategy for 2030 Reaping the Benefits of healthy soils for people, food, nature and climate" (COM, 2021).

1.2 Trends in natural resource consumption: the four main categories of material resources extracted

"Material resources are natural assets deliberately extracted and modified by human activity for their utility to create economic value. They can be measured both in physical units (such as tons, joules or area) and monetary terms expressing their economic value" (UNEP, 2011).

Natural resources provide the raw materials which are then processed and transformed to satisfy basic human needs, like food, shelter, water and medicine, to produce energy for industries, homes and mobility; to build roads and buildings. Although they are vital for human survival, the quantification of natural resource consumption is a hard undertaking, due to both the intrinsic complexity of tracking their flows and the multitude of systems and methodologies employed for the purpose across various countries. Many research centres and government institutions (international, national, and regional) collect and publish data concerning the consumption of natural materials, pollution, and waste production, but they are often difficult to understand and compare, due to a lack of common pro-

ocols. Furthermore, data are usually aggregated by macro-categories of resources, providing the total amount of each of them entering an economic system, while very few or no information is available allowing to quantify separately the shares of extracted, processed, consumed or wasted resources.

Krausmann et al. (2009) studied variations in four primary categories of extracted materials: biomass, fossil energy carriers, ores and industrial minerals, and construction minerals. Their analysis showed that extraction increased eightfold in average between 1900 and 2000, with a huge peak for the construction minerals, which rose by a factor 34, while ores and industrial minerals experienced a growth index of 27, and fossil energy carriers grew by 12. The 2015 OECD report "Material Resources, Productivity and the Environment" confirmed that the quantities of materials extracted, harvested, and consumed worldwide have grown exponentially over the past century. This amount reached nearly 72 billion tons in 2010, that is twice that in 1980, and ten times more than in the early 1900s.

Over two-thirds of the global extracted in 2010 was for non-renewable resources. According to the 2019 OECD's report on natural resource consumption, the overall resource withdrawal is expected to rise to 168 Gt per year by 2060, of which metals are estimated to rise to 20 Gt (8 Gt in 2011), fossil fuels to 24 Gt (14 Gt in 2011), biomass to 37 Gt (20 Gt in 2011), and non-metallic minerals to 86 Gt (37 Gt in 2011) (OECD, 2019).

The resources withdrawn in 2017 were shared as follows: metal 10%, fossil fuels 17%, biomass 24%, and non-metallic materials 49%, of which 32% were sand, gravel, and crushed rocks. Nevertheless, the real consumption of raw resources and related environmental pressures are difficult to account for. Indeed, the international handbook on economy-wide material flow analysis (EW-MFA), jointly developed in 2023 by the United Nations Environment Programme (UNEP), the European Statistical Office (Eurostat), and the Organisation for Economic Co-operation and Development (OECD), aims at describing the interactions between a domestic economy and the natural environment, by focusing on primary material extraction, physical trade (i.e. imports and exports), waste and emissions, which flows are recorded at the country level through a coordinated method of data collection (UNEP, 2023). The survey outputs can be used to identify environmental pressures, and they are suitable to couple with Life cycle assessment approach (LCA) to compare the impacts and productivity of natural resources on a national economy level. According to the global manual on EW-MFA, indicator as:

- Domestic Extraction (DE), that measures the flows of materials that originate from the environment and that physically enter the economic system;
- Domestic material input (DMI), that measures the direct input of materials used in the economy, i.e. all materials which are of economic value;
- Domestic material consumption (DMC), that measures the total amount of material directly used in an economy;

- Material productivity (MP), that indicates the economic value generated per unit of material consumption, also called resource efficiency;
- Raw material consumption (RMC), namely “material footprint”, that measures the primary resources needed to produce a national product,

can be utilised to calculate and describe the physical resources used at various stages of economic activities, including the production of waste and emissions related. Still, when material extracted but unused is considered, the global rate of withdrawn natural resources grows due to unused domestic extraction (UDE) share, which strongly affects resource productivity (Bianchi et al., 2021).

Then, focusing on Europe-27, China and the USA only, and analysing the four main categories of natural resources consumption between 2010 and 2024, as showed in Figure 1, over the past four decades, the overall material (or resources) productivity indicator, measured as GDP/DMC, has increased, while material consumption (Domestic Material Consumption) has decreased. Many scholars attribute this trend to technological innovations introduced in production processes (Fayomi et al., 2019).

1.3 Environmental pressures of non-metallic and construction minerals

In last decades, our economies have changed from biomass-based agrarian metabolism, the largest source of used material (41%), to 33% in 2000 and 26% in 2020. (UNEP, 2024) - toward a mineral-based industrial one. The non-metallic minerals, namely “stone quarries and clay and sand pits; chemical and fertilizer mineral deposits; salt deposits; deposits of quartz, gypsum, natural gemstones, asphalt and bitumen, peat and other non-metallic minerals other than coal and petroleum” (OECD, 2001), are the most extracted natural resources in OECD countries today, with the fastest growth in the last 15 years (OECD, 2019). They are also requested by various economic sectors, including 1) food and nutrition; 2) mobility; 3) the built environment; and 4) energy.

Non-metallic minerals can be difficult to categorise due to their common mixtures and unclear end uses across various sectors, including industry, construction, and agriculture. To address this challenge, the IRP in 2019 proposed a general classification system designed to clarify the dis-

tinct characteristics and potential applications of these minerals (Table 1).

The demand for construction minerals is ubiquitous on a global scale. However, their international trade is limited due to the prevalence of domestically available minerals. The economic value of construction minerals per unit of mass is low and their large volume makes it difficult to transport them over long distances. Although construction minerals are considered non-toxic, with low specific impact on the environment compared to other materials such as industrial minerals or metals, they are often transported by road, causing significant CO₂ emissions. Additionally, other indirect emissions are triggered, mainly due to industrial processes fed by those materials, as in cement production (Winnefeld, 2022), or caused by their extraction as land use, air, water, and loss of biodiversity (Xuhui et al., 2024), even in terms of unused extracted material abandoned in quarry sites.

As this consistent rate of construction minerals unused is progressively growing, we should adopt strategies to promote material efficiency, including but not limited to reduced use of materials through design, material substitution, recovery, reuse, production and lifetime extension (Hertwich et al., 2019; Worrell et al., 2016). This can help reduce environmental impacts from natural resource extraction and consumption, addressing scarcity across various fields (UNEP, 2017). It also contributes to enhancing the decoupling of resources and impacts. (UNEP, 2024).

1.3.1 Non-metallic minerals extracted in Italy and related environmental pressures

According to data from the IRP’s online database (2019), the main categories of natural resources extracted and consumed in Italy are biomass, fossil fuels, non-metallic minerals and metal ores. In Italy, in 2024, the biomasses extracted (DE) accounted for 123,7 Mt, imported (IMP) for 54 Mt and exported (EXP) for 21 Mt; the fossil fuels extracted for 8,7 Mt, imported for 210,9 Mt, exported for 3,1 Mt; non-metallic minerals extracted for 279,7 Mt, imported for 13,5 Mt, exported for 13,6 Mt; metal-ores extracted for 0,0 Mt, imported for 13,6 Mt and exported for 31,4 Mt. It is evident that, except biomasses and non-metallic minerals, the Italian country depends on imports.

According to IRP classification in 2019, non-metallic minerals are divided into 1) ornamental building stones; 2)

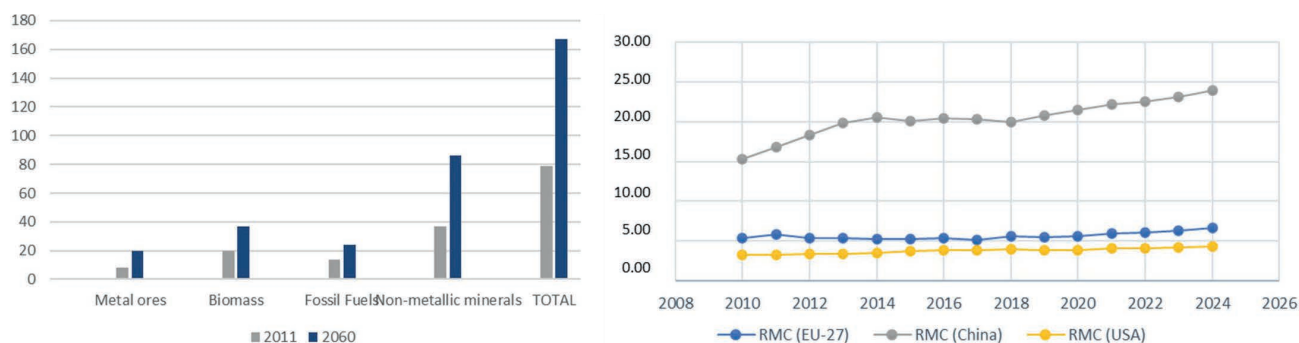


FIGURE 1: a) Consumption rates in 2011 (in Gt) and expected for 2060 (source: authors, based on OECD forecasts); b) Raw material consumption (in Gt) between 2010 and 2024 (source: authors, based on IPR’s flow material database).

TABLE 1: IRP's Domestic extraction (DE) classification system of non-metallic minerals (source: IRP, 2019. Adapted by authors).

Category)	First level	Second level
Non-metallic minerals	Ornamental or building stones	
	Carbonate minerals essential in cement	Chalk0
		Dolomite
		Limestone
	Chemical and fertilizer minerals	
	Salt	
	Gypsum	
	Clay	Structural Clay
		Specialty Clay
	Sand and gravel	Industrial sand and gravel
		Sand and gravel for construction
	Other non-metallic minerals	

carbonatic minerals used in cement production; 3) mineral and chemical fertilisers; 4) salts; 5) gypsum; 6) clays for ceramic industries; 7) sands and gravels. By considering only non-metallic minerals for the construction sector, in 2017, according to the available ISTAT data, the total DE accounted for 149,10 Mt. The aggregate's biggest class by weight includes "limestone, travertine, gypsum and sandstone" which accounts for 68.8 Mt; in second place are "sands and gravels", with almost 59 Mt; the extraction of "porphyry, basalt, tuff and other intrusive rocks" amounts to 9.1 Mt, of which 43.9% is basalt (volcanic rock used in building constructions, road works, etc.); still, the extraction of "cement marl" reaches 6.5 Mt (increasing by 8,8%), as well as "ceramic and industrial minerals", mostly feldspar for the glass and ceramics industry, reaches 4.4 Mt. Lastly, the construction stones extracted, such as marble and travertine reaches over 2 Mt and 500,000 tonnes of granite.

The extraction of non-metallic minerals often requires the removal of large rock masses, which can significantly impact the environment and the surrounding areas of the quarry. This process can lead to issues like pollution, waste, and the gradual depletion of increasingly scarce natural resources. Additionally, it poses threats to surface waters, biodiversity, the landscape, territorial integrity, and public health. Modern survey technologies with high spatial and temporal resolution can assist in mapping the territorial impacts related to excavation and tracking their spatial evolution over time. For example, the Italian Higher Institute for Environmental Protection and Research (ISPRA) has developed a GIS-based monitoring system called GEMMA. This system provides up-to-date and detailed information about various quarry sites across the country, using a set of indicators related to thirty-nine thematic categories. Regional governments and companies contribute to the database update through periodic surveys, enabling better monitoring and management of the quarry's environmental pressures. (Fumanti and Di Leginio, 2023).

A total of 3,316 quarry sites were recorded in Italy in 2021, including both active and inactive sites. A total of 2,158 active sites contribute to the overall amount of domestic extraction (DE). This includes 571 sites in the north-

west, with 288 located in Lombardy alone; 209 sites in the north-east, of which 146 are in Veneto and 186 in South Tyrol; 456 sites in the central regions, including 224 in Tuscany; and 349 sites in the southern regions, which comprise 141 in Apulia, 178 in Sicily, and 95 in Sardinia. There are also 553 inactive sites, distributed as follows: 102 in the north-west, 94 in the north-east, 135 in the central regions, 169 in continental southern areas, and 53 on the islands. Data is unavailable for the remaining 605 sites.

Various statistical indicators have been developed to monitor the extraction of material resources. Some of these indicators are specifically designed to analyse environmental pressures and to describe and interpret the interactions between the economy, land, and the environment. The National Statistical Authority (ISTAT) releases annual data and calculates the Extraction Intensity (IE) and Density of Active Mining Sites (DSE) for non-metallic mineral resource withdrawals in Italy (Figure 2).

The IE index expresses the quantity of mineral resources extracted per square kilometre, and it is available at a regional scale; the DSE indicator records the ratio between the number of active mining sites per municipality and their surface areas. What emerges from the survey is that in Italy the average extraction intensity (IE index) increased by 20,6% between 2017 and 2021, with some difference among the sub-areas (north +20,4%, centre +13%, continental south +25,6%, Sicily and Sardinia + 28,7%.

Two main issues impact the current situation in Italy and other places. Firstly, there is a need for more effective and environmentally friendly management of active quarries, closely linked to the specific social and territorial contexts of quarries; this emphasizes the importance of reducing and valuing quarry waste using high-efficiency extraction technologies; additionally, it calls for a well-planned and progressive approach to environmental recovery, which should be carried out alongside the exploitation of quarry resources from the very beginning. Secondly, the huge amount of quarry waste, temporarily stored in inactive or abandoned sites, must be addressed first, as it contributes to landscape changes and environmental degradation. This can lead to soil loss, harm to biodiversity,

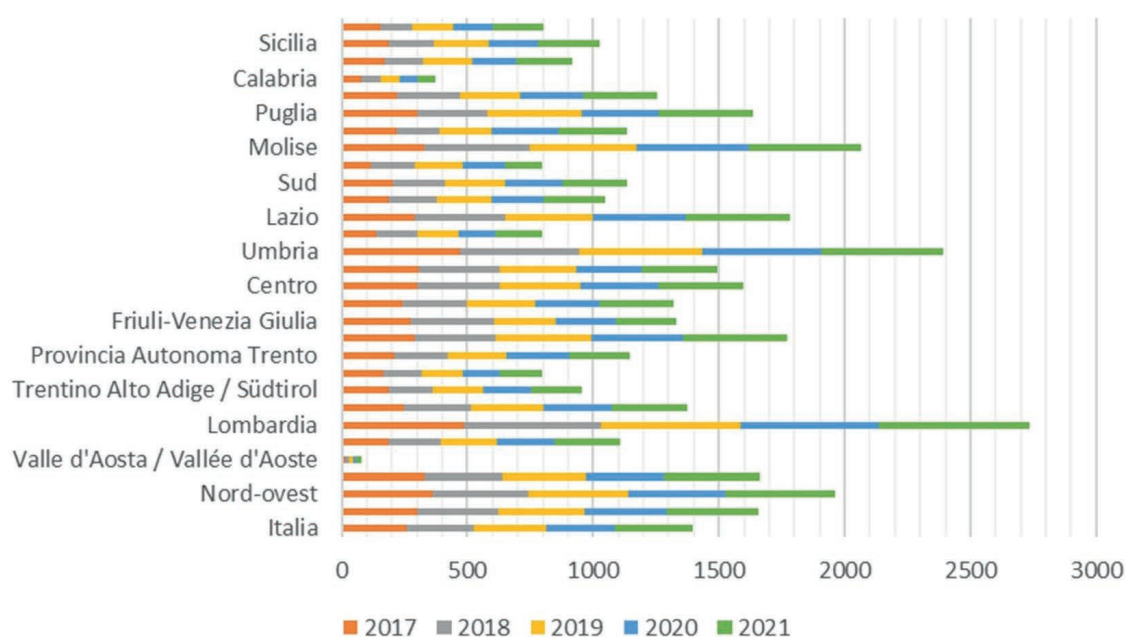


FIGURE 2: Intensity Extraction (IE) in Italy, in m³/km² (Monsù Scolaro A., based on ISTAT's data).

and alterations in underground water, all of which seriously affect the surrounding areas and communities. Waste and scraps can be managed to either reshape the site or be repurposed as secondary raw materials; additionally, these abandoned sites could be transformed into historical parks that hold cultural significance and are open for community use and public enjoyment.

1.4 Granite quarries in Sardinia: a research and development project to valorise abandoned scraps and sites

A comprehensive overview of quarries and mines in Sardinia had been provided by the Regional Plan for Extractive Activities in 2007 (PRAE, 2007), which recorded 397 active quarries, 216 decommissioned (or inactive) ones, and 644 abandoned quarries called "historical" since established before L.R. 30/89. However, as per the most recent ISTAT survey conducted in 2021, the number of active quarries has decreased to 135, of which 95 are productive and 11 not in operation, while the status of the remaining quarries is unknown.

The primary resources extracted in Sardinia are clay, limestone, sand, granite, porphyry, and basalt. From 2017 to 2021, the extraction of non-metallic materials increased by 34%, reaching 134,606 tons in 2021, of which marble 29% to 32%, totalling 2,680 tons; sand 20%, with 1,863 tons; granite 20%, with 1,803 tons; 15% porphyry, totalling 1,395 tons; 13% basalt, with 1,189 tons; and clay, which made up around 2% with 169 tons.

The current regional plan for special waste in Sardinia, in force from 2021 (PRGRS, 2021), highlights significant issues stemming from the production system's insufficient capacity in effectively managing and utilising both before and post-consumption waste materials. The almost total lack of recovery and recycling facilities in the Region is identified as the main cause of this issue. Based on the anal-

ysis carried out, the Plan proposes a survey campaign to identify conditions of environmental degradation linked to waste production. This requires collecting and quantifying waste according to its CER code (European Waste Code) to define potential site-specific management and exploitation scenarios at the territorial scale. Also, due to the precious and very sensitive Sardinian natural landscape, extraction activities cause significant environmental alterations, exacerbated by the accumulation of waste, which affects many areas within the regional territory (Figure 3). The recently adopted new building regulations allow the use of scrap to fill voids from any land excavation, even by moving bulky waste from one site to another, if they are geochemically homogeneous and compatible with the ecosystem.

About one-fifth of Sardinia's territory is characterised by a geological substrate of intrusive granitoid rocks known as the "Sardinian-Corsican Batholith." This formation originated during the Hercynian orogeny and has offered significant economic opportunities to the four main quarry basins where it is found (Buddusò and Alà dei Sardi; Calangianus, Priatu and Bassacutena; Arzachena and Luogosanto; Gavoi and Ovodda), which have been extensively mined for resources.

In 1997, a regional decree grouped the first three areas into the "industrial district of Gallura granite", marking it as the largest Italian extraction area by territorial extent. The district once included 114 quarry sites, 30 of which were in Buddusò and Alà dei Sardi municipalities in Sassari County.

The ISTAT data show the relevance of this basin at the national scale: among 3,911 tons of granite excavated in Italy in 2021, about 2,200 tons are of gneiss, while 88.30% of the remaining 1,700 tons of real granite are extracted in Sardinia.

This data must be located within the general trends of Italian whole granite production, which was 2.06 million tons in 2011 but decreased by 93% in 2023, totalling

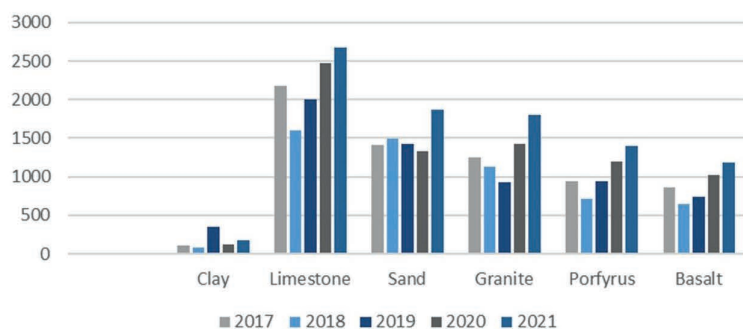


FIGURE 3: a) The non-metallic minerals for construction most extracted (in tons) in Sardinia (Monsù Scolaro A. based on ISTAT's data); b) Landscape alteration by granite quarry activities in Sardinia (Monsù Scolaro A.).

just 143,129 tons. In contrast, the production of blocks and sawn slabs increased by 13%, rising from 474,374 to 534,950 tons during the same period. This growth highlights innovations in extraction processes and a reduction in scrap. The quality in this industry has always relied on the efficiency of the extraction process from the “banks” and quarry faces.

Until the recent past, only 30-40% of the extracted volumes were converted into marketable material, while the remaining became waste, including blocks of various sizes, processing residues, and sludge. Today, diamond wire cutting has gradually replaced the extraction by explosives to help in reducing the waste and its accumulation in mining areas over the years. This reduction is crucial, as the excessive build-up of debris has often strongly limited the productivity of companies by restricting the available space for storing extracted materials while deeply altering landscapes and territories.

1.4.1 Methodological approach

Aiming at finding possible answers to this challenging scenario, the Department of Architecture, Design, and Urban Planning at the University of Sassari carried out a research and development project, entitled “Research for Constructive Solutions for the New Use of Stone Waste and Ceramic Materials for Circular Architecture”. This project took place from 2021 to 2023, having been funded by the Italian Ministry for Enterprise and Made in Italy as part of the “Smart Fabric” initiative. Its primary objective was to explore the potential of granite waste as a secondary resource for several sectors, namely the ceramic or the construction industry or other industrial applications, even by depicting possible production chains based on an industrial symbiosis approach. Furthermore, the project focused on identifying potential methods to regenerate and reuse abandoned quarry sites. The methodological approach, which involved various areas of expertise within a multi-disciplinary framework, was developed according to the following points:

- map sites by using available technical cartography to differentiate between active, inactive, and dismissed quarries and additionally to identify the quantity of granite scrap and waste in each quarry, along with its condition of access, sourcing and withdrawal;

- laboratory analysis of samples of granite residues to determine their chemical and mineralogic composition to assess their suitability for use as secondary raw materials in the ceramic industry or for other applications;
- laboratory analysis of granite scrap and waste by following common market requests and uses, such as for flat surfaces (sawing machine plans), glossy and rough finishes, to evaluate the modifications of thermal reflectance and absorption after applying nano-paints;
- mechanical analysis of granite thin slabs obtained by cutting granite scraps to find the maximum strength by balancing thickness and planar dimensions, enabling the creation of the largest possible stone slab for designing innovative ventilated facades for smart buildings;
- laboratory analysis to check light flow absorption by previous thin granite slabs.

Additionally, we carried out:

- an analysis of dismissed quarry sites from an ecosystemic point of view for identifying alterations and residual natural characteristics to establish the basis for developing potential methods to regenerate abandoned areas for public use and make them accessible to the community;
- a study of industrial and economic territorial conditions, as industrial areas equipped with technological resources, transportation, storage, and conveying systems, reduced VAT, to explore and enhance strategies for managing scrap and waste through industrial symbiosis approaches.

This last is a crucial step toward activating sustainable and systemic processes to address the challenges presented by abandoned sites, which hold significant untapped potential.

However, the findings of the last two points will be summarised to provide a brief overview of the systemic approach adopted to address the issue of abandoned quarries and demonstrate how waste can be transformed into local opportunities on a territorial scale.

2. RESULTS AND DISCUSSION

The mapping activity allowed updating the usage status of the quarry sites by using a cartographic survey,

comparing the current boundaries of the quarries with the topographical ones already authorized, extracted from the regional plan of mining activities released in 2007 (PRAE, 2007). With the aim of valorizing waste and abandoned sites, eighteen quarries have been selected and classified based on five key logistical criteria:

- Current state of use (active, suspended, decommissioned, or already restored);
- Presence and visual assessment of waste volumes, including any possible lithological variations within the waste;
- Accessibility of waste;
- Availability (on-site or near-site) of suitable machineries for collecting and transporting waste;
- Current conditions of the access route to the quarry and distance from the processing plant (owned by the company EURIT Srl) operating in NE-Sardinia, which carries out pre-treatments (crushing, grinding and deferrization by magnetic separation) of raw materials for ceramics.

The laboratory analysis was conducted on the waste samples collected at each abandoned quarry site, identifying two main lithological varieties. The samples were then crushed, granulated, milled and micronised to perform mineralogical analysis by X-ray diffraction (Rietveld method), chemical analyses (by Inductively Coupled Plasma-Mass Spectrometry), and thermogravimetric (TG) analyses. The classification of the samples according to the Streckeisen QAP diagram is reported in Figure 4 (Cerri et al., n.d.). All samples are monzogranites, except for those that correspond to granodiorites and two which have intermediate compositions.

The chemical composition of the materials was compared with that of conventional quartz-feldspathic fluxes used in the production of porcelain stoneware tiles (de'Gennaro et al., 2003), to evaluate whether and which waste can be used as a component in ceramic bodies. From a technological standpoint, the main issue concerns

the iron content, which leads to dark colouration in the fired materials, as observed in the powders of several samples after TG analysis at 950°C (Cerri et al., n.d.). This problem impacts many of the sampled wastes, often compromising their potential reuse in ceramics. Still, lithotypes with favourable chemical and mineralogical characteristics may also require a deferrization treatment.

Considering the logistical aspects assessed for the eighteen quarries and the technological features determined for the twenty waste samples, the investigation focused on three sites: BES, OLU and Senè 1, that is a quarry next OLU containing the same lithotype. The size and abundance of the waste blocks contained in these sites were determined by carrying out a survey using a drone, a satellite system (GPS Top Con Base+Rover) and processing the data with the Meridiana 4.0 software (Geopro.it). In the BES site, 70% of the waste consists of blocks of size $\geq 2 \text{ m}^3$, unlike OLU and Senè 1 where, respectively, 72% and 78% of the blocks have a volume less than 2 m^3 (Figure 5; Cerri et al., n.d.). The volume occupied by waste in the three quarries is estimated to be: BES $\approx 116800 \text{ m}^3$; OLU $\approx 132400 \text{ m}^3$; Senè 1 $\approx 69800 \text{ m}^3$. The tonnages were calculated using an average density for granitoid rocks of 2.7 t/m^3 and assuming a void volume between 15% and 30%. The resulting estimated tonnages are as follows: BES 268-221 kt; OLU 304-250 kt; Senè 1 160-132 kt (Cerri et al., n.d.). Overall, these three sites alone contain at least 0.6 Mt of resources potentially usable as raw materials for ceramics.

Subsequent laboratory analyses were conducted to evaluate the changes in the samples' solar reflectance and thermal absorption. The tests were conducted on a commercially valuable sample of granite known as "pearl grey." After obtaining the reflectance measurements for the three primary colours of the test piece, we formulated three solutions: one that was purely polymeric and two that were hybrids of metal oxide (MO) and polymer at varying concentrations. Preliminary reflectance measurements indicated that the hybrid solutions resulted in a 40% average increase in reflectance while still partially revealing the underlying stone pattern. Subsequently, based on scientific literature, a general coating was chosen to develop a specific coating based on PDMS (polydimethylsiloxane; Sylgard 184) and titania nanoparticles (TiO_2 ; P25) to optimise the reflective effect of solar radiation and the hydrophobic, self-cleaning and photocatalytic effect of the surface. The coating was characterised by infrared spectroscopy. Being the titania of white colour, several mixing ratios were evaluated with PDMS (transparent) to preserve the pattern of the stone materials without sacrificing the functionality of the coating. The different mixing ratios were applied and tested on the surface of stone materials using an infrared thermometer to evaluate the reflectance of solar rays. Finally, measurements were made with a thermal imager to evaluate the possible variations in the thermal behaviour of the different ratios of the PDMS/ TiO_2 coating. The sample was monitored in three different phases to simulate real operating conditions, namely:

- minimum overheating;
- following heating after high sunshine;
- during cooling.

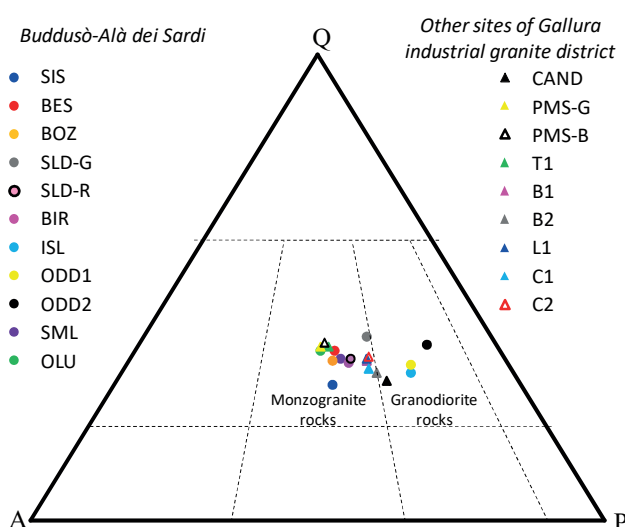


FIGURE 4: Classification of the sampled rocks (Cerri et al., n.d.). Q: quartz; A: sum of Na- and K-feldspar; P: plagioclase.

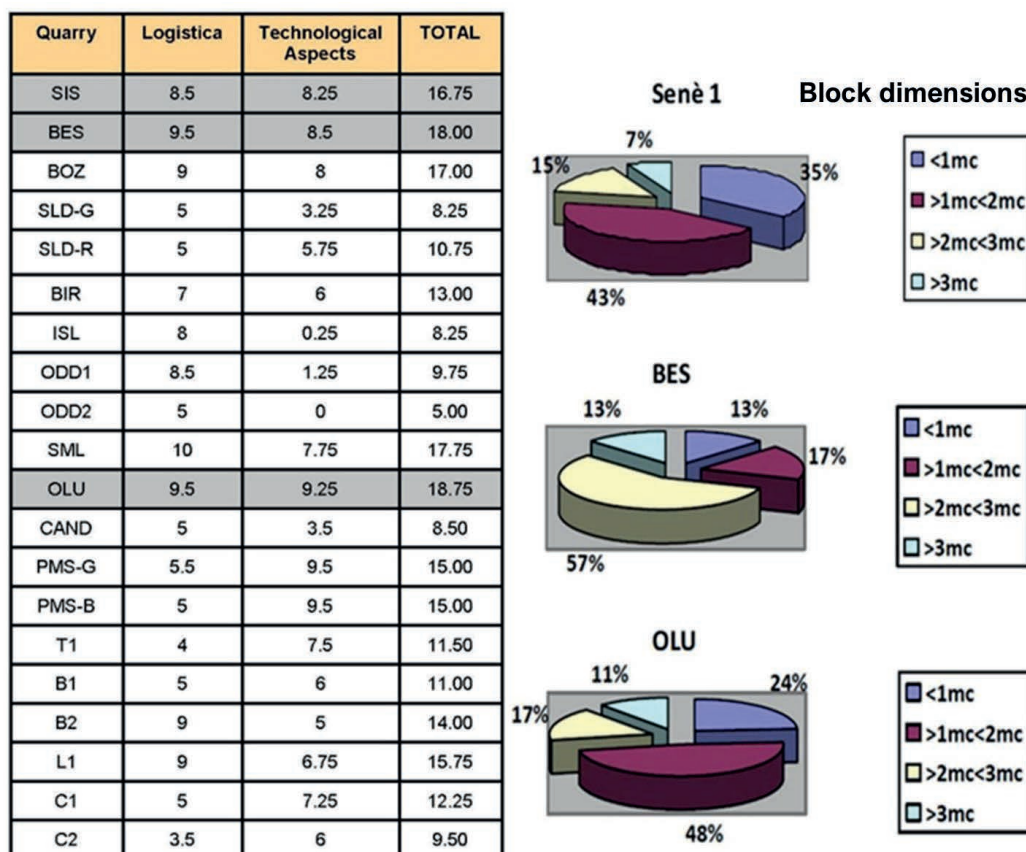


FIGURE 5: a) Final ranking based on the total number of quarries analysed, highlighting (in grey) the three most cost-effective to exploit; b) scrap's dimension of the three quarry most cost-effective (source: Cerri, G. 2023).

The tests were carried out at the end of July to simulate real operational conditions. Finally, in a second thermographic survey, in addition to the PDMS/TiO₂ compound, also tested on polystyrene cans, other types of coating were observed on stone waste: 1) TiO₂; 2) SiO₂; 3) Boric acid; 4) Calcium carbonate (CaCO₃). The various samples, numbered 1 to 6, were created by adjusting the mixture ratio of PDMS to TiO₂, ranging from 4:1 to 4:0.025. In the minimum overheating phase, a rapid increase in temperature is observed on test piece no. 4 (only PMDS) and, to a lesser extent, n.3. The specimens heat up unevenly, perhaps because of the different thickness of the paint layer, while some specimens heat up more quickly during the higher heating phase as showed in Figure 6.

The PDMS acts as a thermal emitter, accumulating infrared radiation and then releasing it in all directions, thus heating both the underlying stone layer and the external environment. Hence, the need to adopt a double emitter/reflective layer to avoid the transmission of heat to the underlying material without altering the natural stone's layer emerged. A second thermographic survey was carried out comparing the PDMS/TiO₂ mixture with other types of compounds, including calcium carbonate (CaCO₃). Scrap coated with two different concentrations of calcium carbonate (CaCO₃) showed excellent reflective properties and indeed the lowest surface temperatures of scrap, both at high and low concentrations.

A mechanical analysis was carried out on thin granite slabs to determine their suitability for use as ventilated cladding on buildings. Firstly, the thin plates - 60 cm by 60 cm and 120 cm by 60 cm according to market requirements - were obtained from a waste block of "pearl grey granite", cut with a special machine equipped with ultra-thin diamond wire, with a diameter between 0,35 and 0.6 mm, not existing in Sardinia. To perform the load test, was consulted the norm UNI 11018-1:2023 "Ventilated Facades - Part 1: Performance characteristics and terminology. The slab was supported along its perimeter by a 1 cm wide metal frame. The load was evenly distributed through nine equi-spaced air pistons (each piston has a thrust area of 12.56 cm²). The piston thrust area has a thin rubber coating to ensure the uniform application of force. The load application is monitored using a pressure gauge on the compressor and a load cell. Plate failures are measured using a millesimal comparator (displacement sensor) positioned near the center of the plate. The load was applied uniformly until failure occurred at a rate of approximately 40 kg/min. The load was applied by simulating the wind load on a ventilated facade slab. Six load tests were carried out on 60 x 60 cm and 120 x 60 cm slabs; three further tests were carried out by applying the load in the middle of the slab as in Figure 7.

The results obtained are shown in Table 2.

The results show that the maximum strength does not always increase directly as thickness, even when minimal,

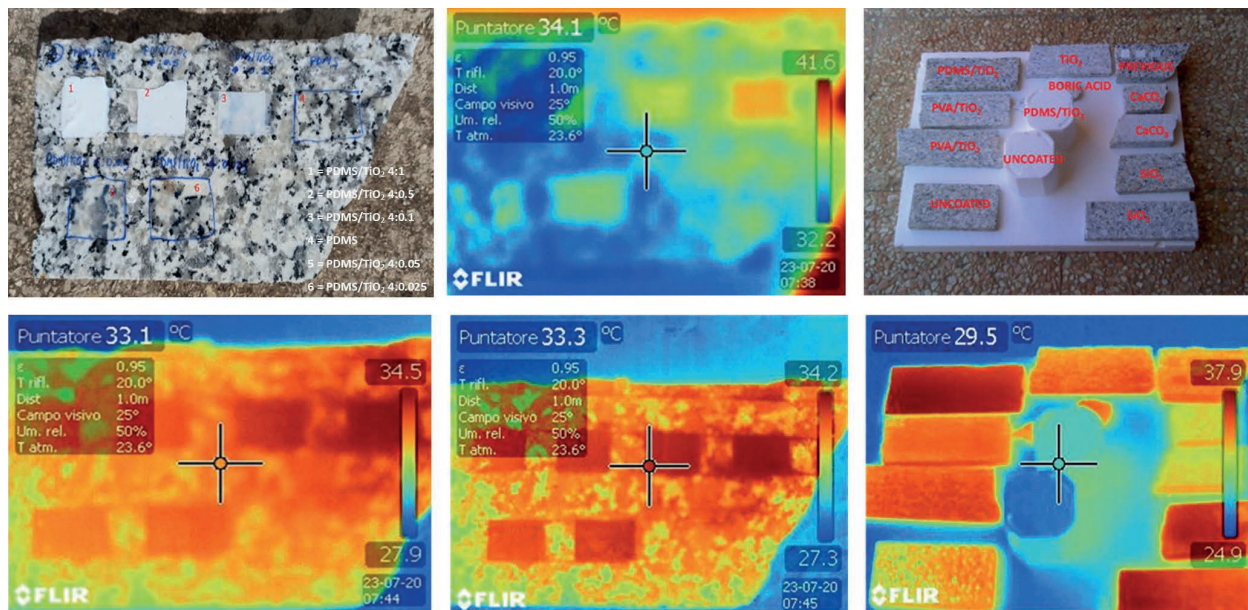


FIGURE 6: top left) the six different coating experimented; top central) minimum overheating; down left) heating after high sunshine; down central) cooling phase. Top right) samples for second thermographic survey and down right) thermal emissivity (source: Malfatti, L. 2023).

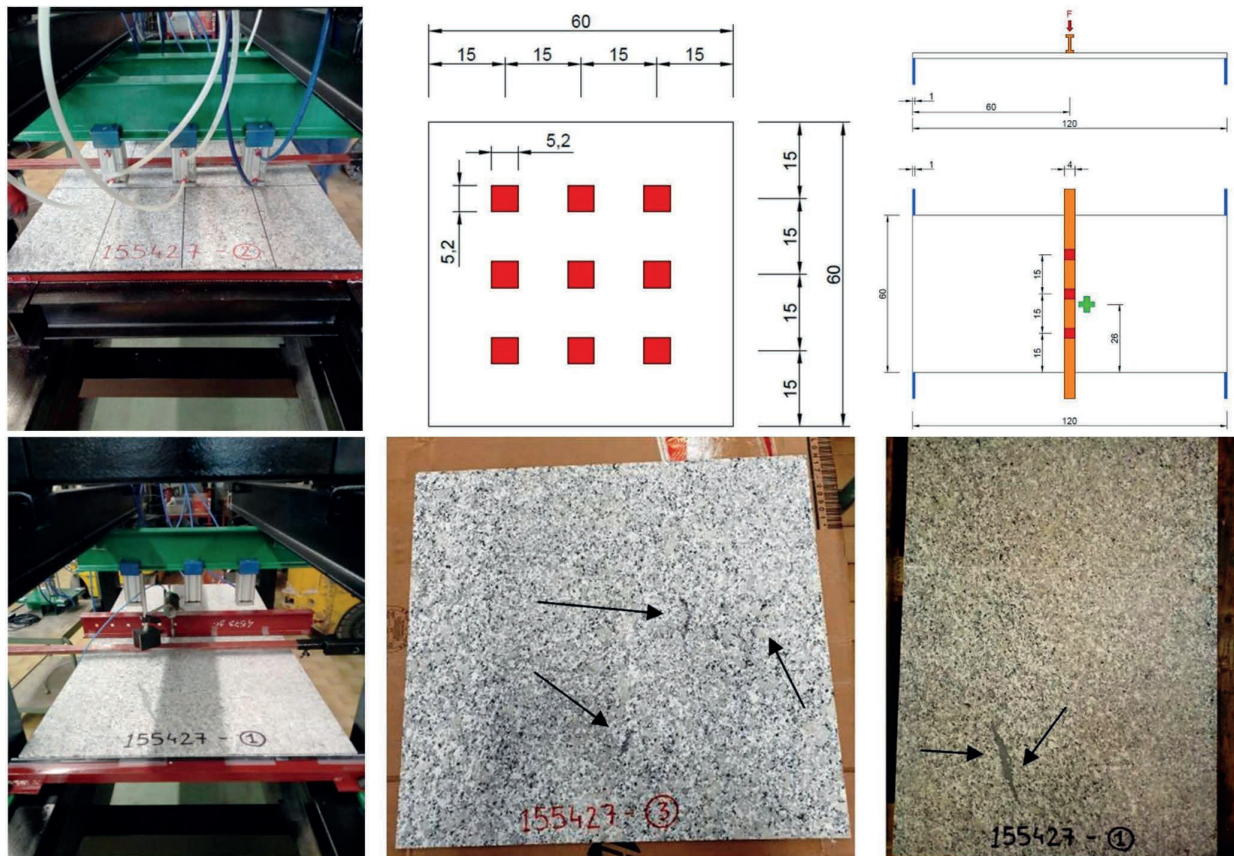


FIGURE 7: top and down, left) three-axis and six-point loading machine; top central) six-point loading scheme on the slab 60x60cm; top right) three-axis loading scheme on the slab 60x60cm; down central and right) arrows indicating defects in the slab (source: Veneta Engineering, 2023).

increases. The 120 x 60 cm slab has the highest breaking load resistance for the lowest thickness, as well as for the 60 x 60 cm slabs. The mechanical behaviour of the slabs,

which is not always linear, can be attributed to irregularities and the presence of imperfections in the stone matrix, due to the smallest iron-micaceous portions (see the bottom

TABLE 2: Load test results (source : Veneta Engineering, adapted by authors).

Dimension (cm)	Slab thickness (mm)	Piston (nr.)	Maximum breaking load (Kg)	Maximum Flexion (mm)
60 * 60	4,96	9	83,43	7,662
	5,05	9	73,53	6,480
	5,21	9	82,35	7,471
120 * 60	5,72	9	109,53	7,828
	5,73	9	103,05	7,451
	5,76	9	100,53	7,541
120 * 60	5,60	3	11,96	12,240
	5,70	3	9,23	14,358
	5,80	3	11,78	12,478

right side of the Figure 7). Ultimately, the maximum flexion occurs at the smaller thickness, as expected. Furthermore, when a three-piston load is applied, both the breaking strength decreases significantly and the deflection at the mid of the slab increases considerably. The non-linear mechanical behaviour of the slabs was considered due to irregularities and the presence of small imperfections in the stone matrix with the smallest iron-micaceous portions.

Finally, the last test was performed to measure the light flow absorbed by a thin granite slab of “granite grey pearl”, dimensions 30 x 30 cm. The test was conducted in a dark room with dimensions of 50 x 30 x 30 cm. Inside the room, slabs of pearl grey granite measuring 300 x 300 x 5 mm were placed (Figure 8). The following equipment was used during the test:

- a LED light measuring 25.5 x 22.9 cm, with a luminous flux of 2,930 lumens.
- a HOBO data logger sensor capable of measuring illuminance in lux.
- a luxmeter (illuminance meter) to measure light levels in lux.
- a plate housing system made of thin black sponge profiles, positioned in the middle of the chamber.

The results in Table 3 show that the medium value of the light density absorbed by the slab, is approximately be-

tween 99,66% and 99,78%, but the effect obtained over the slab could be interesting for design scopes.

3. CONCLUSIONS

In the Buddusò's granite district, there are currently only 5 active granite quarries out of the 42 that were operational in the 1970s. Production is focused primarily on the big granite blocks, sold or cut into slabs, depending on market demand. Over the years, the old extraction techniques have caused huge amounts of abandoned granite waste, now significantly altering landscapes and ecosystems. These scraps are generally used to backfill the roadbed or employed as breakwaters, or reduced in size for secondary building applications, like outdoor pedestrian pavements. However, the research has shown that these waste materials hold remarkable residual performance - both mechanical and aesthetic - promising product innovations in line with circular economy approach and decoupling of resources and impacts. If properly sampled, these wastes could be repurposed as secondary resources for the ceramic industry or innovative products in the construction sector. The thin slabs for ventilated facades, obtained from granite blocks, treated with paints to improve thermal behaviour, need further tests in a climatic room or through prototypes, to evaluate the thermal performance in a real

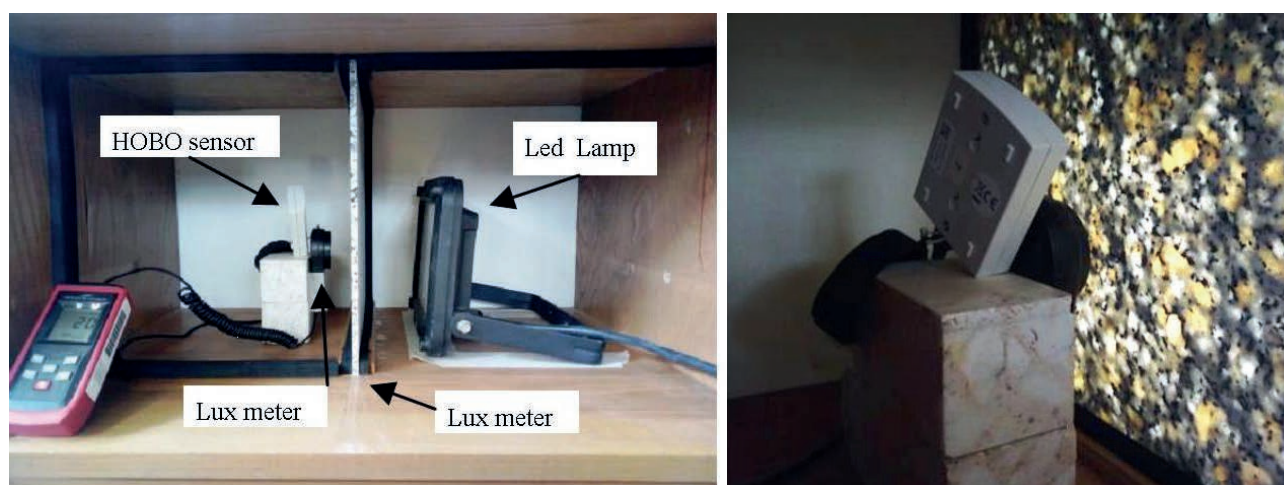
**FIGURE 8:** Lighting test room (source: Veneta Engineering, 2024).

TABLE 3: Illumination test results (source: Veneta Engineering, adapted by authors).

Test	Illumination without slab (lux)		Illumination with slab (lux)		Light absorbed (lux)		Light absorbed (%)	
	E _{min}	E _{max}	E _{min}	E _{max}	ΔE _{min}	ΔE _{max}	ΔE _{min}	ΔE _{max}
1	22.345,8	26.595,9	59,1	74,9	22.270,9	26.536,8	99,66	99,78
2	22.570	27.620,5	59,1	74,9	22.495,4	27.561,4	99,67	99,79
3	23.245,5	27.400,3	59,1	67,0	23.178,5	27.341,2	99,71	99,78
Medium value					22.648,3	27.146,5	99,68	99,78

environment. The use of the slab without additional reinforcement layers would favour end-of-life reuse.

Furthermore, the existence of equipment, storage facilities, processing machinery, and transportation options, along with favourable logistics and special economic zones, is essential for activating potential production chains in industrial symbiosis. Achieving this goal requires a comprehensive, interdisciplinary approach bearing in mind multiple territorial implications - socio-cultural, economic, and environmental- through an active involvement of local communities, administrations, and economic stakeholders to initiate ecosystem rebalancing and social revitalisation processes. Additionally, this approach should aim to redevelop landscapes and restore soil and natural habitats, enhancing their environmental value (OECD, 2021). In this view, the integration of digital technologies, environmental resilience, and a redefined role for humans in industrial processes could potentially lead to a reduction in waste and environmental impact, concurrently enhancing social justice and sustainability (Mondejar et al., 2021).

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MICRO AND NANOPLASTICS IN TERRESTRIAL MAMMALS: CURRENT KNOWLEDGE AND RESEARCH GAPS

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ABSTRACT

Micro and nano plastics (MNPs), originating from environmental pollution, consumer products and industrial processes, are being detected increasingly across numerous ecosystems and food chains. Their pervasive nature elevates the possibility of human and animal exposure through ingestion, inhalation and dermal contact, underscoring the need for scientific investigations into their presence and effects on "One Health". With a view to consolidating the current state of knowledge, identifying similarity between humans and animals, research gaps and paving the way for evaluating the potential toxicological and health implications of these particles, a crucial systematic literature review has been conducted to study the detection of MNPs in specific mammalian (human and animal) tissues, whilst also examining experimental models assessing their toxicological effects. By synthesizing findings across biomedical and environmental research, the review elucidates the pathways through which these particles enter and accumulate in the body. Furthermore, it highlights the potential health risks posed by chronic exposure, including inflammatory responses, oxidative stress, and tissue damage. However, the current evidence remains insufficient to establish a quantitative balance across exposure routes and body tissues due to the lack of harmonized exposure data, heterogeneity in methodologies, inconsistent reporting, and lack of biological/clearance data. Recognition of this constraint is crucial to a more accurate contextualization of current findings, which is key for the purpose of shaping public health policies and regulatory frameworks to mitigate exposure to microplastics. Given the interdisciplinary nature of the topic, the review bridges diverse fields to create a robust foundation for future research.

1. INTRODUCTION

The presence of microplastics in the environment has caused growing concern at all levels of society, ranging from local and regional to global. These contaminants have been detected in numerous environmental matrices including air (Allen et al., 2019; Beaurepaire et al., 2021; Truong et al., 2021), water (Horton et al., 2017; Altuğ & Erdoğan 2022; Gong et al., 2023; Poli et al., 2024), soil (Huerta Lwanga et al., 2016; Kanteraki et al., 2022), organic amendments (Colombini et al., 2022; Edo et al., 2022; Öling-Wärnå et al., 2023; Porterfield et al., 2023) and the food chain (Huerta Lwanga et al., 2017; Oliveri Conti et al., 2020).

Terrestrial mammals have become increasingly exposed to microplastics through ingestion and subsequent crossing of the digestive tract epithelia, inhalation through the respiratory tract and dermal contact through the cutaneous layer.

Based on dimensions, plastics are broken down into mesoplastics (5-25 mm), microplastics (0.001-5 mm), and nanoplastics (10⁻⁶-0.001 mm) (Bandini et al., 2022; EU Commission. 2023; Gigault et al., 2018; Meegoda & Hettiarachchi 2023). Each category is associated with specific characteristics related to environmental fate, uptake, and health risk assessment. Indeed, based on their physical characteristics (particle size, surface chemistry, cell type and biomolecular corona), once meso-, micro- and nano-plastics (MNPs) enter the body by any route, they are able to move, break and establish themselves across a series of organs (Barceló et al., 2023).

Following deposition, accumulation of ample quantities of micro- and nanoplastics throughout a range of tracts could impair functioning of the affected organs. MNPs analysis presents unique challenges for detection and quantification, including particle sizes, diversity in polymer types that require specific detection techniques, and



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the alteration of physical and chemical properties by natural weathering or interference from other substances. The relatively low concentrations of MNPs present in terrestrial, as compared to environmental, matrices necessitate the use of highly sensitive testing methods, added to the potential interference from background contamination. To counter this, approaches such as the Limit of Quantification (LOQ), a contamination adjustment approach used to reliably quantify the lowest concentrations with acceptable precision and accuracy under the required experimental conditions, are applied (Leonard et al., 2024).

Additional variables such as age and gender exposure patterns, are of particular interest as they tend to influence the intake and accumulation of MNPs in organisms (Jenner et al., 2022; Zhu et al., 2024). with specific population groups such as children and the elderly proving potentially more vulnerable to their potential effects. Fetuses start to accumulate MNPs from the mother's blood whilst still in the womb, with their weak detoxification mechanisms potentially resulting in an increase likelihood of exposure (Chen et al., 2023). Likewise, the elderly are rendered more vulnerable due to their reduced immunity and weaker gut barrier and lung epithelia (Fulop et al., 2018). Based on their diverse metabolisms, the impact produced on male and female mammals may differ, thus impinging on the retention and accumulation of MNPs (Björvang & Mamsen 2022; Dominelli et al., 2018; Gade et al., 2021).

Although numerous studies have documented MNP contamination in marine organisms (Cole et al., 2013; Li et al., 2018; Mistri et al., 2022; Moreira-Mendieta et al., 2023; Yin et al., 2022), evidence in terrestrial mammals remains limited and scattered. In this context, a systematic literature review providing for the collection, analysis and comparison of current data relating to the presence of MNPs in human and animal organisms is essential to consolidate the current knowledge on this topic. Accordingly, the present review was undertaken with the aim of: (i) identifying the main sources and exposure pathways of MNPs in mammals; (ii) evaluating the distribution of MNPs across different tissues; and (iii) comparing findings in humans with those from studies on domestic and wild animals, highlighting similarities and differences in bioaccumulation patterns. This comparison may help to determine whether animals can serve as sentinel models for human exposure and to better estimate potential health risks.

Furthermore, the review was intended to help identify any major methodological limitations and research gaps, providing clear directions for future investigations and supporting the development of standardized protocols. The ultimate goal will be to promote a One Health approach that acknowledges the interconnectedness of environmental, animal, and human health in assessing the risks associated with MNP contamination.

2. METHODOLOGY

This systematic review focused on identifying studies that detected MNPs in terrestrial mammals. The search was divided into two parts separating the studies on microplastics (MPs) in humans from other terrestrial mam-

mals. PubMed was used exclusively for the human-focused search as it is powered by MEDLINE, a bibliographic database that contains reliable peer-reviewed articles specialized in biomedicine and health sciences. The search strategy involved combining the terms "microplastic", "human" and 13 target tissues, resulting in the following query: "(Microplastic) AND (human) AND (tissue OR thrombi OR artery OR placenta OR lung OR gut OR blood OR kidney OR brain OR ovary OR breast OR colon OR semen)". On the other hand, PubMed, Web of Science (WOS) and SCOPUS were employed to gather relevant literature on other terrestrial mammals, as both databases provide broad coverage of high-quality peer-reviewed publications. A comprehensive search query was formulated and applied within the field of Title, Abstract and Keywords. The search terms consisted of: "microplastic" OR "nanoplastic" AND common names of 48 animal species spanning 10 of the most common taxonomic orders of terrestrial mammals as seen in Table S1 in supplementary materials.

The search was performed using the 'all fields' option and was limited to original articles published in English. No time restrictions were applied. Review articles, conference proceedings, books, and publications deemed irrelevant or inadequate for data extraction were excluded during the screening process.

Data regarding key findings, sample size, plastics characteristics and origins, as well as laboratory techniques used, were extracted from included articles and recorded in Excel spreadsheets. Following a critical analysis, major trends were observed and discussed narratively, highlighting existing knowledge gaps and challenges in current research. Ethical concerns were considered by ensuring all studies complied with ethical requirements from different institutions concerned.

3. RESULTS AND DISCUSSION

The results of the selection process, including identification, screening, assessment of eligibility and inclusion are illustrated in the PRISMA flow diagram in Figure 1. Specific reasons for exclusion of articles are summarized in Table S2 of the supplementary materials. Following the selection process, we retained a total of 29 papers investigating the presence of plastic particles in the human body and 10 in other mammalian organisms. In addition, 38 studies assessing the effects of various plastic types on various tissues using in vitro models, were incorporated as they provided valuable insights into potential health risks. While the search strategy used through the different databases resulted in an imbalance and could have biased retention, this selection was used so as to maximize the likelihood of retrieving the most relevant studies in humans and other terrestrial mammals. Also, additional searches confirmed that this imbalance reflects the current scarcity of studies. Furthermore, in accordance with PRISMA guidelines for transparency, reproducibility, and scientific rigor, the authors deliberately excluded grey literature from the selection process. Although this inevitably reduced the size of the dataset, it guaranteed the inclusion of high-quality, peer-reviewed, and accessible studies only. This choice enhanced the reliability and compa-

rability of the results, while also underscoring the limited availability of robust peer-reviewed research in this field.

3.1 Micro and Nano plastics in human tissues

The papers reviewed involved participants across a range of age groups (children between 0-15 yrs, adults >18

to 60 years, and the elderly >60 years) and both sexes, as elaborated in Figure 2. Samples from general body tissues were obtained from both male and female individuals, meanwhile studies focused on specific reproductive organs obtained separate samples from each sex as required. Very few studies, however, reported gender analysis and limited

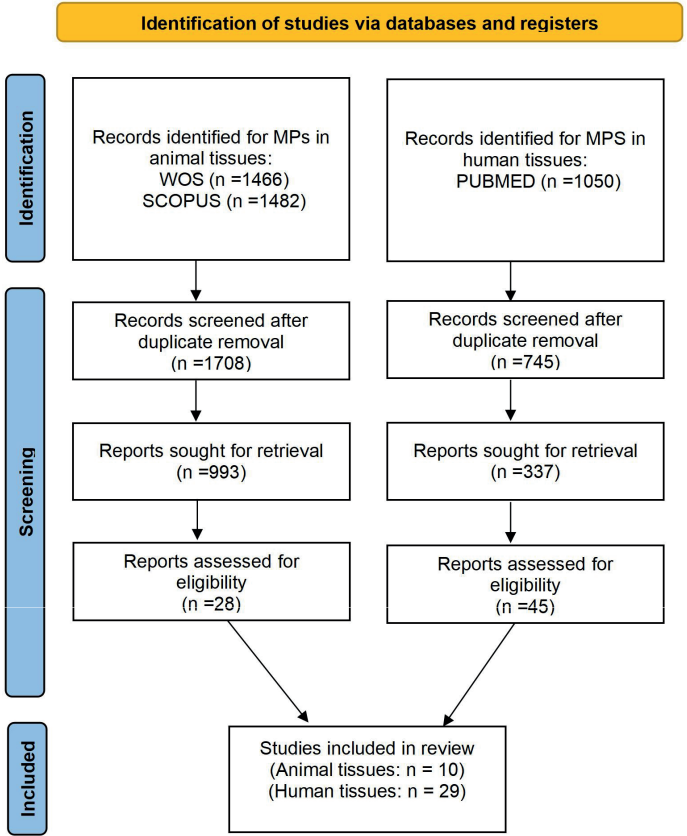


FIGURE 1: PRISMA flow diagram illustrating the study selection process.

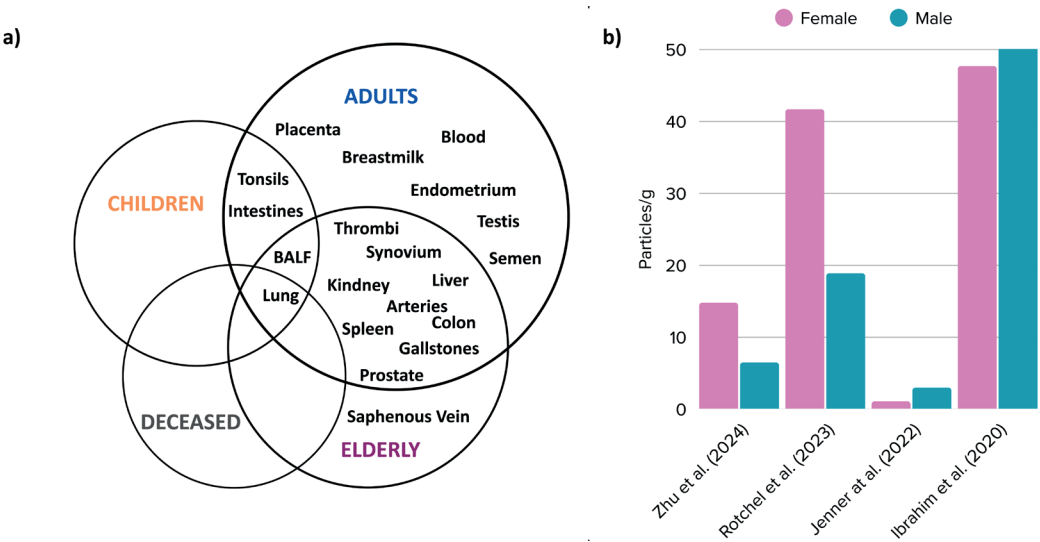


FIGURE 2: Overview of sampled populations and gender-based microplastic abundance comparisons; a) Venn diagram showing the distribution of sampled population groups across studies, with intersections indicating common tissue types analyzed; b) Comparison of some selected studies on general body tissues; lung, tonsil, intestines (Zhu et al. 2024); lung (Jenner et al. 2022); vein (Rotchell et al. 2023) & colon (Ibrahim et al. 2020), highlighting potential gender-related differences in microplastic accumulation.

comparison to concentrations. The small sample size in some studies, constrained by limited control over patient recruitment, coupled with the study of heterogeneous tissues across populations, given the availability in various settings, introduced a risk of bias in the overall assessment of studies and limited age or gender analysis. Thus, highlighting the need for a standardized design in further studies to clarify patterns of MNPs distribution in humans.

In general, samples were collected from both patients with diagnosed pathologies and healthy individuals, as certain studies aimed to investigate the potential associations between microplastic exposure and medical conditions. One study used samples obtained from autopsies and all samples were obtained following written consent and permissions granted by the health infrastructures involved.

MNPs have been extensively detected and quantified in several human body tissues embedded in the circulatory, digestive, lymphatic, respiratory, reproductive, and urinary systems. The first study indicative of this contamination in the human body was conducted in Malaysia by Ibrahim et al., (2020) on patients (age range 36-72 years) scheduled for colectomy (Ibrahim et al., 2020). The study aimed at polymer identification and characterization due to the increasingly reported exposure of MNPs through the food chain. Besides, the authors highlighted easy approval and accessibility to samples from routine surgeries performed in patients with colorectal conditions, thereby minimizing the need for invasive procedures.

Based on the selected papers, the results highlighted the following priority by numbers of studies, which reflects the distribution of scientific attention across human organs in the context of MNP detection (Figure 3): placenta (Amereh et al., 2022; Garcia et al., 2024; Liu et al., 2023; Ragusa et al., 2021, 2021; Weingrill et al., 2023; Zhu et al., 2023); lungs (Amato-Lourenço et al., 2021; Jenner et al., 2022; Zhu et al., 2024); bronchioalveolar lavage fluid (BALF) (Baeza-Martínez et al., 2022; Chen et al., 2023), blood (Leslie et al., 2022a; V. L. Leonard et al., 2024); liver, kidney, urine, spleen (Horvatits et al., 2022; Massardo et al., 2024); meconium, infant feces, breast milk (Liu et al., 2023); knee tissues (Z. Li et al., 2024); thrombi (T. Wang et al., 2024; D. Wu et al., 2023); arteries (Liu et al., 2024); saphenous vein (Rotchell et al., 2023); gallstone (Zhang et al., 2024) and intestines (Zhu et al., 2024); other body fluids such as saliva (Abbasi & Turner 2021), urine, and feces (Abbasi & Turner 2021; Liu et al., 2023; Yan et al., 2022) are also being investigated and have been shown to harbor micro- and nanoplastics. However, the latter primarily provides information on contaminants excreted from the body, which is beyond the scope of this review. These were therefore not included in this review.

3.1.1 Abundance of MNPs in human tissues

Typically, the concentration of MNPs in human tissues was reported in units of microplastic particles per gram of tissue (Figure 4), mainly attributed to ingestion and inhalation (La Porta et al., 2023). The digestive system contains

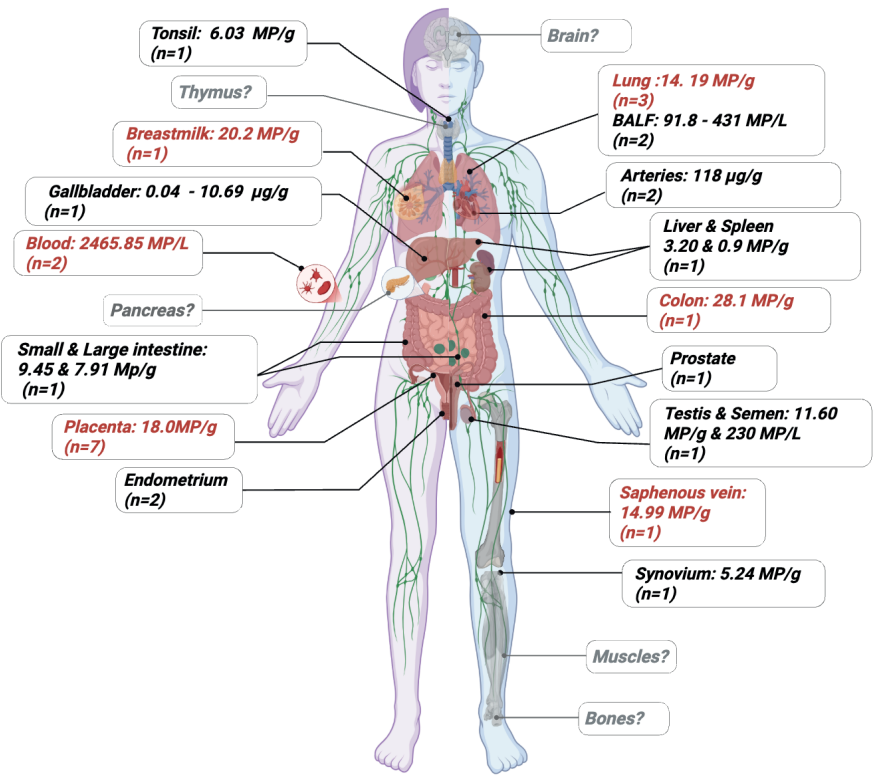


FIGURE 3: Overview of regions assessed for MNPs presence in humans, including specific male and female organs. Analyzed tissues, alongside study counts registered in the reviewed studies are displayed, providing insight into the current research distribution. Tissues highlighted in red indicate the highest concentrations, meanwhile, in grey are currently unexplored pertinent tissues (Created by the authors using BioRender. <https://BioRender.com/xs1b5oc>).

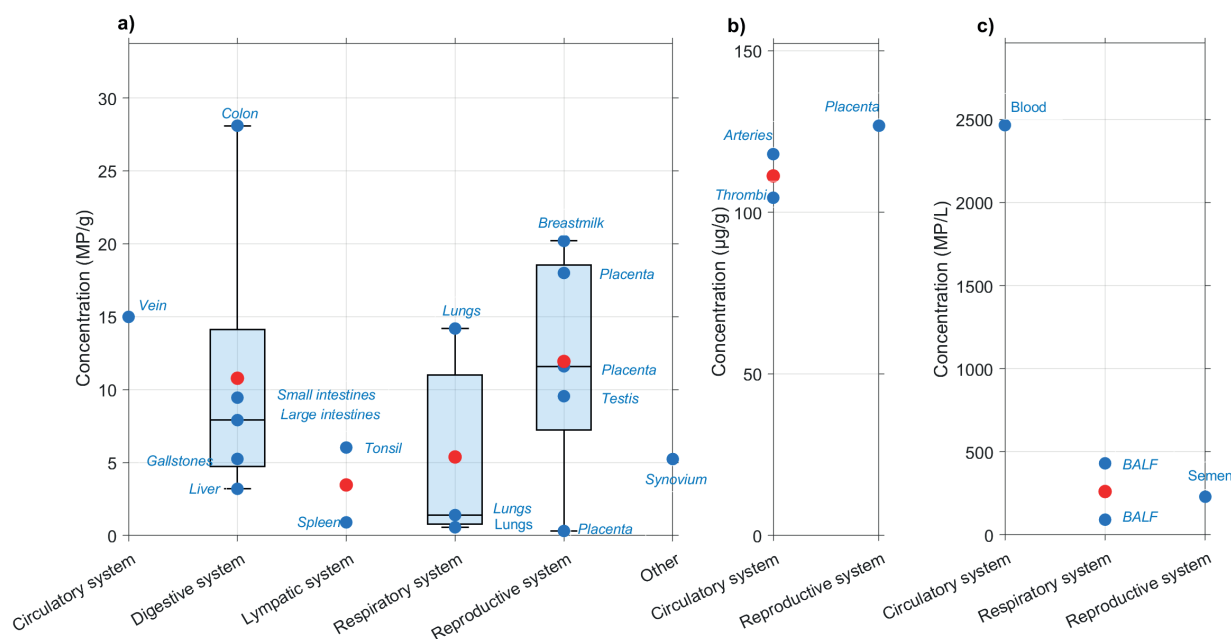


FIGURE 4: Concentration of microplastics across various human body systems. The data illustrate the relative abundance of microplastic polymers using different units of measurement: (a) MNP particles/g; (b) μg/g; (c) MNP particles/L. Each data point represents the outcome of a single study, with the specific organ or tissue labelled adjacent to the corresponding point. For body systems with more than two studies available, the mean value is indicated by a red dot. For systems with three or more studies, a boxplot is included, with the inner black line representing the median concentration.

the colon tissues, from which the most substantial concentration of 28.1 ± 15.4 particles/g was registered. This could be explained by the vital role it plays following the breakdown of large particles alongside food and other ingested compounds through transformation into forms ready for nutrient absorption or compaction of indigestible fraction to feces, which is then transferred to the rectum awaiting expulsion. Smaller concentrations of approx. 9.45 ± 13.13 and 7.91 ± 7.00 particles/g were recorded in the small and large intestines, respectively (Zhu et al., 2024), 3.2 particles/g in the liver (Horvatits et al., 2022) and 5.25 ± 4.88 particles/g in the gall bladder (Zhang et al., 2024). Despite the use of similar digestion protocols in these studies, filters with smaller pore sizes were used in analysis of the colon and liver ($0.45 \mu\text{m}$) as compared to those used for the intestines ($20 \mu\text{m}$), thus a possible justification of differential abundance. Moreover, identification techniques used in the colon-focused study included stereo-spectroscopy, SEM/EDX and μFTIR , meanwhile the latter used LDIR coupled with FTIR for particles. This disparity in methodology coupled with the insufficient number of studies, rendered the comparison challenging, thus highlighting the need for further studies with emphasis on the use of a standard methodology.

The abovementioned study of the digestive system by Zhu et al., (2024) also analyzed tonsil and lung samples on observing discrepancies between loads across age groups and genders. The age group comparison revealed how adults (>18 yrs) featured the highest loads of MNPs compared to infants (0-2 yrs) and children (2-11 yrs), thus establishing a correlation between age and accumulation potential. Moreover, on examining discrepan-

cies according to gender, female samples were reported to be much richer in MNPs compared to males in all tissues, with an average abundance of 14.81 particles/g and 6.47 particles/g, respectively (Figure 4). This finding appears to suggest that in addition to particle properties, clearance mechanisms may also depend on lung physiology and anatomy of the receptor, as females generally tend to have smaller thoracic cavities and narrower respiratory tracts than males, thus featuring a weaker clearance capacity allowing for increased accumulation of microparticles in their lungs (Dominelli et al., 2018). A clinical study conducted in the UK yielded contradictory results as lung samples obtained from males were found to contain an abundance of MNPs, registering 2.09 ± 1.54 particles/g compared to females 0.36 ± 0.50 particles/g. In an attempt to explain this discrepancy, the same physiological differences of clearance capacity were used, thus underscoring the uncertainty and need for standardized analytical techniques and larger sample size to clarify potential sex-related differences regarding accumulation of these particles (Jenner et al., 2022). Across lung compartments, relatively low concentrations of microplastics were quantified (in the upper, mid, and lower parts), with the highest proportion detected in the lower region with 3.12 ± 1.30 MP/g (adjusted to 1.65 ± 0.88 MP/g having subtracted background contamination) compared to 0.80 ± 0.96 MP/g identified within the upper region (adjusted to 0.23 ± 0.28 MP/g) and 0.41 ± 0.37 MP/g within the middle / lingular region (adjusted to 0.33 ± 0.37 MP/g). Moreover, mucociliary clearance in the respiratory tract may cause these particles to be swallowed into the digestive system, thus providing an additional channel allowing en-

try of MNPs into the digestive system (Prata, 2018). High concentrations in the lower regions could favor long-term retention in the lungs, absorption into the bloodstream, or translocation to backup clearance systems such as the lymphatic vessels, all of which are vulnerable to chronic inflammation.

Another notable quantification of MNPs in the human body was recorded by Liu et al., (2023) from the female reproductive/ gynecological system, which encompasses breastmilk (20.2 particles/g) and the placenta (18 particles/g). Earlier reports of MNPs concentration in women's placentas revealed relatively smaller quantities during an extended study of three consecutive intervals by Weingrill, who, per 50g sample, found: 4.1 ± 1.3 MNPs particles in 2006, 7.1 ± 0.9 MNPs in 2013, and 15.5 ± 3.0 MNPs in 2021 (Weingrill et al., 2023), with the more recent data indicating an evident increase over time. In terms of mass concentrations, 126.8 ± 147.5 $\mu\text{g/g}$ were obtained from the placenta (Garcia et al., 2024), while the arteries revealed concentrations of 118.7 $\mu\text{g/g}$ and the thrombi an average concentration of 104.5 $\mu\text{g/g}$ (Liu et al., 2024; T. Wang et al., 2024). These values are relatively comparable and pinpoint the movement of MNPs in circulation. This finding is particularly intriguing, suggesting the potential ability of MNPs to cross the highly vascularized placenta during blood exchange between the mother and fetus.

A recent analysis undertaken by Leonard et al., (2024) studied blood samples obtained from 20 healthy volunteers, recording an average concentration of 2465.85 particles/L. Eight of the samples contained MNPs that met LOQ criteria, obtained following subtraction of instrument sensitivity and background contamination. This suggests significant bioaccumulation in the blood, particularly as

this value was five times greater than the 431 particles/L recorded in bronchoalveolar fluids (Chen et al., 2023). The former study presented an equivalence of concentrations in mass per volume with a range of $1.84\text{-}4.65$ $\mu\text{g/mL}$, therefore an apparently slightly lower level than that obtained two years previously by Leslie et al., (2022), who registered a maximum of 7.1 $\mu\text{g/mL}$ concentrations in blood samples obtained in the Netherlands (Leslie et al., 2022a). However, when applying LOQ to the latter, the majority of concentrations were found to be <1 $\mu\text{g/mL}$. On this occasion, no gender and age-based analyses were performed as personal information relating to volunteers had not been disclosed.

3.1.2 Range of MNP sizes found in human tissues and characterization of polymer types

Across the studies considered, the size distribution of identified particles was solely in the micro-range with dimensions $1\mu\text{m}$ to larger 9 mm fibers in bronchoalveolar lavage fluid (Baeza-Martínez et al., 2022; Chen et al., 2023). Nonetheless, most particles fell in the range of $1\text{-}500\text{ }\mu\text{m}$ as seen in samples from the lymphatic system between 4 and $471.8\text{ }\mu\text{m}$ (Horvatits et al., 2022; Zhu et al., 2024), the reproductive system with particles in the range of 2 and $500\text{ }\mu\text{m}$ (Amereh et al., 2022; Demirelli et al., 2024; Ragusa et al., 2022), and urinary system between 1 and $30\text{ }\mu\text{m}$ (Massardo et al., 2024) (Figure 5). The presence of systemic and more specialized biological barriers could influence the predominance of smaller sizes (ranging between tens of micrometers or below) in the tissues of the reproductive and urinary system, as compared to the systems located behind just one epithelial or endothelial layer, such as the circulatory and lymphatic system. Nanometric particle

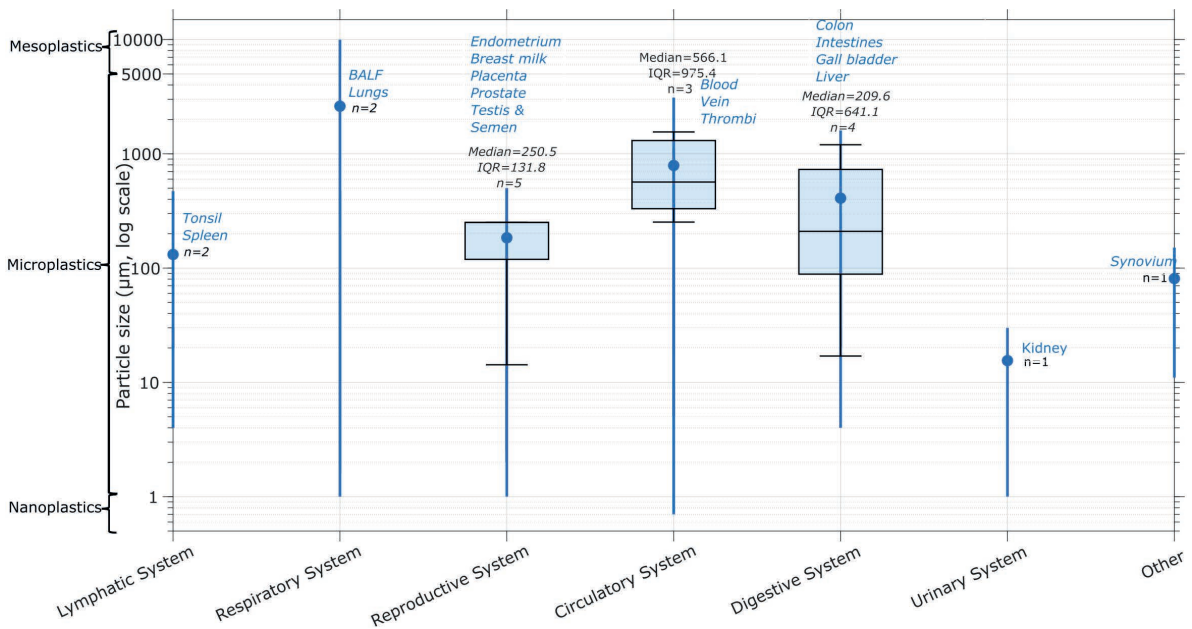


FIGURE 5: Size ranges of microplastics detected across different systems in the human body. Blue vertical lines indicate the range of particle sizes reported, while blue dots represent the mean values for each body system. For systems represented by three or more studies, a boxplot is included, with the inner black line representing the median concentration and the whiskers showing the full spread of the tissue means. The section for the reproductive system contains distinctive tissues from males and females (BALF, bronchoalveolar lavage fluids).

lengths were recorded in the blood (Leslie et al., 2022), yet particle lengths were also recorded in the millimetric range (Leonard et al., 2024) which confirms that the transport function of blood allows a broader size spectrum as compared to other tissues in the same system, such as the arteries and veins, which perform a more size-dependent retention. Nonetheless, the respiratory system, including lungs, BALF, and the colon in the digestive system, which have little or no protective interfaces, facilitates contact between MNPs in sizes of hundreds to thousands of μm through ingestion and inhalation. The predominance of larger particles in these organs reflects their role as primary entry routes, whereas smaller fragments and nanoplastics tend to pass epithelial barriers and accumulate in deeper tissues. These findings demonstrate that particle size distributions across organs are not random but are shaped by the combined effects of exposure route, barrier selectivity, and systemic transport dynamics.

Notwithstanding, the limited quantitative datasets and heterogeneous analytical protocols remained a major constraint, especially in nanoparticle detection, as filtration membranes with large pore sizes were used in various sample digestions. Moreover, even those who used filters in the nano range were further limited by the detection techniques. FTIR and Raman spectroscopy were the most commonly used techniques, although these have a lower size detection ($1\mu\text{m}$) threshold, rendering them inadequate for reliable analysis of nanoparticles. Other thermal analytical techniques, such as Pyrolysis-GC/MS do not provide particle information, rely on complex data, are destructive and are intended for use coupled with other techniques; this however would further increase the complexity and cost (Leslie et al., 2022a; Mariano et al., 2021).

A total of 28 polymer types were detected based on their chemical composition and reference spectral libraries (Figure 6). One study, using Nile red solution for a staining protocol and quantification via fluorescence microscope, was unable to assign some polymers and labelled these as 'unidentifiable' (Horvatits et al., 2022). In the circulatory system, thrombi studies reported the prevalence of Polyamide (PA), a textile/fiber related polymer commonly used in industrial fibers and medical textiles. Wang et al., (2024) recorded a predominant presence of PA (PA66) in the size range of 20-50 μm from patients who had undergone thrombectomy due to Ischemic Stroke (IS), Myocardial Infarction (MI), or Deep Vein Thrombosis (DVT). A percentage of 81.3% PA was registered in thrombi samples obtained from patients diagnosed with IS, 40% for MI, and 100% for DV thrombi. The remaining percentages were Polyethylene (PE) (T. Wang et al., 2024). Similarly, Low Density Polyethylene (LDPE) was the only polymer detected in a further thrombi study (D. Wu et al., 2023). One study conducted on blood samples obtained in the UK reported a prevalence (32%) of PE (Leonard et al., 2024) again, whereas a Dutch study found levels of Polyethylene Terephthalate PET corresponding to 50%, followed by PE (23%) (Leslie et al., 2022a). In the arteries, PET was once again the most widely abundant particle detected (73%) in a study of the arteries, followed by 15.54% PA-66 (Liu et al., 2024). However, venous tissues were richer in alkyd resin (45%) (Rotchell et al., 2023).

Conversely, samples obtained from the digestive system reported Polyvinylchloride (PVC) as the dominant polymer in the small (59.8%), and large intestines (88.4%), in the tonsils (46.2%), and in the lungs (50.8%), followed by smaller amounts of Polypropylene (PP) (Zhu et al., 2024). How-

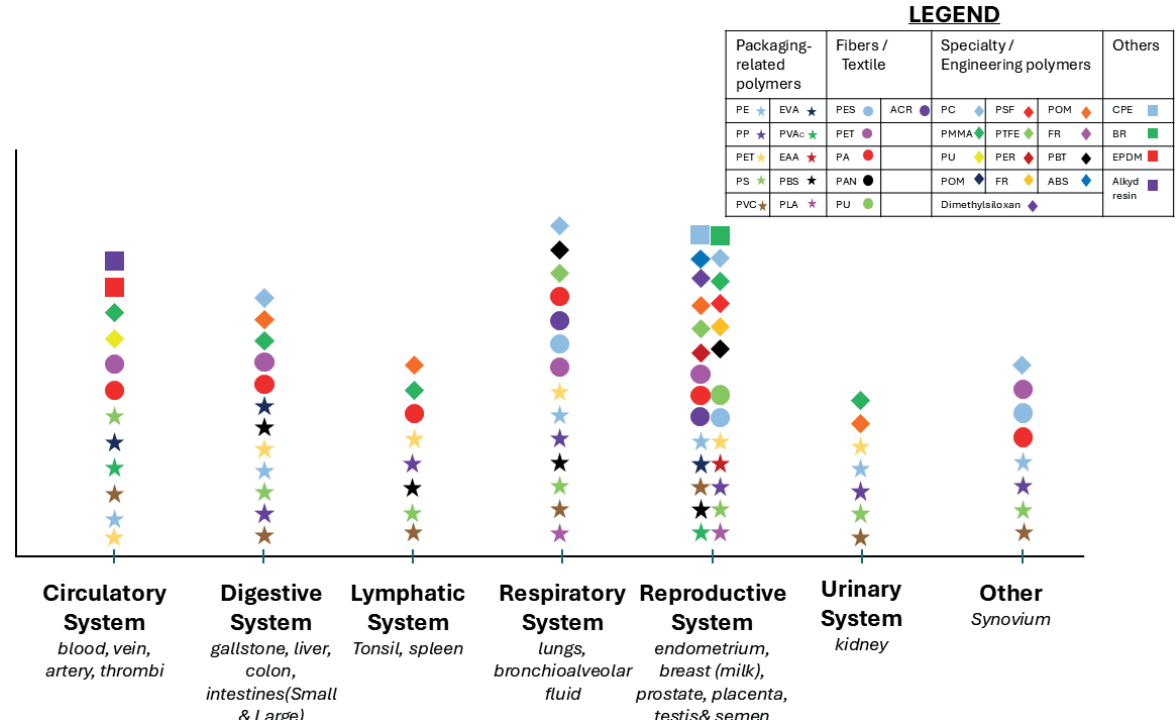


FIGURE 6: Microplastic polymer types detected in various human body systems.

ever, high levels of PP were found in parenchymal tissue from the distal and proximal regions of the left lung (35%) as reported by Amato et al., (2021) in Brazil; this finding was similar to the 23% obtained by Jenner et al (2022) in the UK. Moreover, a study conducted by Chen et al., in 2023 in samples from bronchoalveolar fluid likewise recorded a dominance of PP with a percentage of 41.9%, although an additional study of bronchoalveolar fluids delineated rayon/viscose as the most widely encountered polymer with a prevalence of 40.5%.

Therefore, these findings seem to imply that plastic packaging, including PE, PET, PP, and PVC, is the most widely prevalent polymer in the circulatory and digestive systems, consistent with their environmental availability and dominance in food contact materials. Meanwhile, fibers including PA and rayon are recurrent in the bloodstream and respiratory tissues, likely reflecting inhalation and possible dermal contact with synthetic fibers. The prevalent entry pathway of MNPs into the human body would be water/food contamination derived from the consumption of contaminated fruits and vegetables (Oliveri Conti et al., 2020), seafood, salt, honey, beer, tap water, and contamination from the films and wraps enclosing these beverages combined to air pollution (indoor and outdoor) (Cox et al., 2019). These findings therefore support the notion of ingestion and inhalation acting as the dominant pathways for MNPs transmission (Zhu et al., 2024; Y. Li et al., 2024)

In the male genitalia, PS alone represented 67% of the total amount of polymers detected in the testis, with the same study recording PVC and PE as most abundant in semen (25% each) (Zhao et al., 2023). Another study reported PS as the polymer most widely present in semen with levels of 31% (N. Li et al., 2024). In women, placental tissues were found to be abundant in PE- with levels of 54% detected by Garcia et al., (2024) and 43% by Amereh et al., (2022), Zhu et al. in 2023 found PVC to be more prevalent (43.2%) (Zhu et al., 2023). A further study reported the prevalence of PA and PU with a percentage above 74% in samples from the placenta and breast milk (Liu et al., 2023). Wein-grill et al., (2023) conducted 3 follow-up studies of MNPs in the placenta across three different years, recording PP as the dominating polymer in 2006 and 2013, however, in 2021, PES was the most widely present polymer with a percentage of 13%, thus attesting to an increased concentration and enrichment in polymer diversity produced by exposure over time. The reproductive system appears to be equally susceptible to the translocation of packaging and textile-related polymers from ingestion and inhalation. This is highly influenced by lifestyle, environmental sources, and particle characteristics, which enable the polymers to cross human biological barriers and accumulate in deep sites of maternal and fetal tissues.

MNPs (PA, PU, PE, PVC, PP, and PES) may potentially possess the ability to cross significant biological barriers, such as the blood-placenta barrier, to reach the offspring; further research should therefore be carried out to determine possible health implications for both the mother and fetus, and to bridge a significant gap in our understanding by investigating the susceptibility of other biological membranes such as the blood-brain barrier to contami-

nation (Kopatz et al., 2023; Rajendran & Chandrasekaran 2023).

Furthermore, engineering polymers are less frequent, yet also detected in these tissues with multiple barriers such as PC, POM, and PMMA, in the lymphatic, urinary and reproductive tissues. Although less frequent, their occurrence should set an alert as they are typically used in medical devices, coatings and more robust industrial applications. Their presence may suggest procedural routes, yet their health risks are still unknown.

The heterogeneity of polymers found across the studies examined may be due, at least in part, to regional variations, limited sample sizes, and methodological disparities. This has indeed somewhat hampered comparison with the other metrics studied, including the concentration of MNPs. Accordingly, analytical techniques should be standardized to ensure the yielding of uniform data, to capture the extent, distribution, and potential health impacts of microplastic contamination across diverse populations and environments.

3.1.3 Color and shapes

A wide range of shapes of MNPs have been detected in the human body, including spheres, sheets, pellets, granules, block-shaped, film, fibers, fragments, and irregular shapes. In particular, the detection of fibers and fragments, the presence of which is possibly due to their good mechanical properties (chemical stability and resistance to wear) underlying their widespread global usage (Yang et al., 2023) has been widely reported (Figure 7). Fibers however may prove difficult to remove from the human body, being detected in bronchial fluids as well as to a greater extent deep within the lungs (Baeza-Martínez et al., 2022; Jenner et al., 2022). Samples obtained in Spain from fluid sampled from the lower airways of patients undergoing bronchoscopy revealed a 93% abundance of particles shaped as fibers with sizes between 140 µm-9.96 mm (Baeza-Martínez et al., 2022). In lung tissues, a slightly higher percentage of fibers (49%) was recorded versus fragments (43%) between (12µm and 2mm) (Jenner et al., 2022). Nonetheless, bronchoalveolar lavage fluids also recorded a higher percentage of fragments, with 48.7% > 24.1% of fibers within a smaller size range (1-20µm) detected in Chinese children diagnosed with pulmonary diseases (Chen et al., 2023), and from lung samples obtained in Brazil from 20 coroner autopsies of people who died from pulmonary issues (87.5% fragments and 12.5% fibers within 1 and 5µm) (Amato-Lourenço et al., 2021).

In blood samples, 88% of all particles detected were white fragments (Leonard et al., 2024), similar to observations made in placental tissues in a study conducted in China, which harbored fragments in ample quantities. In this study, the size range of 20-200 µm was most abundant (70%), whilst a total of 50% of microplastics larger than 200 µm were represented by fibers (Ragusa et al., 2022; Zhu et al., 2023). As also reported in an Iranian study, a tendency was observed whereby smaller particles typically presented as fragments, with particles measuring 2.9 -35 µm were dominated by fragments in both normal and IUGR placentas (Amereh et al., 2022). This finding may support

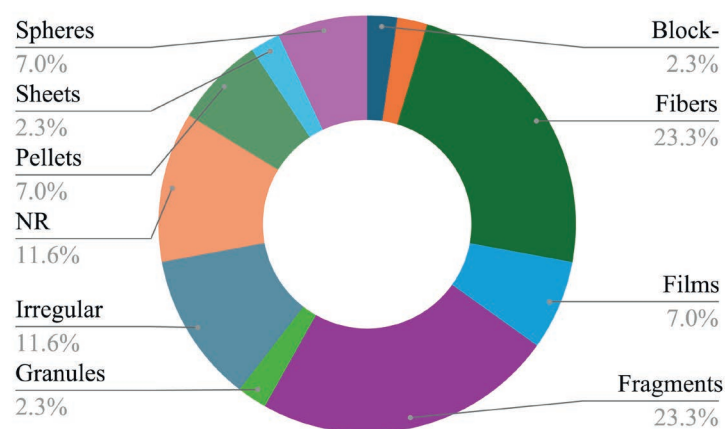


FIGURE 7: Overview of MNP polymer shapes identified within the human body across multiple studies. The figure presents raw frequencies, reporting observed occurrences rather than globally representative proportions. Interpretation should take into account differences in analytical methods, study design and reporting details among studies (NR stands for Not Recorded).

the notion according to which it is the actual structure of the fibers to enable them to resist biological clearance mechanisms, thus rendering the fiber more persistent and suggesting a lower propensity to breaking down and moving into the circulation.

Likewise, fibers/filaments found in the digestive system accounted for over 96% of particles in colon tissues, all measuring in the range of 0.8-1.6mm (Ibrahim et al., 2020). The tonsils, small and large intestines however, recorded a high prevalence of fragments in the size range of 20 -500 μm (Zhu et al., 2024). These findings therefore confirm that smaller particles tended to be fragments, meanwhile fibers were usually longer. Such size and shape distribution suggests distinct exposure pathways and transport mechanisms. Large fibers are likely ingested or inhaled, and after undergoing breakdown, suffer limited shape altering as compared to fragments, which are usually the product of continuous fragmentation.

3.1.4 Effects of MNPs in human tissues

The identification of different polymers in human tissues has raised serious concerns over potential health effects. Numerous studies are currently being carried out to ascertain the fate of these polymers in the body. Factors such as polymer types, particle size, and surface properties promote MNPs movement and interactions throughout the human body and influence the formation of the protein corona capable of providing them with a new biological identity and mediating surrounding interactions (Du et al., 2023). The results of toxicity tests performed by various authors using in-vitro culture mediums have been outlined in Table S4 of the supplementary materials. The different cell lines studied include the ovarian granulosa cell line, macrophages/blood cells, lung, colon, and intestinal cell lines, liver, placenta, kidney, brain, lymphocytes, cardiomyocytes, testicular and neural stem lines. PS represents the most widely investigated polymer across the entire body, although a wide array of polymers such as PET, PP, PVC, PA, and PE, in order of recurrence, is detected in various body tissues. PP is the most widely used, particularly in food

packaging, whilst PVC, featuring a high hazard score and potential to cause health defects, is predominantly used in building and construction and has been detected throughout numerous body tissues (Zhu et al., 2023). Nonetheless, despite the finding of the significant presence of the latter two polymers throughout the human body, their potential for harm seems to be largely overlooked. Indeed, two of the selected studies evaluated the risk associated with PP and PVC in the human body. The first described a molecular dynamic simulation using lung surfactants (LS) (Li et al., 2022), whilst the second investigated the impact of these polymers on human monocytic THP-1 cells (Zingaro et al., 2023). Both polymers proved capable of causing harm, although more extensive studies are needed to allow more definitive conclusions to be drawn with regard to the impact of these polymers on human health.

An in-vitro model in which human M1 macrophages (macrophages responsible for mediating pro-inflammatory response through production of cytokines such as TNF- α , IL-1 β , and IL-6) were exposed to 20 $\mu\text{g}/\text{mL}$ of PP and PVC caused oxidative stress as they increased the lipid content and changes in the cellular components. However, the authors concluded that cell viability remained high (Zingaro et al., 2023), in the same way as the results of a study investigating exposure of human lymphocytes to doses of PE (10-45 μm) exceeding 25 -500 $\mu\text{g}/\text{m}$ - these cells continued to display no significant signs of cytotoxicity although genomic instability was observed (Çobanoğlu et al., 2021). Some degree of discrepancy was observed in two other models when similar bloodlines were exposed to smaller sizes of PS nanoparticles (50-500nm), resulting in cytotoxicity and a loss of mitochondrial membrane potential. At 200 $\mu\text{g}/\text{mL}$ hemotoxicity was observed, with elevated ROS and lactate dehydrogenase production (LDH), consequently leading to cell death, indicative of a size and dose response to PS. In another study, human bronchial epithelial cells (BEAS-2B) were exposed to PS particles with a similar concentration to that used above, with an average size of 100nm, resulting in the finding that positively charged particles induced toxicity and increased ROS (Jeon et al., 2023),

thus highlighting the role of surface charge in addition to size and dose effect of MNPs toxicity in human tissues.

Although considerable advancements have been made in the search for plastics in the blood, further studies are needed to dispel any existing discrepancies with a view to ascertaining the multidimensionality of MNPs and investigate their affinities, interaction, and internalization under a series of direct physiological conditions (such as blood flow, tissue interaction, and cellular environments). For example, the sizes of polymers currently used for in vitro culture mediums and simulations cover only a small portion of the actual sizes detected in the human body. The particles studied are mostly in the ranges of 15 nm-10 μ m, meanwhile, those actually detected are dominantly in the micro range of 1-500 μ m. Regarding the concentrations to which these tissues are exposed, μ g/mL is the most widely used unit of measure, with samples being obtained in liquid suspensions and their mass recorded per volume. Difficulties however may arise during the comparison of loads naturally detected from tissues versus loads tissues are exposed to during experimentation. Nonetheless, among the studies that detected MNPs in the human body, the two studies carried out in blood samples recorded concentrations ranging from 0.1-7.1 μ g/mL (Leslie et al., 2022b; Leonard et al., 2024), considerably lower than the doses to which cells are exposed in culture mediums. For example, the concentration of PS to which lung, colon, and liver cells were exposed was studied by Peng et al., (2023), detecting an abundance ranging from 114 to 568000 particles/mL (Peng et al., 2023), i.e. three orders greater than that found in the blood (1412-10729 particles/l) (Leonard et al., 2024). This disparity underscores the challenge of translating toxicological findings into environmentally realistic scenarios. Nonetheless in vitro models are inherently limited by short exposure durations, whereas real-world exposures often span years, allowing for progressive accumulation and potential long-term effects, even at low concentrations. Therefore, while high-dose experiments remain valuable for elucidating dose-response mechanisms, their outcomes should be interpreted with caution when extrapolating to actual exposure conditions..

As highlighted in the present section, in vitro studies typically employ particles of smaller dimensions—generally nanoplastics—compared to those investigated in human and animal tissues. In general, the reduction in particle size from microplastics to nanoplastics significantly increases their toxicological potential, as smaller particles exhibit enhanced cellular uptake, deeper tissue penetration, and systemic distribution. Nanoplastics are associated with oxidative stress, inflammation, genotoxicity, and endocrine disruption, and can cross biological barriers such as the blood-brain barrier and placenta (Bhardwaj et al., 2025; Leslie et al., 2022; Yong et al., 2020). Dynamic or mechanical models may be able to provide valuable insights into these complex interactions, thus complementing data gathered from biological human models by predicting the transport and toxicity of contaminants across a wide range of scenarios without invasive procedures in animals and humans.

3.1.5 Mechanical modelling of MNPs in the human body

In vitro, in vivo, and ex vivo models have been extensively employed to investigate the kinetics of MNPs across various species, including humans. These models have provided valuable insights and contributed towards advancing knowledge of their potential health impacts on humans as portrayed in the toxicological effects in Table S4 of the supplementary materials. Research has identified the three primary mechanisms through which MNPs exercise toxicity, which include: (1) mechanical stress on cells due to their physical properties, (2) chemical toxicity resulting from the leaching of additives, and (3) serving as vehicles for other contaminants, facilitating entry into the human body. MNPs encompass such a huge diversity of properties and exploring them all may prove too cumbersome and potentially pose ethical issues if explored using animal models alone. Mechanical models are peculiar in that they incorporate parameters such as particle characteristics, biological transport mechanisms, and controlled tissue-specific kinetics. These could provide a more comprehensive predictive framework to assess the potential risks posed by MNPs in the human body.

Early in 2013, a computational study involving coarse grain simulations and molecular dynamics was carried out in France to assess the impact of PS nanoparticles on phospholipid membranes (Rossi et al., 2014). Based on this study, the ability of the PS nanoparticles to penetrate and impact the structural and mechanical properties was predicted. However, the authors pointed out a lack of direct experimental verification and high computational cost of this study over the relevant length and time scales. Boichio et al., (2021) later overcame these issues through use of a combination of experimental techniques (calorimetry, x-ray, and neutron scattering) with atomistic molecular dynamics simulations to study the interactions between short chains of PS and dipalmitoyl phosphatidylcholine (DPPC) lipid bilayers (Bochicchio et al., 2022). Once embedded in the DPPC lipid bilayer core, nano-sized PS decreased their pre and main transition temperatures, stabilizing the lipid fluid phase while reducing system cooperativity in a dose-dependent manner. However, the same data might also have suggested that above a certain threshold, the system would undergo partial phase separation. Using a coarser model compared to earlier experimentation in 2013, molecular dynamics simulation provided an interpretation at an atomic level, confirming PS as producing a mechanical softening and overall disordering effect. PE was also studied using molecular dynamics simulation to understand the effects of nanoscale PE on the DPPC bilayer. Key properties of the DPPC membrane were affected, causing lower density, fluidity changes, membrane thickening and clusters of PE at high concentrations, thus tending to cause pore formation in the DPPC bilayer (W. Wang et al., 2022). As in previous studies, the overall results obtained confirmed the alterations of properties of DPPC, the perturbation of membrane structure and dynamics, and eventual modification of the mechanical properties of these membranes (Bochicchio et al., 2022; Holl  czki & Gehrke 2020; Wang et al., 2022).

Computational fluid dynamics (CFD) simulations include different variants intended to describe the effect of MNPs on airflow and lead to lung infection/disease. This includes the discrete phase model, the Eulerian-Lagrangian approach, single or multiphase models, and laminar or turbulent models. When coupled with molecular dynamics simulation, they have been jointly proposed as a means of predicting what happens following the deposition of micro and nano plastics in the respiratory tract and interaction mechanisms at various levels of complexity (Saha & Saha 2024). Anatomical features have been repeatedly suggested as influencing the accumulation and clearance of MNPs in the respiratory tract; however, definitive conclusions remain limited due to small sample sizes and methodological variability across studies (Jenner et al., 2022; Zhu et al., 2024). Findings from CFD studies on polydispersed particles in the lungs (Islam et al., 2019) confirmed the veracity of this factor alongside other parameters such as gravity, flow rate or breathing pattern, and the chemical and physical properties of the plastic particle (Islam et al., 2023; Saha & Saha, 2024). In addition to the detailed insights into flow dynamics, models such as Multiple-Path Particle Dosimetry (MPPD) and CFD simulations could alleviate the uncertainties in sex-based interpretation by standardizing anatomical inputs and isolating the effects of morphology from other factors, thereby offering a more controlled framework to understand particle deposition and clearance mechanisms. Also, it will facilitate the design and optimization of medical device treatments for lung diseases and could significantly reduce the need for animal testing and human trials, thus saving time, money, and ethical problems. Furthermore, these simulations foster collaboration between several diverse fields to ad-

dress the common issue of micro and nano plastics (Saha & Saha 2024).

3.2 Micro and Nano plastics in animal tissues

As seen in human samples, microplastics have been identified throughout numerous body systems in mammals (Table S3 of the supplementary materials). These include the digestive (stomach, forestomach compartments, liver and intestine), respiratory (lungs), reproductive (epididymis, testicular, prostate and sperm), urinary (kidneys), musculoskeletal, circulatory and lymphatic systems. Animal species investigated include bats (*Chiroptera spp.*) in Brazil (Alencastre-Santos et al., 2024; Correia et al., 2024), cows (*Bos taurus*) and sheep (*Ovis aries*) in Iran (Bahrani et al., 2024), the European brown hare (*Lepus europaeus*) in Austria (Hornek-Gausterer et al., 2021), bulls in Germany (Grechi et al., 2024), pigs (*Sus scrofa*) in China (Li et al., 2023), dogs (*Canis familiaris*) and cats (*Felis catus*) in Portugal and the US (Hu et al., 2024; Prata et al., 2022) and various rodents in China and Italy (Nessi et al., 2022; Yang et al., 2024). The age and gender information of these mammals were cited solely for descriptive purposes, but no statistical analyses were performed.

Only a few studies have reported qualitative and quantitative descriptions of MPs in animals versus human samples, with a range of reporting units being used across studies, although 4 out of 7 papers reported in particles/g, as seen in Figure 8. Among terrestrial mammals, plastics were detected at a concentration of 180 particles/g in the 20.34 µm-916.36 µm size range in the lungs of a domestic pig, and 97 particles/g in a shorter range of 20.34 µm-501.49 µm in pig fetuses (Li et al., 2023). This concentration of MPs in domestic pigs is ten times greater than the

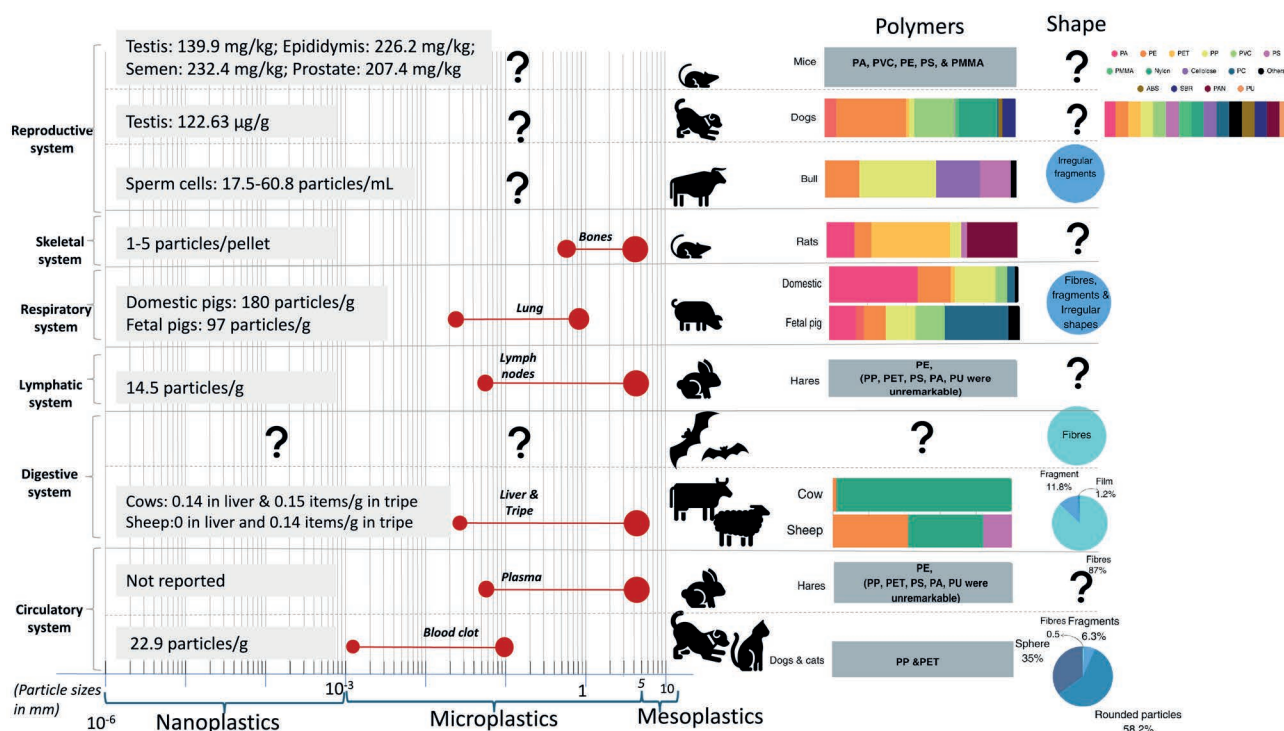


FIGURE 8: Overview of MNPs in animals' tissues. Question marks denote missing and unclearly stated information.

maximum amount obtained from human lungs (14.19 particles/g as captured by Zhu et al., (2024)), considering that both used the laser direct infrared spectroscopy for identification. Although the detectable size range of MPs identified in the pig analysis was broader compared to humans, the considerably higher concentrations suggest that particle size alone cannot account for the difference. However, this study encountered the limitation of sample size given that only one pig was sampled and, therefore, comparison was limited to the fetal pigs also analyzed in this study. However, to strengthen the argument and substantiate the conclusion, a larger sample size would be necessary.

Tissues from the lungs, liver, kidney, ileum and blood were sampled from dogs and cats in a study conducted in Portugal, which reported a median of zero particles (0 particles/g) for the majority of tissues, although recording a median of 22.9 particles/g in a size range of 1-100 μm (Prata et al., 2022) for blood clots. In this sample type, only two polymers (PP and PET) were detected. It is worthy of note that PET was also one of the most widely prevalent MPs in the human circulatory system, possibly since it is typically used in the production of clothing and home textiles in the form of polyester fibers. It is at times combined with cellulose-based fibers such as cotton, linen and hemp, to improve softness, versatility, durability and other properties (Periyasamy & Tehrani-Bagha 2022). MPs were mostly rounded particles and spheres in blood samples from dogs and cats as opposed to fragments (88%) in human blood (Leonard et al., 2024), likely resulting from exposure to microbeads from household items, contaminated pet food as well as pet shampoos and other grooming products. Furthermore, dogs and cats have shorter digestive tracts compared to humans, thus potentially facilitating the uptake of round particles, which present with a more uniform geometry (He et al., 2024). A meaningful comparison of loads could not be achieved due to methodological differences due to the use of different reporting units. Moreover, the use of blood clots may have implied increased microplastic concentrations due to sedimentation and particle entrapment during the coagulation process.

Regarding the lymphatic system, a concentration of 14.5 PE particles/g was quantified in the mesenteric lymph nodes of European brown hares (Hornek-Gausterer et al., 2021). However, when compared to investigations in the human immune system (spleen: 6.03 particles/g and tonsil: 0.9 particles/g), the concentration of MPs detected in European brown hares was significantly higher (Horvatits et al., 2022; Zhu et al., 2024). These findings, however, may have been influenced by detection limits as only particles over the size of 50 μm were recorded in hares, thus providing a broad size range of MPs yet excluding smaller particles. Meanwhile, in human studies, particle size ranged between 4 and 30 μm for spleen samples and between 20.34 and 471.87 μm for tonsil samples, thus allowing for detection of microplastics despite covering an overall smaller size spectrum. Aside from the methodological differences in detection sensitivity, variation in the immune system function across species and greater exposure to the outdoor environment (soil, air and vegetation) might have contributed to the higher concentrations observed in animals compared to humans.

Cow and sheep tissues obtained from a butcher shop in Iran had the smallest concentration of MPs, recording a predominant 86.9% of fibers (liver: 0.14 particles/g in cows and 0.0 in sheep; tripe: 0.15 particles/g in cows and 0.14 particles/g in sheep). Analyses conducted on liver samples recorded the smallest concentration (3.2 particles/g) and smallest size range of MPs among tissues from the human digestive system (Horvatits et al., 2022), but still far greater than MPs detected in animal tissues utilizing similar methodological options (i.e. micro-Raman spectroscopy). Ruminants have a multi-chambered stomach, with fermentation and microbial action that might perhaps break down or trap microplastics more effectively than humans (Quartinello et al., 2021). Again, their longer digestion times in the rumen may allow larger or denser particles to settle, be excreted or become immobilized, thus explaining their low concentrations in the digestive system; more studies would however be needed to corroborate these theories (Dehority 2002; Dufreneix et al., 2019).

Notably, the male reproductive organ of mice revealed elevated concentration of MPs, particularly in semen (232 mg/kg), the epididymis (226 mg/kg), the prostate (207.4 mg/kg) and the testis (139.9 mg/kg) (W. Yang et al., 2024). In comparison, slightly lower concentrations were quantified in dog testis (122.6 mg/kg) although with a higher polymer diversity (Hu et al., 2024), suggesting that dogs might be more exposed to a greater range of polymers whilst displaying a lower bioaccumulation potential compared to mice. However, definitive conclusions cannot be drawn due to a lack of information regarding particle sizes which influence bioavailability, combined with the varying exposure intensities and differences in geographical location. In the same way as human tissues, these results reveal the ability of MNPs to cross the blood-testis barrier and confirm their presence in regions critical for spermatogenesis and hormonal functions, therefore posing risks for male fertility and compromising embryo development (Grechi et al., 2024).

Current research focused on the identification and characterization of MPs in terrestrial animals has provided a valuable investigation baseline. However, MPs detection in animal tissues remains an understudied topic, limiting the understanding of systemic distribution and potential long-term impact on animal health. Moreover, meaningful comparison across studies is complicated by inconsistent methodologies, varying units of measurement and lack of standardized detection thresholds. These laboratory protocols must be harmonized to ensure reliability, comparability and reproducibility of findings across both species and research contexts.

Beyond analytical challenges, a deeper understanding of upstream drivers is increasingly urgent. The presence of MNPs in terrestrial ecosystems is closely linked to the broader context of waste management practices. Plastic contamination in soils and terrestrial food webs can be traced to several sources, including improper disposal (e.g., littering), the reuse of waste-derived amendments such as compost, sewage sludge, and digestate on agricultural land, and emissions from recycling and solid waste treatment processes, particularly through atmos-

pheric depositions and runoffs (Adhikari et al., 2024; Corradini et al., 2019; Meixner et al., 2020; Porterfield et al., 2023). Interestingly, the recent discovery of a pathogenic microorganism capable of degrading PET (Srivastava et al., 2024), identified in certain solid waste treatment facilities, raises important questions about whether such microbial adaptations may be driven by prolonged exposure to plastics, materials that offer relatively hospitable conditions. This connection could be crucial for advancing our understanding of both the persistence of MNPs and potential degradation pathways within terrestrial ecosystems. Moreover, it underscores the need for targeted interventions across the entire plastic lifecycle, from production to disposal.

3.3 Critical methodological limitations in the detection of MNPs in biological tissues

As has been highlighted multiple times in earlier parts of the text, micro- and nanoplastic investigation presents several methodological limitations, including:

- The lack of a standardized analytical protocol: there is no universally accepted method for sample processing, extraction, and identification of MNPs in biological tissues. Studies use different digestion techniques, filtration steps, and detection technologies (e.g., FTIR, Raman, Py-GC/MS), each with specific size detection limits and biases. This leads to inconsistent dimensional ranges, units of measurement, and reporting formats across studies (Ośko et al., 2025);
- Limited comparability due to diverse technologies: in fact, different analytical methods vary in sensitivity, specificity, and resolution. For example, FTIR typically detects particles >10 µm, while Raman can reach ~1 µm, and thermal methods like Py-GC/MS detect polymer types but not particle size. This technological heterogeneity makes cross-study comparisons difficult (Shao et al., 2025);
- Matrix complexity and contamination risks: biological tissues are complex matrices that require careful digestion and purification. Sample loss, contamination from labware or airborne particles, and adhesion to surfaces can distort results (Stock et al., 2020). Contamination mitigation strategies are critical in MNPs analysis, particularly in biological matrices, where environmental and procedural contamination can severely compromise data integrity. Therefore, rigorous preventive measures must be implemented at every stage of MNPs sampling and analysis. This includes avoiding the inadvertent introduction of anthropogenic particles — those not originally present in the environmental sample — via sources such as laboratory surfaces, airborne deposition, analysts' garments, or equipment in use. Such contamination can significantly distort particle counts and compromise data integrity (Poli et al., 2024). Protocols for contamination prevention — such as the use of clean-air cabinets, cotton lab coats, procedural blanks, and pre-rinsed glassware — are essential to avoid overestimation of MP abundance (Huang et al., 2024; Stock et al., 2020). Recent findings demonstrate

that procedural contamination can substantially inflate microplastic counts in biological samples. Aminah & Ikejima, (2023), showed that implementing basic mitigation strategies reduced contamination levels by 70–100%, underscoring the critical need for rigorous protocols in microplastic research. While studies on MNPs often lack the inclusion of rigorous blank controls and contamination mitigation strategies, this was not the case in the selected studies for the present review. All reviewed studies implemented plastic contamination protocols to ensure data reliability, including the use of procedural and/or environmental blank controls. Only a few exceptions were noted where the inclusion of blanks was not explicitly stated: Correia et al., 2024; Ragusa et al., 2022; Zhang et al., 2024;

- Inadequate particle morphological characterization: many studies fail to report key particle attributes such as shape, surface chemistry, polymer type, and aggregation state (Alencastre-Santos et al., 2024; Correia et al., 2024; Hornek-Gausterer et al., 2021; Horvatits et al., 2022; Hu et al., 2024; N. Li et al., 2024; Z. Li et al., 2024; Liu et al., 2023, 2024; Nessi et al., 2022; Ragusa et al., 2022; Wang et al., 2024; Yang et al., 2024; Zhang et al., 2024). These factors critically influence biological interactions and toxicity but are often overlooked due to analytical constraints (Ośko et al., 2025);
- Lack of harmonized units and exposure metrics: studies report MNP concentrations using different units, making it difficult to compare exposure levels or establish dose–response relationships (Huang et al., 2024). This aspect has already been addressed in the manuscript, specifically in Section 3.1.1 and illustrated in Figure 4, where we detail the units adopted across studies involving human samples — namely, number of particles per gram of tissue, micrograms of MNPs per gram of tissue, and number of particles per liter for liquid matrices. Additionally, Section 3.2 Micro- and nanoplastics in animal tissues outlines the units used in animal studies, which predominantly include the number of particles per gram of tissues and, in some cases, milligrams per kilogram, particles per microliter, and particles per pellet. This heterogeneity complicates cross-study comparisons.

Taken together, these methodological limitations significantly hinder the ability to perform structured and meaningful comparisons across studies. The lack of harmonized protocols for sampling, sample processing, analytical techniques, and data reporting not only compromises reproducibility but also obstructs the development of robust dose–response relationships and risk assessments. As the field of MNP research continues to expand, there is an urgent need for the scientific community to establish and adopt uniform standards that ensure consistency, transparency, and comparability across investigations (Poli et al., 2024). Such harmonization is essential to advance our understanding of MNP distribution, toxicity, and potential health impacts in both human and animal systems.

4. CONCLUSIONS

Both humans and other terrestrial mammals have been shown to host microplastics across a wide range of body tissues and cells. While human studies have revealed a broader diversity of polymers, terrestrial animals have been reported to host greater quantities. Moreover, unlike aquatic media in which MNPs could be attributed only to ingestion, the inhalation of airborne particles from both indoor and outdoor environments adds to ingestion as an important entry pathway of MNPs into the systems of terrestrial mammals.

Despite growing evidence, gender and age specific analyses remain scarce. These biological variances may significantly influence MNPs exposure, accumulation and physiological response across species and should be integrated into future research.

Another major challenge lies in the lack of standardized protocols, which may have contributed to the broad variability observed in reported data for both human and other terrestrial mammals. Sample pretreatment methods may influence the recovery of particles while the use of sieve filters imposes arbitrary size thresholds. Furthermore, the variety of analytical instruments, each with differing sensitivity, resolution, and suitability for specific size ranges or polymer types, adds to data inconsistency and hampers accurate exposure assessments. This variability often makes the detection of nanosized particles unattainable and renders cross-study comparisons unreliable. To overcome these limitations, a universally accepted and harmonized analytical framework is urgently needed. However, it is worth noting that quantitative integration across exposure pathways of MNPs remains premature, given the said limitations. A holistic perspective which captures a full quantitative integration would require the understanding of multiple stressors, their co-existence in the exposome, which is summed up in all internal and external exposures, intake rates, and as well as the biological response, such as absorption efficiency in respective tissues. Therefore, addressing these limitations in new studies could pave the way towards a more integrated and realistic assessment of MNP exposure in human and animal tissues.

Health-related risks associated with exposure to MNPs have been documented across a wide range of tissues and cell lines, revealing effects such as induced apoptosis (cell death) due to oxidative stress, disruption of cellular functions, and inhibition of specific bacteria and enzymes. While current findings from in vitro, in vivo, and ex vivo studies provide a solid foundation, further research is essential to fully understand the complexities of these contaminants.

Computational Fluid Dynamics (CFD) simulations represent a promising tool to advance this field, offering the ability to model the fate and transport of particles with different physical and chemical characteristics (e.g. size, polymer type) under realistic physiological conditions. These simulations can predict biodistribution, accumulation, and potential toxicity hotspots across species. When combined with experimental data, CFD can contribute to a more comprehensive understanding of MNP behavior within biological

systems, thereby supporting the development of more effective public health strategies.

In parallel, developing a framework for future studies could provide a practical foundation for advancement in this field. It is recommended that future studies encompass parameters responsible for route-specific estimated intakes of MNPs, as well as clearance and translocation coefficients. In addition, reported abundances should include conversion factors linking particle count to mass-based units, derived from particle-specific properties. This approach would enable more meaningful comparisons across exposure routes, support the development of robust mass-based models, and contribute to the harmonization of exposure assessments concerning the presence of these contaminants in terrestrial mammals and ecosystems more broadly.

Furthermore, considering the close ecological and biological connections between humans, animals, and the environment, an approach encapsulated in the "One Health" framework, there is a pressing need for interdisciplinary and integrative investigations. These efforts will be critical in mitigating the widespread health and environmental impacts of micro and nanoplastics.

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ECO-RESTORATION THROUGH INDIGENOUS MICROORGANISMS: A REVIEW OF ADVANCES IN BIOREMEDIATION AND WASTE CONVERSION

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ABSTRACT

Environmental protection from hazardous and toxic chemical deposition is a critical global concern. Bioremediation, an economical and eco-friendly method, has emerged as a promising solution. Among various strategies, the use of Indigenous Microorganisms (IMOs)—microbes naturally present in a given environment—offers significant advantages. These autochthonous microbial populations play essential roles in biodegradation, bioleaching, biocomposting, nitrogen fixation, toxicity reduction, fertility enhancement, and the synthesis of plant growth-promoting hormones. IMOs contribute to mineral extraction, sustainable agriculture, and effective waste management across various regions. Their absence would negatively impact all life forms and ecosystem functions. This chapter emphasizes the importance of conserving and utilizing indigenous microbes to transform waste into valuable bioresources, thereby facilitating environmental restoration. The diversity of IMOs depends on their collection and isolation methods, which are influenced by specific environmental conditions. Overall, the promotion of IMO-based technologies can bridge the gap between anthropogenic waste generation and ecological balance.

1. INTRODUCTION

Microorganisms possess remarkable diversity and biosynthetic potential that can be modulated by their environment, making them ideal candidates for solving complex biological and environmental challenges. Harnessing native microbial communities for sustainable development offers economic, social, and ecological benefits, reflecting a shift from conventional methods to advanced biotechnological interventions (Birgovan (Rhazzali) et al., 2025; Lofthus et al., 2021). The widespread use of agrochemicals—such as synthetic fertilizers, pesticides, and herbicides—has indeed boosted crop productivity. However, the environmental degradation and decline in soil health associated with their use have prompted a search for eco-friendly alternatives (Derpsch et al., 2024; Firincă et al., 2023; Kundu & Biswas, 2025). One such alternative lies in utilizing soil-dwelling microbes as bioinoculants, which can promote plant growth, enhance nutrient uptake, and offer protection through antibiotic and siderophore production. In recent decades, advances in molecular tools and biotechnological techniques have significantly deepened our understanding of micro-

bial diversity and its application in sustainable agriculture (Athanassiou et al., 2018; Derpsch et al., 2024). The recognition of microbes' vital role in ecological balance has further accelerated interest in exploiting both cultured and uncultured microbial resources. High-throughput sequencing and metagenomic analyses have revealed vast unexplored microbial genomes that harbor novel genes and bioactive molecules, some of which have already been developed into marketable products. Many bio-based chemicals like ethanol, butanol, organic acids, and amino acids, traditionally synthesized using petrochemical processes, are now also obtained via microbial fermentation (Lin et al., 2023; Vassilev et al., 2021).

The cultivation process for IMOs typically involves multiple stages:

- IMO-1: Microorganisms are first cultured on cooked rice in wooden boxes;
- IMO-2: The culture is stabilized with brown sugar;
- IMO-3: The mixture is propagated further using rice bran or wheat mill run;
- IMO-4: The final culture is blended with local soil, such as clay or anthill soil.



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This mature culture is then used in composting or directly applied to agricultural fields. Additional natural inputs like fermented plant juices, calcium from eggshells, fish-based amino acids, and phosphate from animal bones are used in synchrony with plant growth stages to enhance fertility and crop yield. These inputs are environmentally safe and cost-effective, making natural farming a sustainable option. The benefits of using IMO include: reduced input costs (up to 60%), enhanced crop quality and yield, improved plant health and resistance, environmentally safe with zero chemical emissions.

Although much research has been conducted with microbial biotechnology, the coupling of IMOs and waste conversion/eco-restoration is still fairly limited. Most previous studies focus on microbial diversity or biofertilizer elaboration only (Díaz-Rodríguez et al., 2025; Gadd, 2010; D. P. Singh et al., 2016). The current study attempts to address this gap by investigating the potential of locally adapted IMOs for both bioremediation and bioresource recovery. The unique focus on indigenous microbial communities versus laboratory-developed strains and assessment of their applicability for affordable, decentralized environmental treatment has strengthened the scope of this review.

The review is mainly dedicated to the role of Indigenous Microorganisms (IMOs) in soil and waste enriched ecosystems, underlining their potentiality for bioremediation, composting and sustainable production of crops. Through highlighting the role of autochthonous microbial communities rather than exogenously introduced species, our study reinforces that context-specific strategies should be adopted for environmental recovery.

In general, this review intends to establish ecological and functional importance of indigenous microorganisms (IMOs) in waste conversion and environmental remediation. It discusses the cultivation and application of IMOs in various ecological systems, their potential to improve soil health and remediate pollutants, and how to include IMO into practical, sustainable eco-restoration projects. It focuses on naturally evolved microbial consortia. The discussion highlights their potential as low-cost, sustainable tools for enhancing environmental sustainability and circular bioresource recovery.

1.1 List of abbreviations

• ABA	Autochthonous Bioaugmentation
• ATP	Adenosine Triphosphate
• DCE	Dichloroethylene
• DDT	Dichlorodiphenyltrichloroethane
• EDTA	Ethylenediaminetetraacetic Acid
• EMs	Effective Microorganisms
• IAA	Indole-3-Acetic Acid
• IMO	Indigenous Microorganism
• IMOs	Indigenous Microorganisms
• PAHs	Polycyclic Aromatic Hydrocarbons
• PCE	Tetrachloroethene
• RDX	Research Department Explosive (Cyclotri-methylenetrinitramine)
• SVE	Soil Vapor Extraction
• SVOCs	Semi-Volatile Organic Compounds

• TCE	Trichloroethylene
• VOCs	Volatile Organic Compounds
• 2,4-D	2,4-Dichlorophenoxyacetic Acid

2. POLLUTION: A GLOBAL CHALLENGE

Environmental pollution, especially from hydrocarbons, heavy metals, oil spills, and synthetic herbicides like 2,4-D, remains a pressing concern. Microbial biodegradation has emerged as an effective remediation strategy. "Intrinsic bioremediation" refers to the natural breakdown of contaminants by native microbes without external intervention. This process typically involves microbial enzymatic conversion of pollutants into less toxic or inert products. Under aerobic conditions, oxygen acts as the terminal electron acceptor, enabling the complete oxidation of organic compounds. In contrast, anaerobic degradation produces intermediates like methane, organic acids, and hydrogen, with electron acceptors such as nitrate, sulfate, or bicarbonate (Aitken et al., 2004). The success of these processes depends on several factors, including soil pH, moisture, oxygen availability, nutrient content, and microbial diversity. Excellent degradation efficiencies have been observed in the field. For instance, in situ microbes from petroleum reservoirs and deep subsurface have been reported to be efficient in hydrocarbons degradation, reaching more than 99% removal of n-alkanes and PAHs in Dagang oil fields China (Lofthus et al., 2021). Analogous native consortia have been described in other polluted habitats where they orchestrate detoxification of pollutants by mediating enzymatic oxidation, biosorption and redox cycling.

Studies have shown that indigenous microbes are also present in petroleum reservoirs and deep subsurface environments, playing significant roles in hydrocarbon degradation (Magot et al., 2000).

These findings underscore the ecological value of site-specific microbial populations in mitigating pollution. However, the full potential of such indigenous microorganisms (IMOs) remains underutilized, particularly in integrated frameworks combining waste conversion and environmental restoration. Understanding and harnessing these naturally adapted consortia aligns directly with the broader objective of this study—to promote IMOs as sustainable, low-cost agents for achieving pollution mitigation and ecosystem resilience.

3. INDIGENOUS MICROORGANISMS AS BIO-FERTILIZERS

Native Microbial Consortia (Indigenous microorganisms; IMOs) are the local environmental microbial diversity which develop into complex and adaptive ecosystems. In contrast to commercial EMs, IMOs are harvested from indigenous soils in situ with the ecosystemic structure and functional diversity intact. These organisms play a key function in the cycling of nutrients such as biological nitrogen fixation, and phosphate solubilization, posing beneficial effects on soil structure through poly-saccharides production and soil aggregation. They also produce plant growth promoting hormone (indole-3-acetic acid; IAA) and

suppress soil borne pathogens by secreting antibiotics and siderophores (Kumar & Gopal, 2015; Solanki et al., 2023). Similarly, inoculation with fluorescent *Pseudomonas* species increased *Vigna mungo* yield by almost 300% as compared to uninoculated controls. These results demonstrate that locally adapted microbial communities can remediate nutrient imbalance and reliance on synthetic fertilizers.

Apart from nutrients management the ecological resilience of IMO's enhances sustainability in agriculture by maintaining soil health, decreasing greenhouse gas generation and conserving biodiversity. These characteristics of inherent fertility-building and the potential for adaptation make them especially useful for low-input and organic farms. By adopting and promoting the connection between microbial ecology and agronomic performance, IMO's are in essence the focus of this paper—accelerating microbe-based methods for sustainable maintenance of long-term soil fertility and ecological reconciliation (Hayat et al., 2010).

4. BIOLEACHING OF HEAVY METALS

Managing metal-contaminated water from mining and metallurgical industries is crucial both for economic re-

covery and environmental safety. Bioleaching, or microbial-assisted metal removal, is a cost-effective and environmentally friendly alternative to traditional physicochemical methods (Dianatdar & Etemadifar, 2024; Dong et al., 2023; Sarkodie et al., 2022). Indigenous microbes—adapted to the specific conditions of mine sites—often show superior performance in bioleaching under acidic or metal-rich environments. Studies have utilized native strains of *Shewanella*, *Bacillus subtilis*, and *Brevundimonas* to remove calcium, magnesium, and iron from contaminated surface waters. Both living and non-living microbial biomass have demonstrated efficient metal biosorption through passive and active mechanisms. Indigenous strains are preferred due to their inherent tolerance to local environmental stresses and higher biosorption efficiencies (Fosso-Kankeu et al., 2009, 2010; Li et al., 2024; Priya et al., 2021). Figure 1 illustrates the the management objectives and remediation strategies used to address metal pollution. The primary goals—economic recovery and environmental safety—guide the selection of remediation approaches. Among these, bioleaching using indigenous microbes such as *Shewanella*, *Bacillus subtilis*, and *Brevundimonas* represents a sustainable alternative to conventional physicochemical methods.

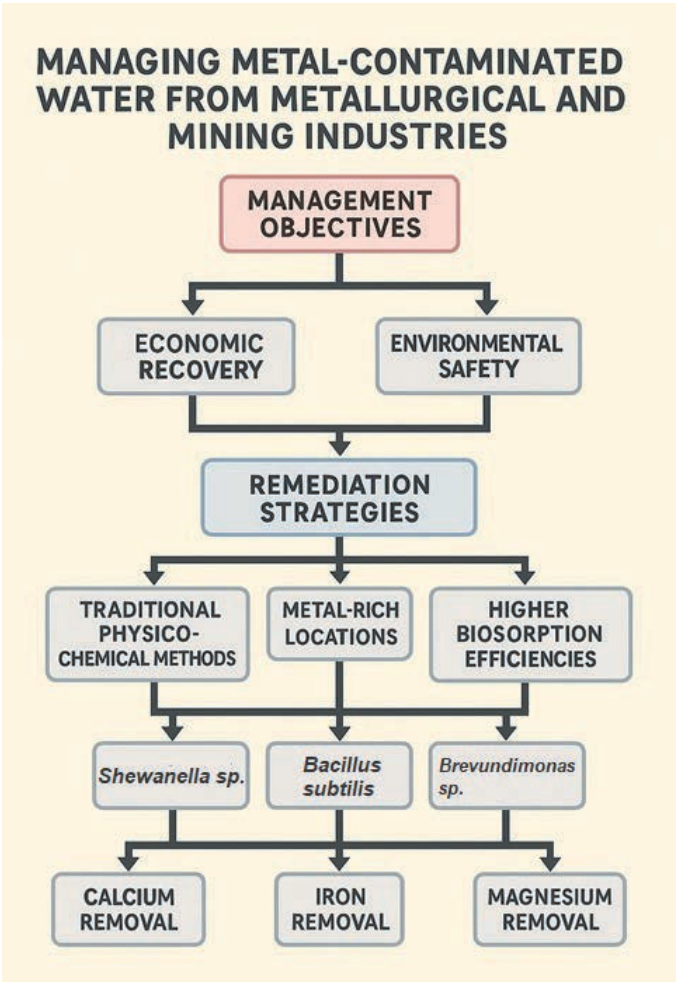


FIGURE 1: Conceptual framework for managing metal-contaminated water. The flowchart illustrates the hierarchy of management objectives and remediation strategies used to address metal pollution in mining and metallurgical industries.

These microbial systems exhibit high biosorption efficiencies in metal-rich environments, facilitating the removal of metals like calcium (Ca), iron (Fe), and magnesium (Mg) from contaminated waters. The process underscores the role of native microbial communities in achieving efficient, eco-friendly metal remediation and resource recovery.

4.1 Metal removal mechanisms by microorganisms

Microbial removal of metals from aqueous solutions occurs through two primary mechanisms: biosorption and bioaccumulation. Biosorption is a passive, energy-independent process in which functional groups—such as carboxyl, hydroxyl, amino, or phosphate groups—on microbial cell surfaces bind metal ions from the surrounding environment. Conversely, bioaccumulation involves the active uptake of metals into the microbial cytoplasm, relying on energy expenditure through ATP hydrolysis (Molaey et al., 2024).

4.2 Bioleaching of heavy metals using indigenous iron-oxidizing microorganisms

The increasing global population has led to a rise in heavy metal concentrations within municipal sewage sludge. Conventional methods such as EDTA-assisted extraction or acid leaching are often inefficient, particularly for elements like Fe, Ni, and Cr, and they pose cost and environmental challenges (Luo et al., 2023). Bioleaching, an

emerging microbial technique, offers an eco-friendly and cost-effective solution for heavy metal removal from sludge (S. Singh et al., 2016). Experimental studies by Chen C, et al. demonstrated that iron-oxidizing indigenous bacteria, when supplemented with FeSO₄, efficiently solubilized metals such as Cu, Zn, Ni, and Pb from anaerobically digested sewage sludge (Chen et al., 2022). More than 80% of these metals were removed after 16 days. Similarly, Pathak et al. observed that indigenous iron-oxidizing strains showed enhanced leaching performance compared to non-inoculated controls (Pathak et al., 2014). Factors such as pH, solid concentration FeSO₄ dosage, and inoculum concentration significantly influenced the bioleaching efficacy. The microbial species *Acidithiobacillus ferrooxidans* and *A. thiooxidans* are widely recognized for their critical role in bioleaching. Iron-based leaching is often more effective than sulfur-based pathways due to its superior sludge acidification capacity. Research by the same group also revealed that parameters such as pH, redox potential, agitation time, and sulfate levels are key in optimizing metal removal from industrial sludge using *A. ferrooxidans* (Zhou et al., 2013). Table 1 presents a comparative overview of various bioremediation techniques employed in environmental cleanup using indigenous microorganisms. It includes aerobic, anaerobic, and cometabolic bioventing, natural farming, and bioleaching among others. Each method is described by its target pollutants, the role of indigenous microbes, oper-

TABLE 1: Summary of current remediation methods using indigenous microorganisms.

Remediation Method	Target Pollutants / Application	Key Microbial Action	Limitations	References
Aerobic Bioventing	Non-chlorinated VOCs, SVOCs, hydrocarbons	Aerobic degradation with added oxygen	Ineffective in highly permeable or overly aerobic soils	(Frascardi et al., 2015)
Cometabolic Bioventing	Chlorinated solvents like TCE, DCE	Cometabolism via monooxygenase enzymes	Limited data; design challenges for shallow or moist soils	(Sui et al., 2006)
Anaerobic Bioventing	Highly chlorinated compounds like PCE, TCE, DDT	Anaerobic reductive dechlorination and cometabolism	Slow process, requires reducing conditions, gas distribution complexity	(Baczynski & Pleissner, 2009)
Natural Farming with Indigenous Microorganisms	Improves soil health, nutrient cycling, structure	Mycorrhizae and beneficial IMO improve soil and plant health	Climatic sensitivity, requires consistent microbial propagation	(B. L. Kumar & Gopal, 2015)
Biocomposting with Indigenous Microorganisms	Composting agro-waste (e.g., corn stalk, paddy husk)	Microbial breakdown of crop waste into humic substances	Quality dependent on feedstock; variable decomposition rates	(B.L. Kumar & Gopal, 2015)
Bioremediation of Organic Pollutants	Organic compounds (e.g., diesel, pesticides)	Indigenous microbes degrade hydrocarbons and VOCs	Dependent on pollutant type and site conditions	(Kuppan et al., 2024)
Bioaugmentation and Bio-stimulation	Hydrocarbons, crude oil	Enhanced degradation via nutrient addition or native microbes	Requires balance of nutrients, environmental parameters	(Nikolopoulou & Kalogerakis, 2016; Yaman, 2020)
Ozonation + Indigenous Microbes	PAHs in diesel-contaminated soils	Ozone oxidizes pollutants, microbes degrade byproducts	Microbial population reduction due to prolonged ozone exposure	(Caniani et al., 2021)
2,4-D Bioremediation using Fe3O4 Nanoparticles + Indigenous Microbes	Herbicide 2,4-D	Enhanced mineralization of herbicide with synergistic action	Requires nanoparticle synthesis, site-specific microbial matching	(Fang et al., 2012)
Arsenic Bioremediation with Indigenous Bacteria	Arsenic (As) in contaminated soils	Arsenic methylation, volatilization under aerobic/anaerobic conditions	Limited to arsenic-tolerant strains; requires enriched conditions	(Majumder et al., 2013; Naiel et al., 2024)

VOCs – Volatile Organic Compounds, SVOCs – Semi-Volatile Organic Compounds, TCE – Trichloroethylene, PCE – Tetrachloroethene, DDT – Dichlorodiphenyltrichloroethane, IMO – Indigenous Microorganisms, PAHs – Polycyclic Aromatic Hydrocarbons, SVE – Soil Vapor Extraction, ABA – Autochthonous Bioaugmentation

ational limitations, and recent supporting references (from the last five years). The table provides a concise framework for understanding the scope, applicability, and challenges associated with each strategy in sustainable environmental remediation.

4.3 Microbial mitigation of arsenic pollution

Arsenic (As), a toxic metalloid of both geogenic and anthropogenic origin, poses severe risks to human and environmental health. Microorganisms have evolved several tolerance mechanisms, such as efflux pumps encoded by the *ars* operon, enabling survival in arsenic-rich environments (Navarro et al., 2022). Bioremediation of arsenic typically involves its conversion into less toxic or volatile methylated forms by bacteria and fungi (Periferakis et al., 2022). Indigenous bacterial strains isolated from submerged paddy soils have demonstrated high tolerance to arsenite and arsenate—the more toxic inorganic forms of arsenic (Majumder et al., 2013). The isolate AMT-08, in particular, was capable of removing up to 16% of arsenic in 60 days when supplemented with organic manure. These microbes outperform strains from other sources (e.g., geothermal sites or gold mines), making them promising agents for cost-effective, eco-friendly arsenic bioremediation (Kaur et al., 2009; Keren et al., 2022). Their enhanced volatilization potential, especially under anaerobic conditions, supports their use as alternatives to genetically engineered organisms for real-world applications (Naiel et al., 2024).

5. OIL SPILL BIOREMEDIATION

Oil spills remain a serious environmental concern, particularly in marine and coastal ecosystems. Bioremediation has emerged as an effective approach, primarily through bioaugmentation (introducing specific degraders) and biostimulation (stimulating indigenous microbes with nutrients) (Nikolopoulou & Kalogerakis, 2016; Tribedi et al., 2018). While native microbial communities may struggle to degrade the complex mixture of hydrocarbons in crude oil, supplementation with specialized oil-degrading microbes can enhance the process (Bôto et al., 2021). A promising advancement in this field is autochthonous bioaugmentation (ABA), which involves isolating, enriching, and reintroducing indigenous strains from contaminated sites to enhance remediation. This method, proposed by Yaman offers superior adaptability and survival compared to external inocula. As proposed by Natural Biodegradation Guidance in 2003, effective bioremediation also depends on optimizing physicochemical conditions like nutrient availability, aeration, and surfactant activity (Yaman, 2020). Field study applications have demonstrated that the adjustment of these factors according to the local site conditions, including salinity temperature, hydrocarbon composition, can promote enhanced bioremediation. Such tailor-made development to ensure that indigenous consortia are kept active metabolic and environment-wise, further contributes towards a better recovery of oil-polluted coastal zones. One such recent study reports the bioremediation of coastal sand contaminated by fuel: over ~106 days the total petroleum

hydrocarbon level dropped from ~5,753 mg/kg to ~37 mg/kg (~98.2% removal) (Ben Laish et al., 2025).

To further enhance degradation, techniques such as ozonation have been applied to increase the bioavailability of hydrophobic compounds. While ozone can break down polycyclic hydrocarbons, prolonged exposure reduces microbial viability. Thus, a combined approach of pre-ozonation followed by biostimulation of surviving indigenous microbes, as studied by Caniani et al., has proven effective (Caniani et al., 2021). This combination of chemical oxidation with biostimulation is indicative of how hybrid nature can be optimized for site-specific remediation. Under field conditions, these strategies support a sequential pollutant degradation: hydrocarbons are first decomposed into simpler intermediates by ozonation and then composed to harmless end product by microorganisms. Indigenous microorganisms are especially well-suited for these successive treatment plans due to their flexibility (Olak-Kucharczyk et al., 2023; Tamizhdurai et al., 2022).

This strategy not only improves the bioavailability of contaminants but also supports microbial regrowth over time. Figure 2 elaborates the combined use of these methods in an explicit way. The combination strategy will enhance the broader mission of developing resilient indigenous microbial systems as a model strategy for engineering practical, sustainable and locally-applicable solutions to oil spill remediation and environmental restoration. The process involves bioaugmentation, where specialized oil-degrading microorganisms are introduced to enhance hydrocarbon degradation, and biostimulation, which optimizes nutrient levels, aeration, and surfactant activity to support indigenous microbial populations. Autochthonous bioaugmentation (ABA) utilizes site-derived microbial strains for improved adaptability and persistence under local environmental conditions. Ozonation is applied as a pre-treatment to increase hydrocarbon bioavailability by partially oxidizing complex hydrocarbons, followed by aeration to sustain the activity and regrowth of indigenous microbial communities, ensuring long-term ecosystem recovery.

6. COMBINED BIOREMEDIATION OF 2,4-D HERBICIDE USING Fe_3O_4 NANOPARTICLES AND INDIGENOUS MICROBES

The combined use of Fe_3O_4 nanoparticles and native soil microorganisms has been shown to improve the degradation of herbicide residues like 2,4-dichlorophenoxyacetic acid (2,4-D). Fang et al. demonstrated that this synergistic approach outperformed treatments using either agent alone. Enhanced breakdown of 2,4-D into its intermediates –2,4-dichlorophenol, chlorophenol, and phenol – was observed, offering a viable solution for remediating pesticide-contaminated soils (Fang et al., 2012).

7. ROLE OF INDIGENOUS MICROORGANISMS IN NATURAL FARMING

In natural farming, indigenous microorganisms (IMOs) support plant nutrition through the development of my-

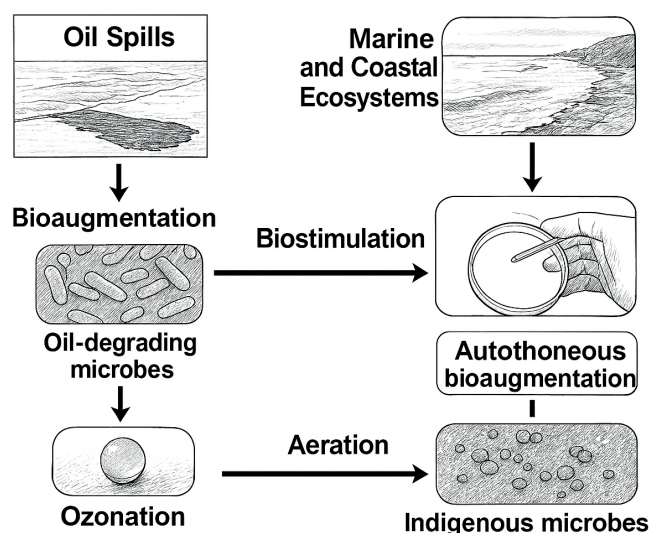


FIGURE 2: Integrated bioremediation framework. Conceptual graphics for microbial degradation of oil spills in marine and coastal environments. The process involves bioaugmentation, biostimulation, and ozonation.

corrhizal fungi, which form symbiotic associations with plant roots and extend root networks via extensive fungal filaments. These fungi convert complex nutrients into simpler, bioavailable forms and promote soil carbon sequestration through glomalin production, which improves soil aggregation and structure. The use of IMO-based inoculants in farms across 30+ countries has shown improvements in soil texture, tilth, and earthworm activity, contributing to plant vigor and ecosystem resilience (Surpura et al., 2025).

7.1 Biocomposting with indigenous microorganisms

Traditional practices of burning agro-industrial residues such as paddy husk or corn stalk release harmful pollutants and contribute to soil erosion. Composting with IMOs offers a sustainable alternative, converting waste into nutrient-rich amendment. Recent research demonstrated that combining corn stalk with IMO inoculant yields compost with significantly better chemical properties compared to paddy husk-based compost. IMOs accelerate composting processes, enrich microbial diversity, and enhance soil fertility—making biocomposting both eco-friendly and economical (Surpura et al., 2025).

8. FACTORS INFLUENCING MICROBIAL BIOREMEDIATION

Environmental parameters critically shape the effectiveness of native microbe-based bioremediation. Key factors include:

- **pH:** Near-neutral pH levels optimize bacterial growth and metabolic activity, accelerating biodegradation rates;
- **Temperature:** Mesophilic temperatures (20-40°C) support efficient microbial activity, while thermophilic species can perform degradation at elevated ranges (60-70°C);
- **Water Content:** Soil moisture levels with water activity

≥ 0.9 are essential for microbial metabolism and contaminant breakdown;

- **Soil Texture:** Porous, well-structured soils enhance oxygen diffusion and microbial mobility, improving remediation kinetics;
- **Nutrient Levels:** Supplementing nitrogen and phosphorus boosts microbial growth and contaminant degradation rates;
- **Electron Acceptors:** Adequate oxygen availability, or addition of oxidants like hydrogen peroxide, supports aerobic degradation;
- **Bioavailability:** Adding biosurfactants or solvents can improve desorption of hydrophobic pollutants from soil particles;
- **Cometabolism:** Some microbes degrade non-growth substrates (e.g., DDT, PCBs) in the presence of growth-supporting co-substrates;
- **Gene Expression:** Sufficient pollutant concentration is required to induce expression of degradative enzymes and operons.

These elements must be optimized to harness the full potential of indigenous microbes for in situ remediation (Sonawane et al., 2022).

8.1 Aerobic bioventing and cometabolic strategies

Aerobic bioventing injects air or oxygen into contaminated unsaturated soil, stimulating native microbial degradation of hydrocarbons and semi-volatile organic compounds like benzene, toluene, phenol, and light PAHs. Cometabolic bioventing involves adding volatile substrates (e.g. propane, methane) alongside air, which induces enzyme systems (e.g., monooxygenases) capable of transforming chlorinated solvents such as TCE or DCE. Successful in situ applications have demonstrated that indigenous microbial consortia can support simultaneous treatment of multiple dilute contaminants when engineered properly, though substrate competition and toxicity must be monitored (Skinner et al., 2024). Figure 3 is a schematic representation

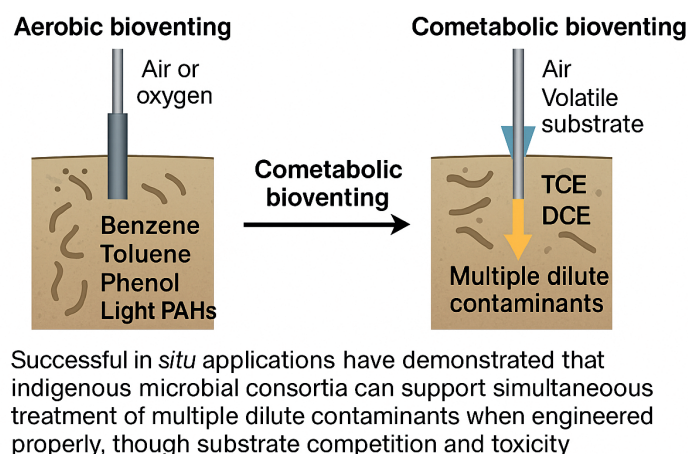


FIGURE 3: Mechanism of Bioventing Strategies. Aerobic bioventing and comatabolic bioventing strategies for in situ remediation of hydrocarbon- and solvent-contaminated soils.

showing the mechanisms of aerobic and comatabolic bioventing. In aerobic bioventing, air or oxygen is injected into the unsaturated zone to stimulate indigenous microbial communities that degrade hydrocarbons and semi-volatile organic compounds such as benzene, toluene, phenol, and light PAHs. In comatabolic bioventing, air is co-injected with volatile substrates (e.g., methane, propane) that induce microbial enzyme systems such as monooxygenas-

es, enabling the transformation of chlorinated solvents including trichloroethylene (TCE) and dichloroethylene (DCE) (CLU-IN, 2020). Field applications have demonstrated that properly engineered systems can support simultaneous degradation of multiple dilute contaminants by indigenous microbial consortia, though substrate competition and toxicity must be managed. Table 2 provides a comparative overview of aerobic and comatabolic bioventing strategies

TABLE 2: Global aerobic and comatabolic bioventing strategies for in situ bioremediation of contaminated soils.

Country / Region	Contaminant Type	Strategy Applied	Key Microbial / Process Insight	Outcome / Efficiency
United States	BTEX (Benzene, Toluene, Ethylbenzene, Xylene)	Aerobic Bioventing	Air injection stimulated native hydrocarbon-degrading bacteria (<i>Pseudomonas</i> , <i>Acinetobacter</i>) in unsaturated soil	>90% BTEX removal within 6 months
Canada	Diesel and light PAHs	Aerobic Bioventing	Continuous oxygen flow increased the oxidation rate of hydrocarbons by indigenous microbes	85–95% reduction of TPH; minimal secondary pollution
Germany	TCE, DCE	Comatabolic Bioventing	Methane-induced monooxygenase activity by methanotrophic consortia facilitated chlorinated solvent degradation	Complete TCE removal in ~150 days
Japan	Aromatic hydrocarbons, chlorinated solvents	Aerobic + Comatabolic Hybrid	Co-injection of methane + oxygen induced <i>Pseudomonas putida</i> and methanotroph enzyme systems	Dual degradation of hydrocarbons and chlorinated solvents; stable microbial population
India (Vadodara, Gujarat)	Petroleum hydrocarbons, phenol, and light PAHs	Aerobic Bioventing	Indigenous consortia (<i>Bacillus</i> , <i>Pseudomonas</i> , <i>Rhodococcus</i>) enriched from refinery soils under oxygen injection	70–95% degradation within 90 days; reduced phenol toxicity and improved soil respiration
India (Assam Oil Fields)	Crude oil and diesel residues	Aerobic + Bioaugmentation	Integration of oxygen sparging with native <i>Alcanivorax</i> and <i>Marinobacter</i> strains	Enhanced hydrocarbon breakdown in 45 days; increased biosurfactant production
China (Dagang)	TCE, DCE in industrial soil	Comatabolic Bioventing	Methane + oxygen injection promoted <i>Methylosinus trichosporium</i> activity	80% solvent removal; reduced substrate competition under optimised ratios
Norway (Arctic Marine Sediments)	Weathered crude oil	Aerobic Bioventing	Oxygen diffusion supported <i>Alcanivorax</i> and <i>Cycloclasticus</i> spp.	>99% n-alkane and >90% PAH removal
Australia	Hydrocarbons, semi-volatile compounds	Aerobic Bioventing	Nutrient-supplemented oxygen injection enhanced microbial metabolism	Efficient degradation with minimal volatilisation
Poland	Chlorinated pesticides (DDT, HCH)	Comatabolic Bioventing	Propane-induced cometabolism under controlled aeration	Slow kinetics; >60% DDT transformation after 12 weeks
South Korea	Mixed hydrocarbon and chlorinated solvent contamination	Sequential Aerobic–Comatabolic	Alternating oxygen + methane pulses enhanced cometabolic enzyme expression	Simultaneous degradation of BTEX and TCE achieved >85% removal efficiency

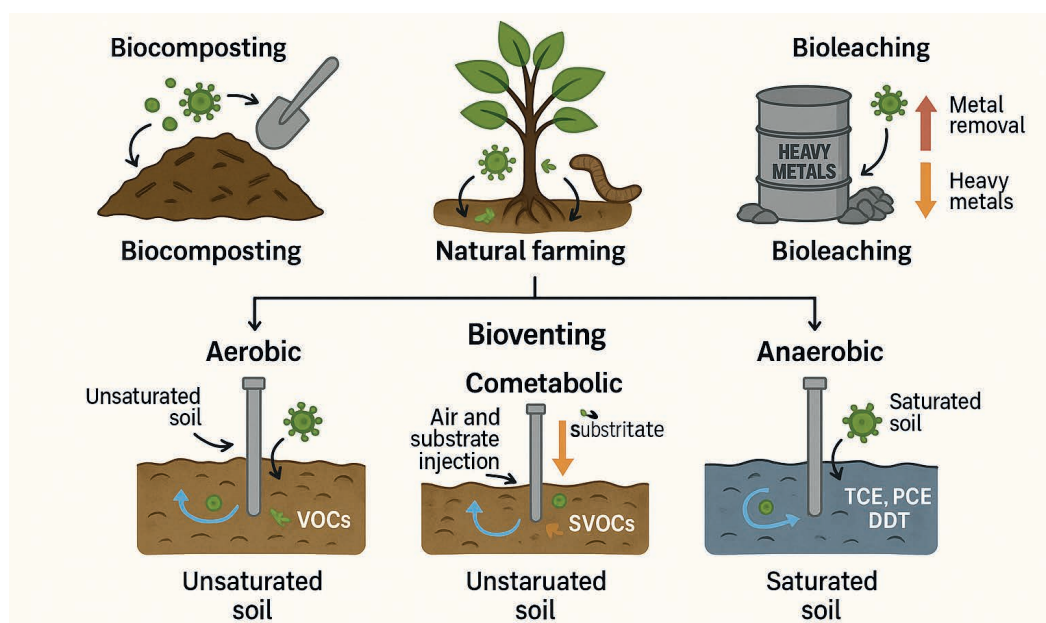


FIGURE 4: Bioremediation with Indigenous Microorganisms. This figure summarizes the multifaceted roles of indigenous microorganisms (IMOs) in environmental remediation strategies. It illustrates the three primary bioventing approaches— aerobic, cometabolic, and anaerobic—highlighting their applications in removing hydrocarbons and chlorinated compounds under varying subsurface conditions.

implemented worldwide, emphasizing in situ applications that use indigenous microbial consortia for the degradation of hydrocarbons and chlorinated solvents. Indian case studies demonstrate the potential of refinery-derived native microbes and soil bioaugmentation under oxygen-enriched conditions to achieve high hydrocarbon removal efficiency while maintaining ecological stability.

8.2 Limitations of cometabolic bioventing

Cometabolic bioventing faces key challenges related to gas distribution in subsurface environments. In low-permeability soils or those with high moisture content, it becomes difficult to uniformly deliver both oxygen and co-substrate gases. As per Federal Remediation Technologies Round table (FRTR), newer tools such as Technology Screening Matrix can improve the designing of Bioventing Designing systems to effectively reach shallow contamination zones without causing vapor intrusions or inadequate aeration presents significant engineering complexities (<https://frtr.gov/matrix/bioventing/>). The technology is relatively novel and lacks broad field experience. While cometabolic bioventing has shown promise in limited pilot projects, its performance across diverse contaminant profiles remains uncertain. Regulatory acceptance and public familiarity are still evolving, and comprehensive treatability and pilot studies are essential before full-scale deployment. As more applications are pursued, these constraints may diminish (Fascari et al., 2015).

8.3 Anaerobic bioventing

Anaerobic bioventing extends the concept of gas delivery into subsurface zones by injecting inert gases like nitrogen, along with electron donors such as hydrogen or CO_2 , to establish reducing conditions conducive to anaerobic dechlorination. This method is particularly suited for treat-

ing highly chlorinated solvents, such as PCE, TCE, RDX, and DDT, which resist aerobic breakdown. However, the effectiveness of this approach heavily depends on the ability to uniformly distribute gases within the soil matrix. Soils that are waterlogged or low in gas permeability may require complex system designs and extended remediation timelines—often spanning multiple years—making it less suitable for time-sensitive cleanup efforts (Fascari et al., 2015). Figure 4 depicts the beneficial impacts of IMOs in natural farming, bio-composting, and metal removal, showcasing their ecological compatibility and synergistic potential in improving soil structure, enhancing nutrient cycles, and detoxifying pollutants. The interconnected arrows emphasize the integration of these microbial processes into sustainable environmental management. Key elements shown:

- Mycorrhizal symbiosis improving plant-soil interactions;
- Bioventing systems using oxygen or alternative gases for in situ soil remediation;
- Microbial cometabolism pathways utilizing added organic substrates;
- Application of IMOs in agro-industrial waste management and bioleaching of heavy metals;
- Future prospects for climate-resilient, microbe-driven sustainable agriculture.

9. CONCLUSIONS

Environmental sustainability has become a research priority, driving interest in technologies that minimize ecological impact. Indigenous microorganisms offer multifunctional benefits, including contaminant degradation (hydrocarbons, chlorinated solvents, herbicides), nutrient cycling (nitrogen fixation, phosphorus solubilization), soil fertility enhancement, and natural hormone production

for plant growth. Because IMO's consist of diverse, acclimated microbial consortia, they integrate effectively into soil ecosystems and often outperform cultured strains in resilience and ecological compatibility. When inoculum density is adequate, IMO's establish positively associative communities that can exert control over native soil microbiota, promoting ecosystem stability and crop productivity. However, further research is needed to optimize strain selection, application protocols, operational conditions, and field-scale performance metrics. Addressing these gaps will enable IMO's to play a central role in future sustainable agriculture and environmental restoration strategies. The review highlights the ecological and functional contribution of Indigenous Microorganisms (IMO's) in promoting waste conversion, soil fertility improvement, and pollutants treatment. The discussion enriches that IMO's, in consideration for locality, as a sustainable and resilient tool for ecological restoration. Despite this, several major knowledge gaps still exist, including the best practices regarding the selection of strains, standardization of method application across different ecosystems and understanding long-term ecological effects under field conditions. Future research needs to focus on the integration of microbial diversity with quantifiable restoration results through field level assessments. Reinforcing these avenues for research would push towards the practical use of IMO's in large-scale, cost-effective bioremediative and sustainable agriculture systems that contribute to global agenda on environmental resistance and resource circularity.

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BRIDGING THE GAP: HOW GHG ACCOUNTING TOOLS CAN SUPPORT BEER COMPANIES IN THE IMPLEMENTATION OF CIRCULAR ECONOMY STRATEGIES

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ABSTRACT

This study presents a greenhouse gas (GHG) accounting tool developed for Italian breweries aimed at the quantification of the carbon footprint (CF) of organizations, thus enabling a self-assessment of their contribution to climate change. The tool builds on a Microsoft Excel interface that allows breweries to input site-specific data related to materials, energy, emissions, and waste across their life cycle in accordance with both the GHG Protocol and the ISO 14064-1 standard. A methodological framework is developed based on 6 steps, which include: i) organizational and operative boundaries definition, ii) identification of the databases for the calculations of emission sources and emission factors, iii) calculation of CF which allows iv) to perform a screening study and the hotspot identification, followed by v) a check with literature to vi) perform a CE strategies assessment. This procedure made it possible to build a valuable and reliable tool, in line with international standards and easy to replicate in other contexts. The tool indeed enables companies to identify environmentally relevant processes (hotspot analysis) and to implement relevant circular ecosystem innovation. This is possible thanks to its ability to create collaboration within ecosystem actors and allow a trial-and-error process.

1. INTRODUCTION

1.1 The role of GHG accounting tools as enablers of circularity

The awareness of living in a linear economy, where we take resources, make products, use them and discard them, while recognizing the limits of planetary resources and energy use, in a world that is a "system" threatened by pollution and waste, build the basis for circular economy (CE) thinking (Bocken et al., 2016; Konietzko et al., 2020).

The CE's main principles are the maximization of resource value and the minimization of emissions, resource consumption, waste, and pollution (Ghisellini et al., 2016; Kirchherr et al., 2017). Among the several studies concerning the integration of CE into businesses, the one of Konietzko et al. (2020) identified four main strategies supporting and enabling CE in business: narrowing (use less material and energy), slowing (extend products and components lifespan), closing (reuse products and components) and regeneration (avoid toxic materials, use renewable energy and regenerate ecosystems). This approach highlights the importance of implementing CE throughout

the supply chain from input materials to final products, representing an opportunity and challenge for businesses, including businesses in the beverage industry (Ho et al., 2024). Actors in the supply chain indeed play a crucial role in the implementation and success of the CE, serving as the foundation for driving the necessary changes (Hazen et al., 2020).

Among the instruments which help businesses in making CE-related decision along their supply chain, the Carbon Footprint (CF) is a highly useful decision-making tool that allows organizations to have a deeper knowledge and, consequently, to exert a greater degree of control over the potential environmental impacts of their activities in relation to Greenhouse Gases (GHG) emissions (Valls-Val and Bovea, 2022). GHG accounting operates as the quantitative backbone of CE strategy design and monitoring. Carbon accounting within the CE has emerged as a key priority for professional organisations, as climate change represents a pressing global challenge and carbon emissions continue to rise worldwide. Although research on carbon accounting is still relatively recent, both public and private entities are

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increasingly required to quantify their GHG emissions, not only to comply with sustainability goals but also to reduce operational costs and enhance overall efficiency (Ionescu, 2023). At the organisational level, inventories built under the GHG Protocol Corporate Standard and ISO 14064-1 standard make visible direct and indirect emissions across the value chain, thus enabling firms to prioritise interventions and coordinate actions with ecosystem partners.

Recent organisational CF tools (e.g., for universities) further show how routine accounting, dashboards, and scenario screening can guide internal governance and external reporting (Valls-Val & Bovea, 2022). In line with circular ecosystem innovation principles, emphasising experimentation, collaboration, and platformisation, tools translate measurement into decision-relevant insights, supporting strategies that narrow, slow, close, and regenerate material and energy flows (Konietzko et al., 2020).

Nevertheless, the existing body of literature provides limited in-depth analyses on the potential of GHG accounting and reporting tools explicitly designed to support the identification and implementation of CE strategies. To address this gap, the present study initiates a research stream at the intersection of GHG accounting and CE strategies, proposing the development of accessible and user-friendly tools to facilitate their practical adoption.

In addition, GHG accounting tools, which build on the same logic and methodological assumptions of Life Cycle Assessment (LCA) tools, are among the simplification strategies following the logic of LCA standardisation (Beemsterboer et al., 2020). Their appeal is particularly strong for non-expert users, as they typically combine a user-friendly and customisable interface with the ability to generate immediate feedback (Beemsterboer et al., 2020). These features make them a powerful instrument for informing strategic decision-making and advancing the transition towards CE practices.

1.2 The Carbon Footprint accounting tool in the agrifood sector

In agri-food value chains, CF complements product-level LCA by providing comparable, year-on-year monitoring of operations and supply chains. In particular, the brewing industry is one of the biggest consumers of water and energy and one of the largest industrial producers of wastewater, solid waste, by-products, and emissions to air (Olajire, 2020). In addition, within the food industry, the brewing sector occupies a strategic position due to its high volumes of production, reaching 1,88 billion hl in 2023. Moreover, beer is the most popular alcoholic beverage worldwide, and the third-most popular drink overall after water and tea (Statista source, 2025).

The use of GHG accounting tools provides valuable support for monitoring value chain activities, responding to the urgent need for reporting and management improvements emerging in the agri-food sector, which accounts for 37% of global GHG emissions (Alromaizan et al., 2023).

Among agri-food companies, the brewing sector is particularly challenging with respect to the reduction of climate-changing emissions. Literature consistently iden-

tifies hotspots in agriculture (malting barley and hops), packaging (glass and aluminium), thermal energy for brewing and utilities, and distribution logistics (Olajire, 2020).

European Beer Product Environmental Footprint Category Rules (PEFCR) guidance and industry sources stress the importance of life-cycle thinking and harmonised datasets for comparability across formats (The Brewers of Europe, 2020; Beer PEFCR). However, most applications provide an up-to-date snapshot of the impacts on climate change (Bowler et al., 2024) without converting CF evidence into a structured, repeatable process to screen CE strategies for breweries.

1.3 Research gap and aim of the study

Despite growing interest in CE within brewing, as demonstrated by several international studies exploring the integration of CE strategies within the beer industry (Ho et al., 2024), some barriers remain, and a practical, standards-aligned organizational CF tool tailored to breweries and explicitly geared toward CE strategy assessment is still missing. The agri-food supply chain is a highly complex network, and effective CE strategies must engage all the actors of the supply chain (Esposito et al., 2020). In addition, there are sector-specific barriers, like internal company barriers or market barriers, to overcome (Angova, 2023).

In this contest, to spread the use CF methodology to support beer companies in the assessment of their CE strategies, the development of a calculation tool could be a valuable path to pursue. Therefore, with this paper, we aim to fill this gap by addressing the following research questions:

- RQ1: How is it possible to build a valuable GHG accounting tool supporting beer companies in CE strategies assessment?
- RQ2: In which way could a GHG accounting tool help beer companies to overcome the barriers in the adoption of CE practices?

The overall objective is thus to demonstrate how to develop a reliable and complete GHG accounting tool supporting Italian breweries in the transition towards a more sustainable and CE model. The tool facilitates companies in GHG accounting activities; the user-friendly interface is designed to be attractive for users and encourages data collection in the framework of a wider company commitment to measure and reduce GHG emissions. It allows for pre-assessing the potential effect on climate change resulting from the implementation of CE strategies, to compare results, and to replicate the study over the years, since it is based on the same standardized methodology.

The remainder of this paper is organized as follows. Section 2 illustrates the materials and the methodology behind the GHG accounting tool, with an introduction to the reference standards and the description of the assessment process. Section 3 provides the results of the GHG accounting tool design and testing, as well as outlining the possible barriers to the development of CE strategies in the beer sector, the opportunities given by a GHG accounting

tool, and future developments. Finally (Section 4), conclusions are provided addressing the research questions.

2. MATERIALS AND METHODS

2.1 Methodological framework

The methodological framework developed in the context of the present study is detailed as follows and illustrated in Figure 1.

The first activity is the analysis and comparison of available GHG accounting standards (GHG Protocol and ISO 14064-1); then there is the definition of organizational and operational boundaries; the database with the identification of emission sources (ES) and emission factors (EF) is then built, and calculation formulas are set. The GHG accounting tool is then tested by input inventory data collected from a sample of selected representative companies. This screening study allowed the identification of environmental hotspots. The reliability of the results was checked by reviewing studies within the available literature. After a positive check, the priority CE strategies to be assessed using the GHG accounting tool were identified.

2.2 GHG Protocol and ISO 14064-1

GHG Protocol and ISO 14064-1 standard provide requirements for quantifying the GHG impact of an organization (Gao et al., 2014), or in other terms, its CF.

The GHG Protocol Initiative is a multi-stakeholder partnership of businesses, non-governmental organizations (NGOs), governments, and others convened by the World Resources Institute (WRI) and the World Business Council for Sustainable Development (WBCSD) (WRI, 2004). In 2004, the Initiative published the GHG Protocol Corporate Accounting and Reporting Standard, a document providing a step-by-step guide for companies to use in quantifying and reporting their GHG emissions. It provides sector-specific and general calculation tools and deals with the quantification of GHG reductions resulting from the adoption of mitigation methods in its project protocol (Gao et al., 2014).

ISO 14064-1 standard details principles and requirements for designing, developing, managing, and reporting

organization-level GHG inventories released in 2006 by the International Standard Organization (ISO). It includes requirements for determining GHG emissions and removal boundaries, quantifying an organization's GHG emissions and removals, and identifying specific company actions or activities aimed at improving GHG management. It also includes requirements and guidance on inventory quality management, reporting, internal auditing, and the organization's responsibilities in verification activities (ISO, 2018).

During the development of the two assessment standards, efforts were made to harmonize the methodologies, but some minor differences remain. Table 1 summarizes the different aspects of the two methodologies.

Both standards require quantifying direct GHG emissions separately for all Kyoto Protocol GHGs, such as CO₂, CH₄, N₂O, NF₃, SF₆, and other appropriate GHG groups (HFCs, PFCs, etc.) in tonnes of CO₂e and GHG removals. The two standards require defining the organizational boundaries following the same two alternative approaches: control and equity share. On the contrary, reporting boundaries are set in a different way in the two standards. As for the quantization of CF, the two standards give several different quantification methods. However, the quantification based on GHG activity data multiplied by GHG emission or removal factors is recommended and widely used (Gao et al., 2014).

In this study, the CF tool for Italian breweries has been developed according to both the GHG Protocol and the ISO 14064-1. Both guidelines follow an approach based on the LCA methodology to assess GHGs corporate inventory annual emissions.

2.3 Organizational and operational boundaries

As for ISO 14044, the first step of an LCA is the definition of the goal and scope, which includes the identification of the system boundaries of the study. In the context of a corporate CF assessment, the company shall define its organizational boundaries. The organization may comprise one or more facilities, and emissions or removals may originate from one or more GHG sources or sinks. Organizational boundaries setting is the way a company consolidates its facility-level GHG emissions and removals

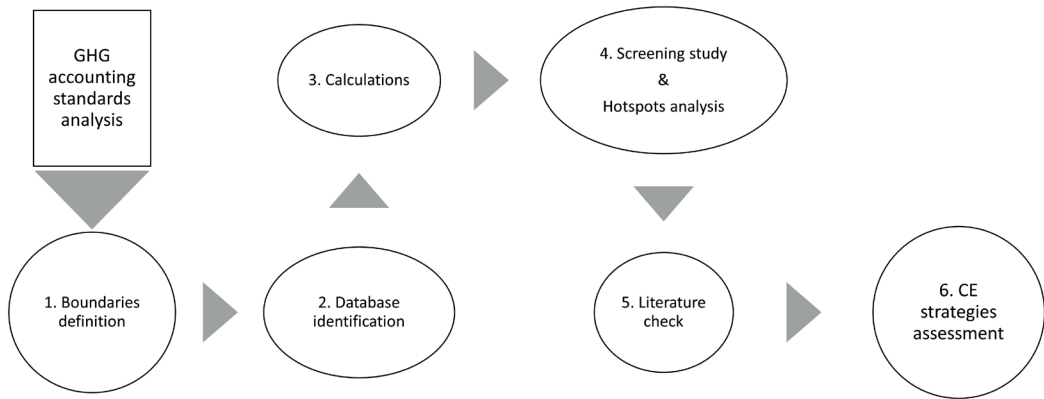


FIGURE 1: Methodological framework based on a 6-steps approach followed in the study. Procedural steps followed to design a GHG accounting tool allowing the assessment of possible circular economy strategies within the beer industry sector.

TABLE 1: Comparison of ISO 14064-1 and GHG Protocol.

	ISO 14064	GHG Protocol
Publisher	ISO	WRI&WBCSD
Date	2006, 2018 (update)	2004, 2011 (revise)
Goal	To define principles and guidelines for the accounting, reporting and verification of an organization's GHG inventory	
Principle	Relevance, completeness, consistency, accuracy, transparency	
GHG	Kyoto Protocol GHGs	
GWP	Latest IPCC's GWP	
Organizational boundaries	Same approach for consolidating GHG emissions: equity share and control (financial and operational)	
Operational/Reporting boundaries	Category 1 (direct emissions), Category 2 (energy indirect emissions), Categories 3-6 (other indirect emissions)	Scope 1 (direct emissions), Scope 2 (energy indirect emissions), Scope 3 (other indirect emissions)
Indirect imported energy emissions accounting method	Compulsory location-based approach, optional market-based approach	Compulsory dual reporting (location and market-based approaches)
Treatment of biogenic emissions and removals	Anthropogenic biogenic emissions and removals shall be quantified and reported separately. Non-anthropogenic emissions and removals may be quantified and reported separately.	Emissions from biomass or biologically sequestered carbon shall be reported separately from the scopes

according to one of the following two approaches: equity share and control.

Under the control approach, a company accounts for all GHG emissions from operations and facilities over which it has control. Control can be defined in either financial or operational terms, and companies shall choose one of the two criteria. The financial control takes place when a company controls the operation of another company by directing the financial and operating policies of the latter with the goal of having economic benefits from its activities. The operational control is realized when a company controls the operations of another company with the full authority to introduce and implement its operating policies.

The equity shares approach lets a company account for GHG emissions and removals from operations according to its share of equity in the operation. This approach reflects the company's economic interest in terms of risks and rewards flowing from an operation, and it is aligned with the company's percentage ownership of that operation. Thus, both the GHG Protocol and ISO 14064-1 suggest the same approaches to define organizational boundaries. In the context of the present study, organizational boundaries follow the operational control approach.

Operational boundaries consist of the identification of the emissions associated with the company operations, included in the organizational boundaries, and categorizing them as direct and indirect emissions.

According to the GHG Protocol, emissions are classified in scopes:

- Scope 1 considers all direct emissions arising from stationary combustion, mobile combustion, industrial processes, and fugitive emissions;
- Scope 2 accounts for indirect emissions from imported energy flows.
- Scope 3 includes all indirect emissions generated from other sources. Indirect emissions are further categorized into 15 categories, and the company should perform a screening study to determine which are the

most relevant according to its business goals and following some relevance criteria. The criteria are the size of the contribution to the overall impact, the influence on potential emissions reduction, the risk exposure of the company, the importance for stakeholders, the outsourcing of activities, the presence of sector guidance, and the correlation with spending or revenues for the company. These criteria indicate the level of detail and the type of effort that should be dedicated to each accounting category.

ISO 14064-1 classifies emissions in 6 main category groups, further detailed in many subcategories. The first and second categories correspond to respectively first and second scopes of the GHG Protocol, the remaining categories account for all other indirect emissions, considering the same level of analysis as the GHG Protocol. Also, the ISO standard indicates some criteria to evaluate in a screening step the significance of indirect emissions, which are: magnitude of the impact, level of influence, contribution to the company's risk or its business opportunity, presence of sector-specific guidance, outsourced activities and employee engagement to emission reduction.

The CF assessment tool is designed to generate results in line with both GHG Protocol and ISO 14064-1 requirements.

2.4 Database

2.4.1 Emission sources

In order to identify the emission sources to be included in the calculation tool, an interview with the representatives of the main brewery companies involved in the study was conducted.

Three representative companies have been reached: one craft, one medium, and one big company, thanks to the collaboration of Fourgreen srl, an intermediary company providing services to HORECA businesses. Their involvement aimed to gain a reliable and comprehensive

understanding of all processes that could vary depending on the company's size. This screening process is necessary to design a GHG accounting tool covering all potential processes and activities and being representative of the Italian brewery sector. The interview was guided by a draft checklist for input data collection that had been customized according to the breweries' suggestions.

In some cases, where information on beer life cycle processes was not available, secondary data were obtained thanks to the Product Environmental Footprint (PEF) screening report (The Brewers of Europe, 2015) and PEF Category Rules (PEFCR) of beer (The Brewers of Europe, 2020) and the PEF screening report of Packed Water (The European Federation of Bottled Waters, 2015). Generic processes, like an average meal delivered at the canteen, have been modelled thanks to information obtained from an LCA study of an international conference series in Europe (Neugebauer et al., 2020).

A complete list of the default emission sources included in the CF tool database is reported in the Appendix. The tool can be further customized by adding other ES to adapt to the specific requirements of the case study under analysis.

2.4.2 Emission factors

The database includes default emission factors (EF) corresponding to the default emission sources. Since the tool is designed for Italian companies, some EF are specific to the Italian context and come from governmental Italian sources such as the natural gas emission factor (ISPRA, 2022). Other EF are provided by European authorities such as the Association of Issuing Bodies (AIB) that each year publishes the annual results of the calculation of residual mixes (AIB, 2024) and the rest are taken from international institution like the UK Department of Environment, Food and Rural Affairs (DEFRA), the US Environmental Protection Agency (EPA) or commercial database like Ecoinvent 3.10, Agribalyse 3 and the World Food LCA Database (WFLDB) using the life cycle impact assessment method IPCC 2021 GWP100 – total based on the latest version of the IPCC report "AR6 Climate Change 2021: The Physical Science Basis" through the LCA software SimaPro 9.6.

The detailed list of default Scope 1 and Scope 2 EF used in the GHG accounting tool is presented in the Appendix.

The list of default emission factors for Scope 3 categories is not included in the present paper. The sources of indirect emission factors are the databases Ecoinvent 3.10., Agribalyse 3 and the World Food LCA Database (WFLDB).

2.5 Calculations

The calculations behind the results implemented in the tool consider for each emission source the corresponding EF multiplied by the activity data, which are defined as the quantitative information related to any life cycle process inventoried within the system boundaries, directly measures emissions and consumptions of all life cycle activities. Examples of activity data are the kg of gaseous emissions, the m³ of water withdrawal, the kWh of electric energy consumption, or the km of vehicle transport, given as quanti-

tative information related to the emission sources in the input data spreadsheet.

The formula set in the tool that automatically calculates CO₂e emissions is the following:

$$CF = \sum_{i=1}^n AD * EF \text{ [ton CO}_2\text{e]} \quad (1)$$

Where:

i represents the number of emission sources (ES),

AD is the activity data collected for each ES,

EF is the emission factor relative to each ES,

CF is the total carbon footprint expressed in tonnes of equivalent CO₂.

3. RESULTS AND DISCUSSION

3.1 Tool design and testing

The tool was designed using Microsoft Excel and structured in two main modules: i) input data, containing the collection of all activity data, and ii) output data, presenting the results of the calculation, as described in sections 2.4 and 2.5.

Figure 2 illustrates the Instruction sheet in which the user can find useful information on how to fill in the document and how to read the results shown in the output spreadsheets.

The tool was tested by asking the three breweries involved in the design of the tool to collect data and compile the input spreadsheet. After the testing period, the tool has been improved in order to be clearer and easier to compile for non-expert users.

3.2 Input data

Input data required by the tool comprises some general information about the company, the scope of the study, like the organizational and operational boundaries, the reporting period, and the data owner involved in data collection. The specific input data spreadsheet corresponds to the life cycle inventory analysis (LCI) phase, as conceived by ISO 14040 on LCA methodology, and guides the collection of all activity data for every scope (GHG Protocol approach) or category (ISO 14064-1), as defined in the operational boundaries.

Figure 3 shows an extract of the input data spreadsheet.

3.3 Output data

After compilation of input data, the tool automatically calculates the CF of the organization and shows the results in different ways, one for each output data spreadsheet, both in table and graph format, in order to make them easier to interpret.

General results are first represented in two different tables: the first, in one spreadsheet, is referred to GHG Protocol and reports results following both location and market-based approaches, the second, in another spreadsheet, is referred to as ISO 14064-1, reporting only location-based results. Both tables show the amount of CO₂e emissions related to Scope 1 or Category 1, distinguished for each relevant Kyoto Protocol greenhouse gas. Indirect emissions (Scope 2 and Scope 3, or categories from 2 and on) are

Il Tool è composto da diversi fogli di lavoro, di cui uno, il foglio "Dati di Input", permette l'inserimento dei dati in Input da parte dell'Utente e gli altri sono fogli di Elaborazione dati e Output dei risultati visualizzabili solo in lettura.

Per determinare i processi e le attività dell'Organizzazione su cui effettuare lo studio, in linea con la Norma ISO 14064, il Tool fornisce all'Utente uno strumento per effettuare una **Analisi di Significatività preliminare**, con l'obiettivo di individuare i processi e le attività dell'Organizzazione significative.

L'Analisi di Significatività è fornita nel Foglio di lavoro "Significatività". I processi che risultano avere una significatività bassa possono essere esclusi dallo studio della CFO.

Foglio di lavoro "Dati di Input"

Il foglio di lavoro "Dati in Input" permette all'Utente di fornire in ingresso le informazioni necessarie per il calcolo dell'impronta carbonica di Organizzazione secondo gli standard GHG Protocol e ISO 14064.

La compilazione dei dati in ingresso richiede la conoscenza di informazioni dettagliate relative alle diverse attività aziendali.

Come informazione preliminare viene richiesto di fornire dati sui volumi di vendita dell'anno di rendicontazione, sia in termini aggregati in hl, sia in termini più dettagliati specificando il numero di pezzi venduti o i kg per ogni tipologia di prodotto.

Queste informazioni sono funzionali per il calcolo di dati necessari per diverse attività, come, per esempio, la fase d'uso dei prodotti venduti o il loro fine vita.

In seguito, la schermata di Input si sviluppa secondo la classificazione prevista dal GHG Protocol, suddividendo le attività aziendali in Scope 1, Scope 2 e Scope 3. Lo Scope 3 è a sua volta suddiviso nelle Categorie ritenute significative.

Per rendere più chiara la richiesta del dato da inserire, il Tool fornisce, nella colonna Note, informazioni o spiegazioni aggiuntive con l'obiettivo di agevolare l'Utente

Il dato va inserito nella corrispondente cella di colore chiaro indicata di seguito dalla freccia. Va inserito SOLO la quantità espressa in numero, NON inserire unità di misura o altre informazioni

Combustione fissa	Quantità	Unità di misura	Note
Gas naturale (per caldaie e cogeneratore)		smc	
GPL (es: per riscaldamento)		l	
Diesel		l	

FIGURE 2: Instruction sheet extract of the Greenhouse Gases (GHG) accounting tool. The sheet presents detailed instructions for completing the data input sheet, as well as guidance on how to interpret the resulting outputs (revised caption; high-resolution image to be provided by authors).

SCOPE 1

Note

Combustione fissa	Quantità	Unità	Note
Gas naturale (per caldaie e cogeneratore)		smc	Inserire i consumi annuali dei combustibili impiegati negli impianti di combustione fissa (es. caldaie)
GPL (es: per riscaldamento)		l	
Diesel		l	
Biomassa (cippato di legno)		ton	
Biodiesel ME (Estere Metilico)		l	
Biodiesel ME (da olio da cucina usato)		l	
Biodiesel ME (da sego, grasso animale)		l	
Biodiesel HVO		l	
Bioetanolo		l	
Biometano compresso		kg	
Biometano liquefatto		kg	

Combustione mobile			Note
Auto diesel	l	km	Inserire il valore richiesto in litri o, in alternativa, in km percorsi
Auto ibrida diesel	l	km	
Auto ibrida benzina	l	km	
Auto benzina	l	km	
Auto gpl	l	km	
Auto metano	kg	km	
Furgone diesel	l	km	
Furgone benzina	l	km	
Furgone ibrido/elettrico	l	km	
Furgone gpl	l	km	
Furgone metano	l	km	
Camion diesel	l		
Altri macchinari a combustione interna - diesel (es. tosaerba, mezzi movimentazione carichi)	l	km	

...

Istruzioni

Significatività

Dati di input

Risultati GHG Protocol

Risultati ISO 14064

GHG Biogeniche

DashBoard

Input Incertezza ISO

Risultati Incertezza

FIGURE 3: Input data sheet extract of the GHG accounting tool. The sheet contains the list of all input and output processes inventoried to be filled with the corresponding activity data by each company (revised caption; high-resolution image to be provided by authors).

expressed in terms of tons of CO₂e without any distinction among specific GHGs. A third table in the following spreadsheet contains results related to biogenic CO₂ emissions that shall be reported separately from the results of the other scopes or categories.

Detailed results relative to direct, indirect from imported energy, and indirect from other sources emissions are presented in different spreadsheets, with the details of each emission source inventoried.

To conclude, a dashboard summarizing all the results in graph format is presented at the end of the document, and you can see an extract in the following Figure 4.

3.4 Selection of CE strategies in the beer ecosystem

After having performed a screening study on beer production for the three selected representative companies, a literature check was conducted to verify the compliance between our results and those of similar studies. Results suggest that, considering the environmental hotspots identified, possible CE strategies may include more efficient water consumption, via the prevention of water losses and the reuse of treated wastewater; energy efficiency improvements, like energy recovery implementation and renewable energy usage; gaseous emissions and solid waste reduction, both process and packaging waste (Olajire, 2020).

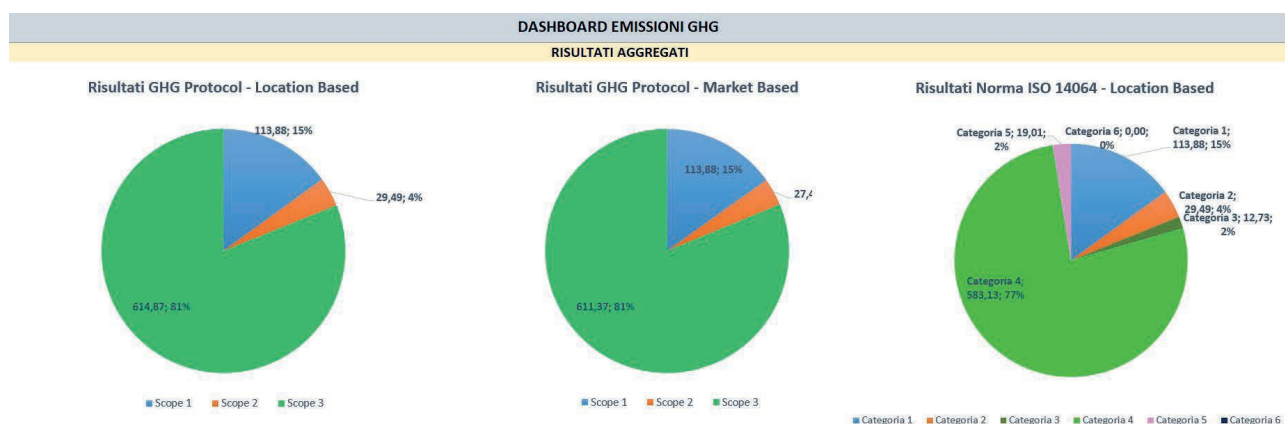


FIGURE 4: Dashboard summarizing results of the GHG accounting tool. The company carbon footprint assessment, according to both GHG Protocol and ISO 14064 standard, are presented in the form of pie charts, with varying levels of detail regarding the contributions (revised caption; high-resolution image to be provided by authors).

Studies demonstrate that the main waste generated by the brewing process is grain husks, yeast, and CO₂, but out of the three, spent grain is the most important one, making up around 85% of the total waste of a brewery. Literature results show that the gasification of spent grain minimizes the total volume of residue to be disposed of and can be used to produce syngas for heat generation at breweries, substituting fossil fuels (Ortiz et al., 2019). Consistently, another study has investigated the opportunity to convert malt residues into fuel and colorants as substitutes for raw materials (Jackowski et al., 2021).

An interesting experiment is the symbiotic solution created by a rooftop hydroponic aquaponic model integrated with breweries. The authors demonstrate that this new agroecological model allows the recycling of nearly all waste resources (malt residues, yeast residues, wastewater, rainwater, heat, electricity, and gaseous emissions), reducing costs and improving system sustainability (Horn et al., 2020).

CE strategies can also be applied to packaging management, like those aiming at building a continuous loop packaging system supported by the Cradle to Cradle® (C2C) certification program (Niero et al., 2017). Aluminium cans closed closed-loop recycling, or the use of returnable packaging, could be other solutions to be tested by the GHG accounting tool.

Echoing Konietzko et al. (2020) categorization proposal of CE strategies and after discussions on the feasibility of CE solutions with the beer companies that participated in the development of the screening study, we have identified and classified six different strategies, illustrated in Figure 5.

3.5 Discussion on opportunities arising from the tool

GHG accounting approach encourages companies to broaden their view to include all relevant actors in their value chain, from suppliers up to end users. In the same way, effective CE strategies require to involvement of different company stakeholders, like raw materials producers, distributors, or customers. Both innovation approaches

support the adoption of an ecosystem perspective, going beyond the product system and the business model. A business ecosystem is defined as a set of stakeholders, including producers, suppliers, service providers, and end users, who contribute to a common goal (Konietzko et al., 2020). Some studies have identified three main groups of principles guiding circular ecosystem innovation (Konietzko et al., 2020):

1. Collaboration within the ecosystem actors;
2. Experimentation via a trial-and-error process to implement greater circularity;
3. Platformization of interactions among organizations.

There is a need for a collaborative approach to catalyze the transition to circular business models and to implement circular practices efficiently. Engaging value chain partners in this transition starts with a good understanding of the processes and their potential for sustainable improvement. Based on the widespread assumption that only what is measurable is manageable, there is a clear need for a standardized, consistent, and high-quality method of measuring and reporting the environmental performance of organizations and products. Furthermore, standardization, considered as one of the simplification approaches used in LCA to manage the complexity of product systems and to meet user expectations regarding LCA results, while remaining consistent with the resources available to the analyst (Beemsterboer et al., 2020), can be another opportunity arising from the GHG accounting tool.

From the brewery sector perspective, the advantage is twofold: the first lies in the possibility to evaluate the relative sustainability of brewing processes by considering the CF among other sustainability parameters (Olajire, 2020). The second advantage is represented by the use of a shared assessment tool, enabling the comparison of circular performances and practices among the different stages of the life cycle of the brewing process and involving all the sectoral actors (Esposito et al., 2020).

Designing a GHG accounting tool that considers a wide range of emission sources, with relative emission factors, throughout the beer value chain, and enabling the pre-as-

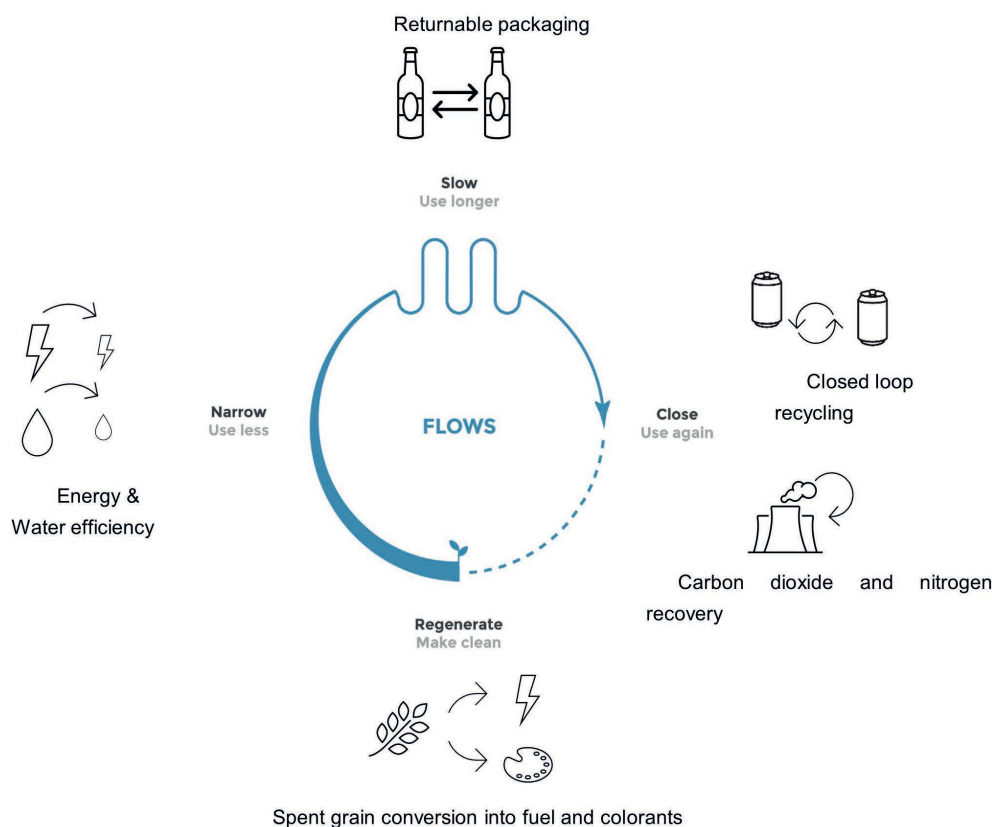


FIGURE 5: Circular economy (CE) strategies in the brewing sector. Readaptation from (Konietzko et al., 2020). Slowing, closing, regenerating and narrowing strategies applied to the beer sector with some suggestions on possible feasible solutions.

assessment of potential CE strategies is our proposal to support beer companies in the implementation of CE strategies.

The tool will increase companies' awareness of the potentially most impactful processes and the effectiveness of improvement actions with a view to the CE, leveraging greater involvement of value chain partners.

Practical implications include: (i) prioritizing supplier engagement for high-impact categories; (ii) integrating CF outputs into procurement Key Performance Indicators (KPIs); (iii) using the dashboard for internal communication and external reporting; and (iv) coupling the tool with CE pilots (narrowing, slowing, closing, regenerating) to enable iterative learning.

Our testing confirms that Scope 3 categories, particularly packaging, upstream agriculture, and energy supply, dominate organizational CF, in line with prior findings in brewing and food sectors (Olajire, 2020; PEFCR Beer). Beyond hotspot identification, the tool supports CE strategy formulation by quantifying potential improvements (e.g., renewable electricity market-based reductions; closed-loop aluminium; spent-grain valorization), and by structuring multi-actor dialogues around material and energy loops.

From a managerial point of view, the GHG accounting tool helps to develop the CF of breweries, thus allowing those companies to have critical data for external communication. As demonstrated also by (Rossi et al., 2025), an ecodesign tool, as well as a GHG accounting tool, can

be considered as green marketing instruments, providing the foundation for developing an emission-reduction management strategy which, when effectively communicated through suitable indicators and KPIs, can move beyond a purely technical function to become a valuable instrument of strategic communication with stakeholders.

Implications for practitioners and value chain actors relate to the multiplicity of applications for the tool: managers can employ it to establish baselines, identify emission hotspots, compare alternative CE scenarios, and track progress over time, while large buyers may leverage harmonized reporting from suppliers to foster alignment and accelerate coordination across the ecosystem.

Transferability is high for process-agnostic modules (e.g., energy, logistics), while product- and context-specific modules (e.g., ingredients, packaging formats) require sectoral tailoring. We therefore suggest modular deployment for other agri-food subsectors.

4. CONCLUSIONS

The goal of the study is to show that a GHG accounting tool can support beer companies in the assessment of CE strategies. The tool presented here builds on both the GHG protocol and the ISO 14064-1 standards and is based on 6 subsequent steps which include: i) organizational and operative boundaries definition, ii) identification of databases for the calculations of emission sources and emission factors, iii) calculation of the CF, iv) performing a screen-

ing study and hotspot identification, v) conducting a check with literature to vi) perform a CE strategies assessment.

This made it possible to build a valuable and reliable tool, in line with international standards and easy to replicate in other contexts. The tool enables companies to identify environmentally relevant processes (hotspot analysis) and to implement relevant circular ecosystem innovation. This is possible thanks to its ability to create collaboration within ecosystem actors and allow a trial-and-error process.

Possible limitations could arise due to data collection effort, necessary to fill the input data spreadsheet, and manual data entry, which may introduce inconsistencies. Most of the time, companies have neither the time nor the knowledge to fulfil these tasks. Another limit could be the lack of automated adjustment of emission factors in case of any assessment method updating.

Future developments could address this limitation by providing training courses to companies' data owners on CF accounting and CE principles. The design of a more user-friendly web-based version with automated data ingestion (Application Programming Interfaces (APIs) to utilities, Enterprise Resource Planning (ERPs), and metering systems) can be a solution to address both company involvement raising and dynamic EF updating, thanks to the adoption of another simplification strategy following the automation logic (Beemsterboer et al., 2020).

Finally, the development of the 7-step methodological framework provides a basis for future development and adaptation of the assessment process to other agri-food subsectors to test transferability and benchmarking.

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CIRCULARITY OF TIMBER CONSTRUCTION ACCORDING TO THE LIFE-CYCLE POTENTIAL OF WOOD BY-PRODUCTS

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ABSTRACT

This scoping review examines the integration of life-cycle assessment (LCA) and circular economy principles - specifically practices such as recycling, reuse, reclamation, and upcycling - within wood-based architectural design, building, and construction industry. The aim is to provide a detailed evaluation of the environmental impacts associated with wood and timber materials in the context of sustainable building practices. By analyzing recent studies, this review focuses on how these materials perform environmentally across various life-cycle stages (A1: raw material supply, A3: manufacturing, C1: deconstruction, and C3: waste processing). Each selected study's materials were assessed mainly for Global Warming potential (GWP) and, where data was available, for other environmental impact indicators based on established LCA frameworks. In addition to environmental assessments, the review documents relevant metadata, including material origin, year of publication, and study sources, to construct a comprehensive dataset. The findings underscore that reclaimed and recycled wood materials offer significant potential for reducing greenhouse gas emissions, aligning with the goals of a circular economy by minimizing resource extraction and waste generation. However, several challenges impede widespread adoption. These include concerns regarding the durability and long-term performance of recycled or reclaimed wood, the economic feasibility of implementing circular practices at scale, and regulatory or policy barriers that can limit the reuse and upcycling of wood materials. Despite these challenges, the study emphasizes the untapped potential of upcycled wood products and the need for further research into their environmental benefits and structural performance in architectural applications. This review provides critical insights for advancing a sustainable and resource-efficient construction industry, laying the groundwork for future investigations into innovative wood material practices within the framework of a circular economy.

1. INTRODUCTION

The construction industry, one of the largest contributors to global carbon emissions and resource consumption, faces mounting pressure to adopt sustainable practices to mitigate environmental impacts. A growing body of research emphasizes the need for sustainable materials and building methods, especially through the integration of renewable resources such as wood and timber (FAO, 2018; EC, 2019; Gresham, 2020; Galluccio et al. 2024). Wood, as a renewable and carbon-sequestering material, has gained significant interest in sustainable architecture, particularly when combined with circular economy principles like recycling, reuse, reclamation, and upcycling (Beyer et al., 2011; Braungart & McDonough, 2013; Campbell et al. 2020). These approaches aim to extend material life cycles, reduce waste, and lower emissions throughout a building's

life span. IPCC (2022) highlights that the building and construction sector accounts for a significant share of global greenhouse gas emissions, especially through energy use and material production.

UNEP (2020) reports that buildings and construction are responsible for nearly 40% of global energy-related carbon dioxide emissions and resource consumption, underscoring the urgency for sustainable practices.

EN 15804 provides the framework for Environmental Product Declarations (EPDs) in construction, defining the different stages of a product's life cycle (A1-A3 for product stage, B1-B7 for use stage, C1-C4 for end-of-life stage, and D for benefits beyond system boundary). However, this work focuses on A1 (raw material supply), A3 (manufacturing), C1 (deconstruction), and C3 (waste processing) as these stages are often emphasized because they repre-

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sent the most significant contributions to global warming potential (GWP) and other environmental impacts for wood-based materials, while also aligning with current literature on timber life-cycle assessments. Since Life-cycle assessment (LCA) has become a critical methodology for evaluating the environmental impacts of wood and timber across their full life cycle, from raw material extraction to disposal or reuse (El-Haggar, 2007; Hauschild et al., 2018; Yu & Fingrut, 2022), this review focuses on assessing recent applications of LCA in wood-based architectural and construction projects to better understand the global warming potential (GWP) and environmental impacts of wood materials across specific life-cycle stages, including A1 (raw material supply), A3 (manufacturing), C1 (deconstruction), and C3 (waste processing). Furthermore, the review compiles data on various wood materials identified in relevant literature, emphasizing GWP, life-cycle stage impacts, location, year, and source, thereby providing a robust database framework. GWP is an index that quantifies the relative climate impact of different greenhouse gases over a specified time horizon (typically 100 years), expressed in terms of carbon dioxide equivalents (CO₂e). This makes it a widely adopted and comparable metric across LCA studies. Justifying the focus on GWP would enhance methodological transparency, as it highlights that GWP captures the most critical environmental impact category linked to construction materials, while also acknowledging that other impact categories (e.g., acidification, eutrophication, resource depletion) could provide complementary insights.

Specifically, this review contributes critical research on environmental data essential for building a first “prototype” of a comprehensive database to support LCA-type evaluations within digital design and manufacturing processes. By emphasizing innovative and circular materials, this research aims to support sustainable digital design practices that leverage LCA insights to guide material selection and resource-efficient construction. As such, this review attempts at forming the foundation for future environmental databases in digital design, thereby advancing circular design practices in architecture and construction. We also discuss ongoing challenges to implement these practices, such as economic viability, material durability, and regulatory compliance, and identify research needs for optimizing wood’s role in sustainable architecture. Specifically, this review contributes critical research on environmental data essential for building a first “prototype” of a comprehensive database to support LCA-type evaluations within digital design and manufacturing processes. In this context, “digital design” refers to emerging practices such as BIM-based modeling, parametric workflows, and fabrication-oriented approaches, which increasingly shape architectural production both technically and culturally (Perriccioli & Ruggiero, 2024). By emphasizing innovative and circular materials, this research aims to support sustainable digital design practices that leverage LCA insights to guide material selection and resource-efficient construction.

This paper is organized in 5 paragraphs: “Introduction”, “Research and Methodology”, “Results”, “Discussion”, “Conclusion”. In “Research and Methodology”, we conduct a systematic literature review, analyzing studies on LCA

and circular wood materials from 2010 onward. Key environmental metrics, such as GWP and life-cycle stages, are extracted to compare the impact of reclaimed, recycled, and new wood. A database is developed to support sustainable material selection in digital design and manufacturing. In “Results”, we highlight that wood-based materials have a lower environmental impact than concrete and steel, with circular strategies (reuse, recycling, upcycling) reducing emissions. However, material durability, recyclability issues, and regulatory barriers limit adoption. In “Discussion”, we analyze LCA data across life-cycle stages, highlighting the benefits of circular practices and carbon sequestration. Challenges include regional processing differences and logistical constraints, requiring regulatory support and standardized LCA methodologies. In “Conclusions” we emphasize that circular economy practices in wood construction can significantly lower emissions. Future efforts should focus on LCA standardization, digital databases, and optimized reuse strategies to advance sustainable, low-carbon construction.

2. RESEARCH METHODOLOGY

2.1 Literature search and selection criteria

The initial step involves a systematic search of peer-reviewed journals, conference proceedings, and industry reports related to the keywords: “life-cycle assessment” (LCA), “recycle,” “reuse,” “reclaim,” “upcycle,” “architectural design,” “building,” “construction,” “wood,” and “timber.” Key academic databases, including Web of Science, Scopus, Google Scholar, and ScienceDirect, are searched to ensure a comprehensive selection. The search focuses on studies published from 2010 onward to capture recent advancements in LCA and circular material practices, while priority is given to publications from regions where sustainable construction is highly regulated, such as Europe, North America, and parts of Asia (EC, 2003; ISO, 2006a;b; Rigillo et al., 2023) (Figures 1-3). Key academic databases, including Scopus, were searched to ensure a comprehensive selection. The review focuses on studies published from 2010 onward, a starting point chosen to capture the period following the introduction of influential policy and conceptual frameworks such as the EU Roadmap to a Resource Efficient Europe (EC, 2011) and the wider diffusion of circular economy paradigms through the Ellen MacArthur Foundation (2012) (Figure 4). These milestones mark a shift toward sustainability-driven construction research and provide a strong rationale for this temporal boundary (Figure 5). Priority is given to publications from regions where sustainable construction is highly regulated, such as Europe.

2.2 Data collection

From each selected study, specific data points are extracted to evaluate the environmental impact of wood-based materials across different life-cycle stages, as follows:

1. Global Warming Potential (GWP) - Among the indicators used in LCA, global warming potential (GWP) is particularly critical because it quantifies greenhouse

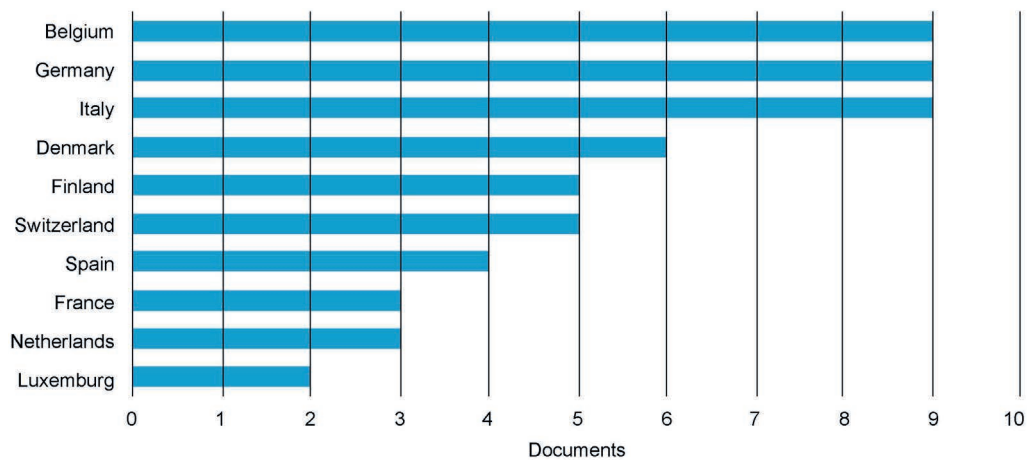


FIGURE 1: Data on the number of documents categorized by countries or territories.

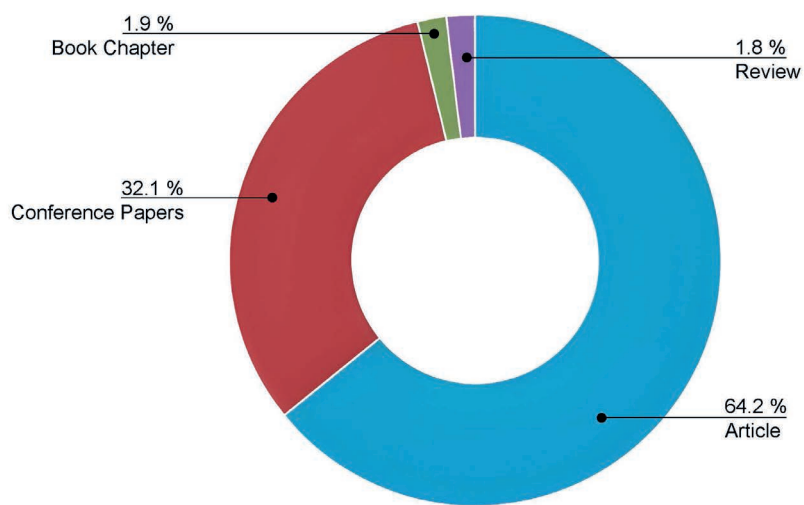


FIGURE 2: Data on the percentage distribution of documents by type.

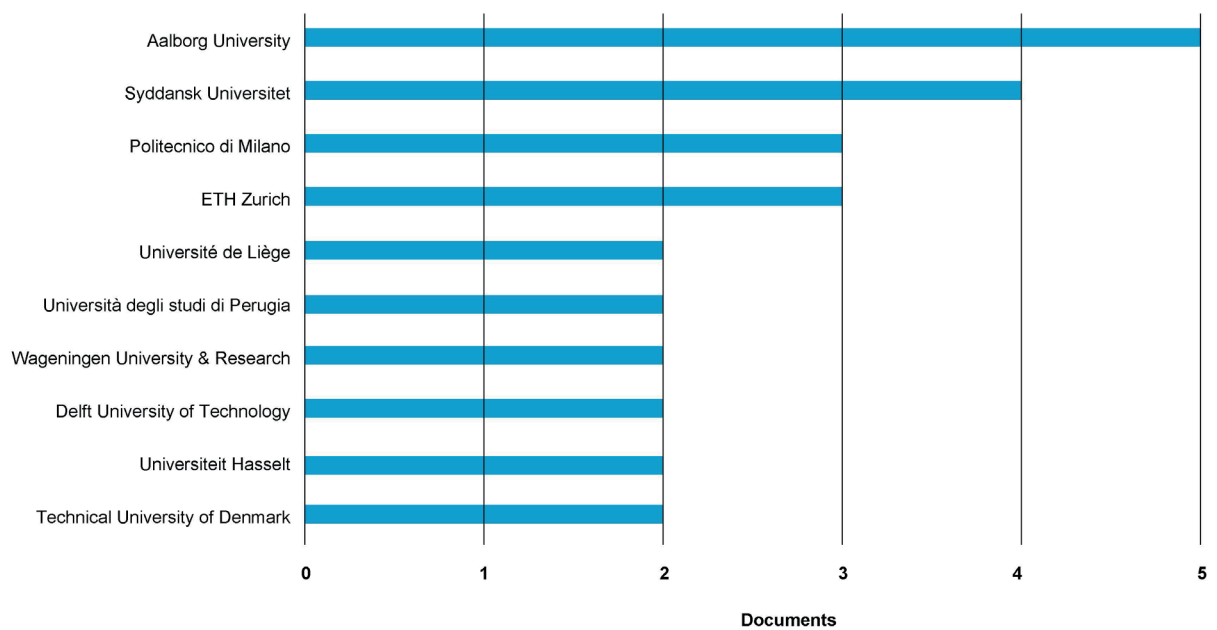


FIGURE 3: Data on the percentage of documents categorized by affiliation.

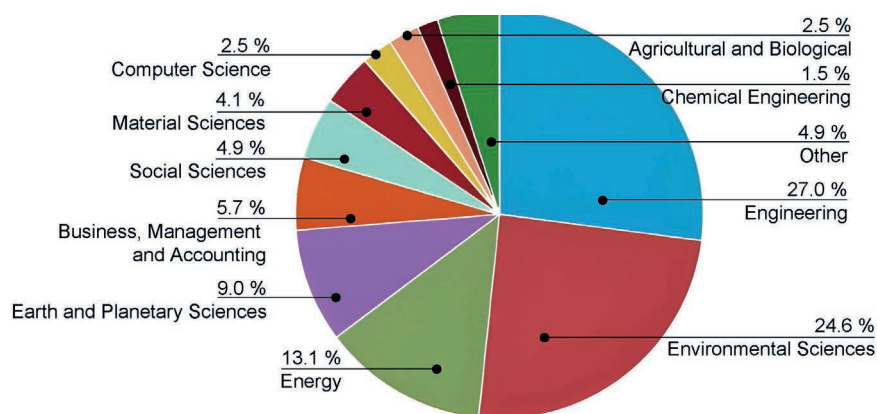


FIGURE 4: Data depicting the percentage distribution of documents across various subject areas.

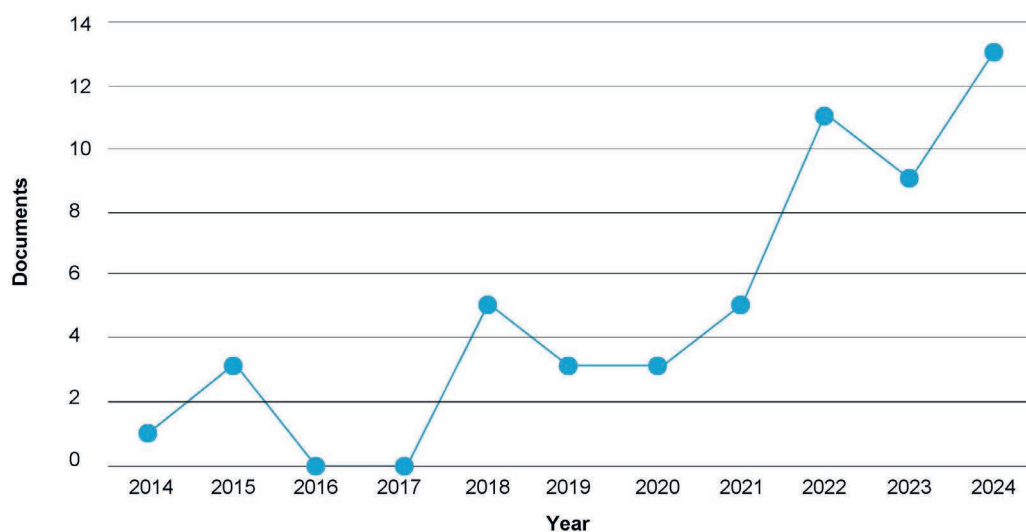


FIGURE 5: Number of documents (Papers) produced annually from 2014 to 2024.

gas emissions across different life-cycle stages in terms of their contribution to climate change. Beyond serving as an environmental indicator, GWP has direct design implications: it enables architects and engineers to compare the carbon intensity of alternative materials (e.g., timber versus concrete), evaluate the impact of design strategies such as modularity, prefabrication, or reuse, and align construction practices with broader climate targets and net-zero pathways (IPCC, 2022; UNEP, 2020). In this way, GWP functions as both a scientific measure and a decision-making tool that informs sustainable material selection and resource-efficient architectural design. A key metric that quantifies the greenhouse gas emissions associated with the material over its life cycle, measure in kg of CO₂ equivalent;

2. Life-Cycle Stages - Impacts were evaluated specifically at stages A1 (raw material supply), A3 (manufacturing), C1 (deconstruction), and C3 (waste processing) to assess the material's environmental footprint;
3. Material Source, Year, and Location - Information on the origin of each material (region or country), year of study, and publication source was documented to allow for comparisons across different contexts.

This data is organized into a structured datasheet, allowing for easy reference and future use in creating databases for LCA-type evaluations within digital design and manufacturing processes. The goal is to define a foundation that could support the growing demand for open LCA-driven tools in digital construction, facilitating the selection of materials with lower environmental impacts (Augusti-Juan et al., 2016;2017; EC, 2021; Ke Yin Lee et al., 2023; Galluccio et al., 2025).

2.3 Evaluation criteria and analysis

The extracted data are analyzed using a comparative approach, focusing on the environmental benefits and drawbacks of circular practices with wood materials in construction. Comparative analyses are conducted across GWP values and life-cycle impacts for different wood materials (e.g., reclaimed versus virgin wood) to understand how recycled and reused wood fares against new timber in sustainable design contexts. Each study is also evaluated according to its methodology to assess the reliability of reported LCA values, considering factors such as the inclusion of end-of-life stages, completeness of system boundaries, and geographic relevance. Further, each study's methodol-

ogy is scrutinized to identify common gaps, such as the lack of comprehensive end-of-life data for reclaimed wood, that could inform future LCA research. The methodological workflow could be summarized in the following steps:

1. Literature Review. A systematic search was conducted across major academic databases, including Scopus, Web of Science, Google Scholar, and ScienceDirect. The search employed targeted keywords such as “life-cycle assessment,” “wood,” “timber,” “reuse,” “recycle,” and “upcycle.” Inclusion criteria required publications from 2010 onward, peer-reviewed status, and direct relevance to life-cycle assessment (LCA) and circular wood applications. Studies not meeting these criteria were excluded.
2. Data Extraction - Selected studies were analyzed to extract key variables, including:
 - Material types (e.g., virgin wood, reclaimed wood, engineered timber);
 - Life-cycle stages addressed (A1, A3, C1, C3);
 - Reported environmental impacts (e.g., global warming potential [GWP], other impact categories);
 - Geographic origin and publication year;
 - Methodological parameters (e.g., system boundaries, end-of-life scenarios, underlying assumptions).
3. Database Construction. Extracted data were organized into a structured comparative framework. Units of measurement, impact categories, and life-cycle modules were standardized to ensure consistency and facilitate cross-study comparison.
4. Comparative Evaluation and Synthesis. The compiled dataset was used to:
 - Compare GWP values across different wood material types (e.g., virgin vs. reclaimed);
 - Evaluate the methodological rigor of each study, focusing on system boundaries, end-of-life modeling, and regional specificity;
 - Identify recurring patterns, inconsistencies, and gaps in reported outcomes.

This synthesis enabled the identification of environmental advantages and limitations associated with circular wood practices. It also revealed common methodological shortcomings — such as incomplete end-of-life data — and informed recommendations for future research, including the development of sector-specific LCA guidelines and improved integration of circularity metrics in sustainable wood construction.

2.4 Database development for digital design and manufacturing

The synthesized data will be instrumental in developing a database to support digital design and manufacturing processes with circular wood materials. This database will enable architects and designers to evaluate materials based on LCA metrics, facilitating sustainable material selection. The data compilation also considers scalability for use in digital tools like Building Information Modeling (BIM) software, which integrates LCA data for holistic environmental assessments during the design phase (Cays,

2017). The synthesized data will be instrumental in developing a prototype database to support digital design and manufacturing processes with circular wood materials.

The database is structured as a relational framework, organized around key parameters including material type (virgin, reclaimed, engineered timber), life-cycle stages covered (A1, A3, C1, C3), reported GWP values and other LCA indicators, geographic location, year of study, and methodological characteristics such as system boundaries and treatment of end-of-life scenarios. To ensure comparability, data were standardized in terms of units, functional units, and impact categories before being organized into the database.

While currently developed as a conceptual prototype, the structure has been designed with scalability in mind, allowing future integration into digital tools such as Building Information Modeling (BIM) software. This integration would enable architects and designers to query material options based on environmental metrics, facilitating evidence-based material selection and more holistic environmental assessments during the design phase (Cays, 2017).

3. RESULTS

This data highlights the significant environmental benefits and challenges of integrating various wood materials into sustainable construction practices. Many wood-based materials, like Cross-Laminated Timber (CLT) and Glulam, exhibit lower global warming potential (GWP) compared to conventional materials like concrete and steel, with some even demonstrating carbon sequestration capabilities. Reclaimed, recycled, and upcycled wood products show strong potential for reducing emissions and supporting circular economy principles through reuse and energy recovery. However, factors such as limited recyclability (e.g., due to adhesives), high emissions in certain life-cycle stages, and regulatory or logistical barriers can complicate widespread adoption. Despite these challenges, the data underscores the opportunity to optimize wood use across life-cycle stages, driving a more sustainable, resource-efficient construction industry while reducing overall carbon footprints. The study finds that integrating life-cycle assessment (LCA) with circular economy principles in wood-based architectural design and construction offers significant environmental benefits.

Key results include:

1. Global Warming Potential (GWP) Reduction. Reclaimed and recycled wood significantly reduces greenhouse gas emissions compared to virgin timber, especially in life-cycle stages A1 (raw material supply) and A3 (manufacturing);
2. Enhanced Sustainability Metrics. Circular economy practices, such as reuse, recycling, and upcycling, extend material life cycles, decreasing overall environmental footprints in construction;
3. Challenges Identified. Issues such as material durability, regional processing differences, and regulatory barriers persist, limiting the widespread adoption of circular practices.

3.1 Evidence from Reviewed Literature

The reviewed studies highlight both the environmental benefits and challenges of integrating various wood materials into sustainable construction practices. Materials such as Cross-Laminated Timber (CLT) and Glulam consistently demonstrate lower global warming potential (GWP) compared to conventional construction materials like concrete and steel, with some studies even documenting carbon sequestration capacities. Literature on reclaimed, recycled, and upcycled wood products further underscores their potential to reduce emissions while supporting circular economy principles through reuse and energy recovery. A concentration of studies in 2023 emerges from the analysis and could be interpreted as a signal of a rising trend of research on the topics of LCA and forest-based materials in architecture and design. Since international, European and even local regulations are pushing industries to adopt environmental impact metrics to assess their processes and products and a significant emphasis is being placed on the widespread use of bio-based renewable resources such as timber, it is expectable that the number of studies will even grow in the future.

3.2 Dataset Composition and Structure

The dataset (Table 1) compiled from the reviewed sources incorporates variables including material type (virgin, reclaimed, engineered timber), life-cycle stages analyzed (A1, A3, C1, C3), reported GWP values, geographic location, year of publication, and methodological features such as system boundary definitions and end-of-life treatments. This structure enables systematic comparison across case studies and facilitates the evaluation of both environmental performance and methodological robustness.

3.3 Environmental Performance Comparison

Comparative analysis reveals that reclaimed and recycled wood significantly reduce greenhouse gas emissions compared to virgin timber, particularly in the A1 (raw material supply) and A3 (manufacturing) stages. Circular practices, including reuse, recycling, and upcycling, extend material lifecycles and reduce the overall environmental footprint of construction projects. Engineered timber products (e.g., CLT, Glulam) consistently show lower GWP than conventional alternatives, positioning them as effective strategies for lowering emissions in sustainable design.

3.4 Critical Factors Limiting Circular Adoption

Despite these environmental benefits, several challenges persist. Issues such as limited recyclability due to adhesives, high emissions in specific life-cycle stages, and regional differences in wood processing methods introduce variability into results. Moreover, regulatory and logistical barriers restrict the widespread adoption of circular wood practices. These recurring limitations highlight the need for standardized LCA methodologies and improved end-of-life data for reclaimed wood to advance the integration of circular design in construction.

4. DISCUSSION

The findings of this review emphasize the pivotal role of integrating life-cycle assessment (LCA) and circular economy principles in advancing sustainable wood-based architectural design and construction. The data collected from recent studies reveal substantial environmental benefits associated with wood materials, particularly in their capacity to reduce global warming potential (GWP) compared to conventional materials such as concrete and steel. Materials like Cross-Laminated Timber (CLT), Mass Timber, and Glulam not only demonstrate significantly lower GWP values but also possess carbon sequestration capabilities, contributing to net-negative carbon impacts in certain scenarios (Abdolmaleki et al., 2025).

Key insights from this review underscore the transformative potential of integrating life-cycle assessment (LCA) and circular economy principles into wood-based architectural design, revealing significant environmental advantages over conventional materials. Engineered wood products such as CLT, Mass Timber, and Glulam not only exhibit lower global warming potential (GWP) but also contribute to carbon sequestration, offering net-negative carbon outcomes in certain applications. However, the path to widespread adoption is impeded by technical, economic, and regulatory challenges. Adhesive use, regional processing disparities, and high costs associated with reclaiming wood limit recyclability and scalability. Moreover, the absence of standardized certification and quality assurance frameworks hinders market development for recycled wood. Despite these barriers, circular strategies — particularly reuse and upcycling — demonstrate substantial emission reductions, with some materials achieving negative GWP values. The findings highlight the urgent need for harmonized LCA methodologies, improved end-of-life data, and supportive policy reforms to unlock the full potential of circular wood practices in sustainable construction. In the following four sub-paragraphs the insights and learnings from the review are examined.

4.1 Critical Factors Limiting Circular Adoption

Despite these environmental benefits, several challenges persist. Issues such as limited recyclability due to adhesives, high emissions in specific life-cycle stages, and regional differences in wood processing methods introduce variability into results. Moreover, regulatory and logistical barriers restrict the widespread adoption of circular wood practices.

To provide a more complete picture of these challenges, it is essential to acknowledge the significant economic and market barriers that hinder the scalability of circular wood practices. For instance, the costs associated with processing, sorting, cleaning, and re-engineering reclaimed wood often make it less competitive than new, virgin materials. The difficulty of establishing a stable, structured market for recycled wood is another major hurdle, exacerbated by the lack of standardized certification and quality assurance protocols that are necessary for its reliable integration into large-scale construction projects. These recurring limitations highlight the need for standardized LCA

TABLE 1: Summary table of all the wood materials mentioned in the papers and their Impact Global Warming Potential, A-1, A-3, C-1, C-3, Location, year and source. Notes: "Included" = stage explicitly modeled; "Limited" = partial coverage; "Not reported" = no data available; "Reuse potential" refers to end-of-life recovery options.

Wood product or by-product	A1 Raw Material Supply	A3 Mnft.	C1 Dem./Decn.	C3 Waste Processes	GWP Impact (A1+A3+C1+C3)	Location	Year	Source
Cross-Laminated Timber (CLT)	Included	Included	Energy recovery considered	Some reuse potential	0.314kg CO ₂ e/kg	Europe, Denmark, USA, Japan	2021-2024	Discover Applied Sciences, IOP Conf. Series
Oriented Strand Board (OSB)	Limited reuse potential	Moderate emission	High reuse potential if not incinerated	Inciner. possible	Between 600 and 680 CO ₂ e/kg per m ³ .	Europe, Global	2023-2024	Discover Applied Sciences, Sugahara et al.
Plywood (PaB)	Adhesive limit to recycl.	Moderate emission	Inciner. provides energy offsets	Inciner. modeled	Approximately 1.07 kg CO ₂ e per kg, lower GWP impact through pulpwood substitution	Europe	2024	Discover Applied Sciences
Light Wood Framing	Included	Included	High impact if not reused	Recycling debated	0.46 kg CO ₂ e per kg, Varies based on end-of-life scenario	Southern USA	2020-2024	Passarelli, IOP Conf. Series
Engineered Wood (LVL, Glulam, OSB)	Included	Included	High impact if landfilled	Recycling & reuse exist	338 kg CO ₂ e per cubic meter, Positive CO ₂ sequestration but supply chain emissions	Europe, USA	2024	IOP Conf. Series
Mass Timber	Included	Included	High reuse potential	2.5x lower than concrete	582 kg CO ₂ per cubic meter	Multiple locations	2023	Kanyilmaz et al.
Timber-Concrete Composite (TCC) Floors	30-50% lower emissions than RC	Low primary energy demand	Higher environ. benefits in reuse	Full reuse recomm.	0.159 kg CO ₂ e per kilogram, One-third of reinforced concrete (RC) emissions	Europe	2023	Research Journal
Deconstructed Timber Buildings	Carbon sequestr.	Embodi. carbon storage offsets emissions	Can be reintrod. into cycle	Circular approach ensures negative CO ₂ balance	General Timber (excluding sequestration): 0.46 kg CO ₂ e per kg Glue-Laminated Timber: Approximately 0.87 kg CO ₂ e per kg	Europe	2023	Research Journal
Kraft Lignin	Biogenic carbon storage	Moder. emiss.	Reuse in bio-based applic.	Inciner. or reuse possible	Not reported	Latvia	2024	Constr. and Building Materials
Wood Fly Ash (WFA)	Utilized in asphalt filler	Moder. emiss.	No reuse	Reduces cement demand	Not reported	Latvia	2024	Constr. and Building Materials
Softwood (38x200mm)	Included	Included	Not explicitly stated	Not explicitly stated	0.46 kg CO ₂ e per kg	Belgium	2022	E3S Web of Conf.
CLT Timber Structure	Included	Included	Not specified	Not specified	0.43 kg CO ₂ e/kg	Belgium	2022	Sustainab. Journal
Pine Timber (Structure)	Included	Included	Can be dismantled and reused	Reuse or inciner.	-382,101 kg CO ₂ stored	Belgium	2021	Obaidy et al.
Timber Flooring	Included	Included	High reuse potential	Reuse or inciner.	73.5% of total structure weight	Belgium	2021	Obaidy et al.
Timber-Based Façade (Staples)	Included	Included	98% longer duration than best option	75% reusability, rest disposed	Highest among options due to waste	Italy	2022	Juaristi et al.
Timber-Based Façade (Screws)	Included	Included	35% longer duration than best option	100% reusability	Lower than staples, easier disassembly	Italy	2022	Juaristi et al.
Timber-Based Façade (Geometrical Assembly)	Included	Included	Best option, fastest disassembly	100% reusability	Very low GWP, best for reusability	Italy	2022	Juaristi et al.
Glulam (Glued Laminated Timber)	Included	Included	Included	Included	-656 kg CO ₂ e with biogenic uptake	Sweden	2016	Buildings Journal
Structural Timber (Glulam Beams)	Included	Included	Reused with minor processing	Energy recovery by incineration	30% lower impact than virgin production	Finland	2021	Niu et al.
Recycled Wood Fiber (CDW)	Included	Included	Moderate reuse potential	Incineration or recycling	-1.79 to -2.25 kg CO ₂ e (avoided impact)	Finland	2021	Sormunen et al.
Wood Waste Panel	Included	Included	Included	Included	-656 kg CO ₂ e (Glulam with biogenic carbon uptake)	Sweden	2021	Sustainab.
Cross-Laminated Timber	Included	Included	0.16 kg CO ₂ e/kg	Incineration or reuse	-1.78 kg CO ₂ e/kg	Belgium	2022	Binderholz EPD

methodologies and improved end-of-life data for reclaimed wood to advance the integration of circular design in construction.

4.2 Environmental performance across life-cycle stages

The LCA analysis of wood materials across life-cycle stages (A1: raw material supply, A3: manufacturing, C1: demolition, and C3: waste processing) illustrates distinct environmental trade-offs. While raw material supply and manufacturing processes can generate emissions, these are often offset by carbon sequestration during growth and energy recovery at the end of life. For example, Timber-Concrete Composite (TCC) floors exhibit a GWP impact of only 0.159 kg CO₂eq./kg, significantly lower than reinforced concrete, while reclaimed and recycled materials further extend the environmental benefits through waste reduction and reuse potential (Ivanica et al., 2022).

4.3 Potential for circular economy practices

Circular practices such as reuse, recycling, reclamation, and upcycling emerge as crucial strategies for enhancing the sustainability of wood-based materials. The data show that reclaimed timber and recycled wood fiber can drastically reduce emissions, with some materials even achieving negative GWP values. For instance, recycled wood fiber can avoid emissions ranging from -1.79 to -2.25 kg CO₂eq., illustrating the environmental savings possible through circularity (Abdolmaleki et al., 2025).

However, the feasibility of circular practices depends on several factors. Adhesives and treatments, commonly used in plywood and engineered wood, can limit recyclability, while the logistics of deconstruction and transportation may add emissions and costs. Yet, when circular economy principles are properly implemented, materials such as deconstructed timber buildings and geometrically assembled timber facades can achieve nearly 100% reusability, promoting material longevity and reducing waste (Xie et al., 2024).

4.4 Challenges and barriers to implementation

Despite the promising potential of wood materials in sustainable construction, several challenges hinder widespread adoption. Material durability and long-term performance are persistent concerns, especially for reclaimed wood. Additionally, regional variations in wood processing techniques and supply chain emissions introduce variability in environmental impacts, complicating the standardization of LCA practices. Furthermore, regulatory and policy limitations in some regions may restrict the reuse and upcycling of wood materials, emphasizing the need for updated legislation that encourages circular design. Beyond these issues, the economic feasibility and market maturity of circular practices present significant barriers. The high costs and labor involved in deconstruction, as well as the need for specialized equipment to process, sort, and clean reclaimed wood, often make these practices economically unviable when compared to using new timber. Additionally, the construction industry lacks a stable market with structured demand for recycled wood, which is further compli-

cated by the absence of widely accepted certification and quality assurance standards. Without these systems, architects and engineers face difficulties in verifying the quality and reliability of reclaimed materials, limiting their use to smaller, non-structural, or niche applications.

5. CONCLUSIONS

The study concludes that adopting circular economic practices in wood-based construction can significantly reduce the environmental impact of the industry. Reclaimed and recycled wood demonstrate measurable benefits in reducing GWP and resource consumption. However, economic, regulatory, and methodological challenges must be addressed to fully realize their potential.

Future efforts should focus on standardizing LCA methodologies and expanding digital databases to support sustainable design and construction practices. However, challenges remain in fully adopting circular economy practices at scale, including issues related to material durability, inconsistent regional processing methods, and regulatory barriers that hinder widespread reuse and upcycling. The findings emphasize the need for continued research into the environmental benefits and structural performance of upcycled wood products, as well as the development of standardized LCA frameworks to better assess and compare materials across life-cycle stages. The creation of comprehensive LCA databases will be crucial in supporting sustainable, resource-efficient construction practices, enabling designers to make informed material choices that align with circular economy goals. In conclusion, while barriers to implementation persist, the ongoing integration of LCA and circular economy principles offers a transformative path forward for the construction industry, fostering more sustainable building practices and contributing to the global effort to reduce carbon emissions. Future research and technological advancements will be pivotal in overcoming current limitations, optimizing the use of wood in architecture, and advancing the transition to a circular, low-carbon construction sector.

Several limitations were encountered, including data variability due to regional differences in wood processing methods, and a limited number of studies covering comprehensive LCA stages for reclaimed wood. Future research is recommended to close these gaps, with emphasis on creating standardized LCA practices for circular wood materials in construction. This review also highlights the need for advanced LCA databases capable of supporting digital and circular design processes to optimize wood's role in sustainable architecture and construction.

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EMPOWERING FOUNDRIES FOR SUSTAINABILITY: A USER-FRIENDLY ECO-DESIGN TOOL FOR ENVIRONMENTAL SELF-ASSESSMENT

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ABSTRACT

Given the role of aluminum as a strategic resource for several decarbonization strategies, its global demand has experienced an exponential growth. However, aluminum production remains highly energy-intensive and environmentally impactful. Our objective is therefore to support aluminum foundries in addressing the challenge of reducing their environmental burden. For this purpose, this study introduces a software (named Eco-design tool) developed for Italian small and medium aluminum foundries. The tool enables self-assessment of environmental performance based on the Life Cycle Assessment (LCA) methodology, which evaluates the impacts of the foundry process – including emissions, energy use and waste management. Through a user-friendly interface, the tool collects site-specific data from seven production phases to calculate key environmental indicators. The main output is a preliminary environmental profile that helps companies identify critical impact areas and supports informed decision-making, strategic planning and continuous improvement, helping companies align with circular economy principles and reduce environmental impacts. The Eco-design tool is particularly valuable for small and medium-sized companies lacking the resources for full-scale LCA studies and can guide them toward environmental certifications. Future work will focus on expanding the dataset, improving usability, and validating the tool across a broader sample of foundries.

1. INTRODUCTION

In recent years, sustainability has become a crucial objective for companies aiming to maintain their competitiveness. Industries face constant pressure to enhance the environmental performance of their products and manufacturing processes by reducing waste production and preventing pollutant emissions (Appolloni et al., 2022). Aluminum production is a strategic asset to boost environmental sustainability in several industrial sectors, such as the automotive one (Pauliuk, 2018). In fact, as a major metal, it enables transportation, communication, housing, and an infinite number of products and services (Elshkaki et al., 2018). However, this industry is encountering multiple challenges due to intensifying global competition, strict environmental regulations, emissions, and waste issues, all of which affect sustainability performance (Madan and Singh, 2023). Among these challenges, the high demand of bauxite, classified as a critical raw material by the European Commission (European Commission, 2023), is particularly significant as it is unlikely to be sustainably satisfied by the known reserves (Xiao et al., 2023). Moreover, the production of primary aluminum is hugely energy intensive

compared to other metals, thus affecting the environmental performance of aluminum products. Hence, recycling aluminum becomes strategic to reduce its environmental impacts (Raabe et al., 2022).

Given this context, foundries are under increasing pressure to quantify and to improve their environmental performance, yet often lack the tools and resources to do so effectively.

Life Cycle Assessment (LCA) has emerged as a key methodology for quantifying the environmental impact of products and processes in the aluminum industry. In fact, it is frequently used in scientific literature to quantify the environmental performance of primary and secondary aluminum (Liu and Müller, 2012) and has been widely endorsed by sectoral associations such as the European Aluminium Association (EAA, 2024), the International Aluminium Institute (IAI, 2023), and the Aluminum Association (AA, 2023). LCA offers a comprehensive “cradle-to-gate” or “cradle-to-grave” perspective, allowing foundries to evaluate multiple environmental impact categories, such as Climate Change, Acidification, and Resource depletion. This holistic approach makes LCA particularly suitable for aluminum foundries (Milovanoff et al., 2021), where environmental



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burdens are spread across upstream raw material extraction, energy-intensive production, and downstream waste management (Rossi et al., 2025). Furthermore, the recent adoption of the Product Environmental Footprint (PEF) approach, including category rules for metals (European Commission, 2016; European Aluminium, 2022), further strengthens LCA's relevance by enhancing consistency and comparability across assessments.

Despite its advantages, a major limitation of LCA in the aluminum sector remains the scarcity of site-specific primary data. As noted by several studies (Brogaard et al., 2014; Luthin et al., 2021), LCA models often rely on generic industry databases, which may not reflect regional or national production conditions. This lack of geographical representativeness can lead to misleading conclusions and hinder the development of tailored sustainability strategies. While some authors have conducted LCA based on primary data collected at specific sites – such as Grimaud et al., (2018) who focused their study on an aluminum recycling plant located in France, and of Liu et al., (2021) and Salonitis et al., (2016) who have investigated the energy consumption of aluminum production in two aluminum production and die-casting sites in United Kingdom – most studies still depend on secondary datasets.

Thus, since several LCA studies available in the literature are based on industry-generic databases which are not fully representative of the geographical context in which the production site is located, Rossi et al. (2025) strongly encourage future research to fill this gap and overcome this issue by developing country- or regional- specific databases. To overcome this limitation, recent research efforts - such as the GRINS - “Growing Resilient, INclusive and Sustainable” project funded under the EU NextGenerationEU initiative - have focused on collecting and processing quantitative data from Italian aluminum foundries to build nationally representative datasets (Grins Foundation, 2023).

Another promising solution for improving geographical representativeness of LCA analyses is the development of digital tools and calculators tailored to specific industrial contexts. These platforms allow foundries to enter their own operational data and receive an immediate estimate of their environmental footprint. This is especially relevant for small and medium-sized enterprises (SMEs), which dominate the Italian foundry sector (Assofond, 2025), and often lack the resources – in terms of time, budget, organizational capacity, or knowledge – to perform full LCA studies (Testa et al., 2017; Singh et al., 2021). Moreover, since the last decade, sustainability assessment tools have been designed for multinational or large firms but, typically, they are not suitable for SMEs (Gómez-Garza et al., 2023; Ortiz et al., 2023). While these tools do not replace the expertise required for certified environmental labeling (e.g., EPDs, EU Ecolabel, or Made Green in Italy (MGI)), they do offer a low-cost, accessible entry point into sustainability assessment and strategic decision-making.

Given this background, the specific research gap addressed by this study concerns the lack of practical, site-specific, and user-friendly tools that support Italian aluminum foundries - especially SMEs - in conducting pre-

liminary environmental self-assessments. Thus, the aim of this paper is to fill this gap presenting a novel aluminum Eco-design tool developed within the framework of the “BRIEF - Biorobotics Research and Innovation Engineering Facilities” project, funded by the Italian National Recovery and Resilience Plan (NRRP), Mission 4 Component 2 Investment 3.1 of Italian Ministry of University and Research funded by the European Union – NextGenerationEU, providing a discussion on its possible managerial implications for the Italian aluminum foundries. In particular, this paper is focused on presenting the managerial implications of the tool, that enables companies to estimate their environmental impacts based on primary and site-specific data, offering managers a practical means to align with circular economy goals and anticipate environmental certification requirements.

2. METHODS

The Eco-design tool presented here has been developed in collaboration with the Italian Foundry Industrial Association-Assofond (Assofond, 2025). Such self-assessment tool is a calculator based on LCA that allows aluminum foundries to perform a screening calculation of their environmental impacts. The foundry process is composed of 7 steps, including melting, casting, molding, piece removal, finishing, thermal treatments, quality checks. The development of the calculator has followed 3 stages: 1) Data collection, 2) Impact factors quantification, and 3) Eco-design tool development.

Figure 1 is a flowchart which presents an overview of the methodology, combining a summary of the three stages of Eco-design tool development (shown in the top part of the Figure) with the seven steps of the foundry process (shown in the bottom section). The figure also highlights that the tool's LCA results are calculated and displayed for each step of the process.

Concerning 1) Data collection phase, a checklist has been designed and sent with the support of Assofond to hundreds of foundries located throughout the Italian territory during the GRINS project. The checklist is an excel spreadsheet in which each module corresponds to one of the stages of the foundry process. Every module contains tables where materials and energy flows are listed row-by-row, and the corresponding amounts have to be filled in the columns, together with comments, notes, and transport distances. The materials and energy flows are the following:

- Ingots (primary and secondary);
- Alloy elements;
- Modifying agents;
- Refining agents;
- Fluxing agents;
- Degassing agents (e.g. nitrogen, argon);
- Fuels (e.g. natural gas);
- Waste (slag, dust, refractories);
- Electricity (e.g. from renewable or fossil sources, grid mix);
- Moulds and cores (including sand, resins, and other additives);

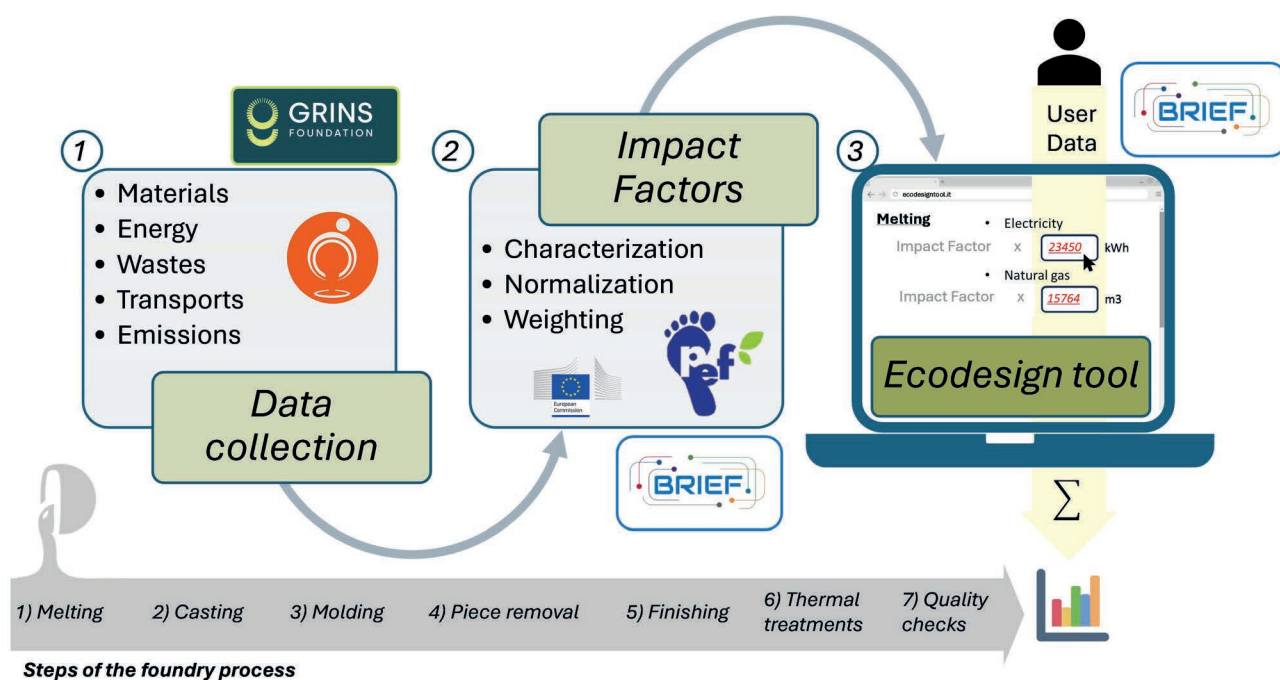


FIGURE 1: Sketch of the methodology followed to define the Eco-design tool for aluminium foundries.

- Water;
- Direct emissions.

The checklist has been filled by 15 foundries with quantitative data and descriptions about the materials consumed over the 7 steps of the process, as well as energy consumption, direct emissions to the atmosphere, transports and waste management (Figure 1). Firstly, 4 volunteer pioneer foundries have tested the effectiveness of the checklist; then, other 11 factories filled the final version. Since energy and material flows vary significantly from foundry to foundry, the checklist has been developed in excel format using a flexible and generic classification of the inputs and outputs of the process. For instance, with the support of Assofond, materials input in the melting furnace have been classified in the classes listed in the above bullet point list. Based on this general classification, foundries could fill the checklist with more specific information: for example, they could add aluminum EN AC-45300 alloy ingots for the class 'ingots', copper for the 'alloy elements', or aluminum-titanium-boron for the 'refining agents'. A similar approach has been used to classify energy sources (e.g. most of foundries declared the use of natural gas, gasoline, and liquefied petroleum gas) and waste recycling and disposal (e.g. most of foundries reported disposing refractory waste and recycling dross and dust).

As illustrated in Figure 1 (more precisely from first box on the left), based on the information collected from the respondent foundries, it has been possible to identify which materials and energy sources are consumed by the foundries and which waste and emissions are produced. In the context of the BREF project, environmental impact factors were associated with these energy and materials flows following the LCA approach (central box of Figure 1). Impact factors calculation is based on the PEF meth-

od: this implies that the use of secondary materials and the recycling of waste are modeled using the Circular Footprint Formula, in accordance with PEF recommendations for parameter setting. Using the software SimaPro version 9.6, the background database ecoinvent 3.9.1 cut-off version (Frischknecht et al. 2005), and the Life Cycle Impact Assessment (LCIA) method Environmental Footprint 3.1 (Bassi et al., 2023), 16 environmental impact indicators are calculated (including characterization, normalization and weighting steps). For instance, in case foundries add copper as alloying element, the corresponding impact factor is calculated as the environmental impacts of 1 kilogram of copper from the ecoinvent dataset "market for copper, cathode". Even though the eco-design tool provides the LCIA results considering all 16 impact categories, it is worth mentioning that the most relevant impact categories in the metals sector are, according to (Santero and Hendry, 2016):

- Climate Change: measures the effects of GHG emissions to Climate Change [kgCO₂ eq];
- Acidification: measures the effects of acidic substances on the environment, particularly on soil and water, which can disrupt ecosystems and harm living organisms [mol H⁺ eq];
- Eutrophication: measures the level of nutrients released into water bodies through human activities, leading to excessive algae growth, oxygen depletion, and ecological imbalances [kg P eq];
- Photochemical ozone formation: measures the emissions of pollutants, such as nitrogen oxides which contribute to the formation of ground-level ozone and smog, impacting human health and the environment [kg NMVOC eq];
- Ozone depletion: evaluates the release of substances, such as chlorofluorocarbons (CFCs), that deplete the

ozone layer in the stratosphere, allowing harmful ultraviolet radiation to reach the Earth's surface [kg CFC11 eq].

In LCIA, the characterization step quantifies the contribution of different emissions and resource uses to environmental impact categories, expressing – for instance - all greenhouse gas emissions as CO₂-equivalents. Normalization contextualizes these impacts by comparing them to a reference, for instance the total environmental load of a person or a region, highlighting their relative significance. Weighting assigns relative importance to different impact categories, enabling their aggregation into a single score to support decision-making (Hauschild et al., 2018).

The developed Eco-design tool is composed of a foreground and a background layer (right box of Figure 1) and it is based on the environmental impact factors associated with the materials and energy flows related to the foundry process that were identified in the context of the GRINS project. The foreground layer is a graphical user interface in which the users can enter their data; the background layer is composed of mathematical formulas – invisible for the user – converting input data into output results.

In the foreground layer, users can enter site-specific values for the foundry under investigation. For instance, in case the annual copper additions amount to '100' kg, the users are required to insert this value in the calculator. In the background layer, the amounts entered by the user are multiplied by the corresponding impact factors. The calculation is performed for each of the steps of the foundry process which are inserted by the user. The impacts of all the relevant steps are then summed up and the final out-

put provided by the Eco-design tool is the environmental burden of the foundry related to 1 kg of aluminium produced, i.e. the functional unit. Following up with the example above, the amount '100' kg of copper is multiplied by the environmental impact of 1 kg of copper resulting from the dataset "market for copper, cathode", i.e. 6.86 kg CO₂ eq / kg copper. A beta-version of the calculator is now online and available for testing by the aluminium foundries associated with Assofond. Seven foundries have provided feedback on the tool: five of them had previously participated in the GRINS data collection campaign, while the other two did not contribute with data in the initial phase. Interestingly, all the foundries that had provided data in GRINS have now considered the tool developed in BRIEF as complete and comprehensive; their main concerns were focused instead on the graphical user interface. In contrast, the 2 foundries that had not contributed with data to the development of the calculator, identified some missing materials and energy flows. The next steps in the development of the tool will involve integrating additional data collected from the testers and expanding the access to the calculator across all foundries affiliated with Assofond. Figure 2 summarizes the geographical distribution of the foundries that contributed to the data collection and the tool validation. These companies are concentrated in the center and north of Italy, thus reflecting the actual distribution of Italian aluminum foundries according to Assofond. In Figure 2, blue labels indicate foundries that participated only in data collection, red labels indicate those involved only in tool validation, and yellow labels indicate participation in both parts of the Eco-design tool development.

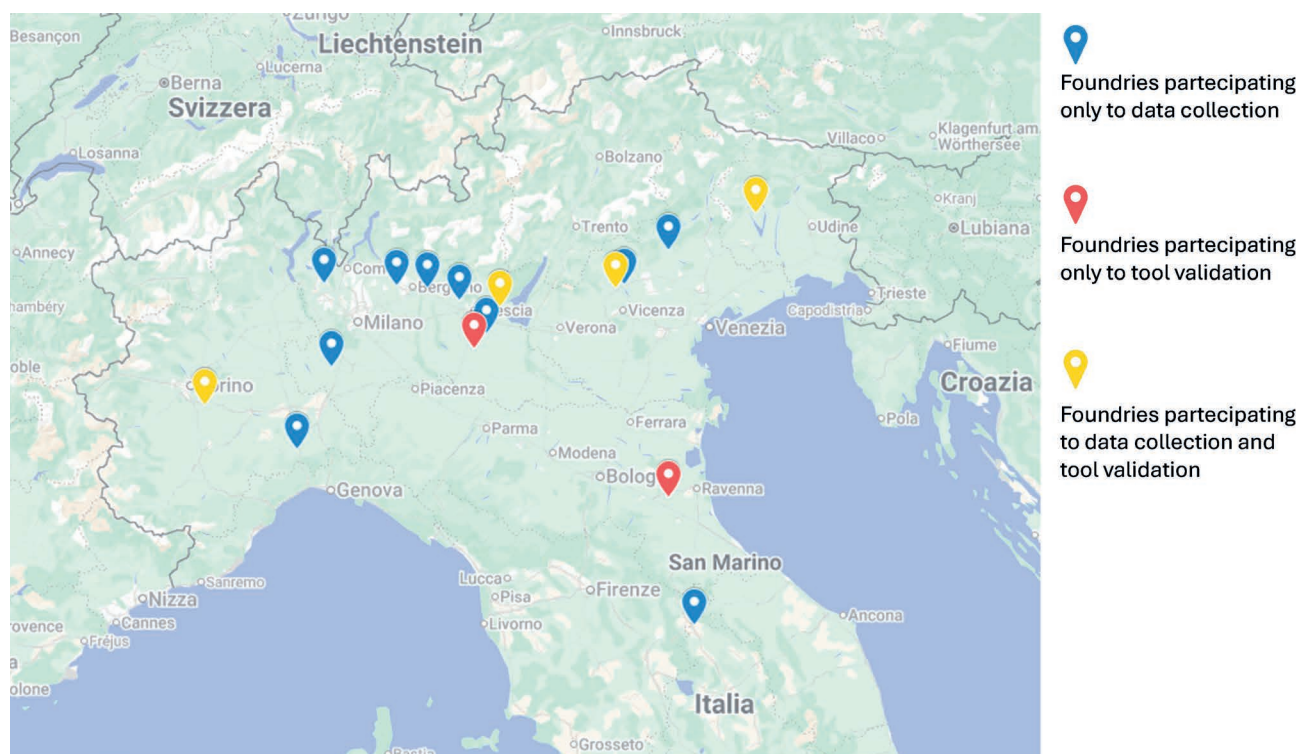


FIGURE 2: Geographical distribution of the foundries participating to data collection and tool validation.

3. RESULTS AND DISCUSSION

The Eco-design tool (developed in Italian) has a modular structure, as illustrated in the dashboard shown in Figure 3. The dashboard becomes accessible after creating a profile on a portal (available via a specific link) and consists of three panels. The left panel allows users to navigate between the modules of the tool, which correspond to different phases of the foundry process. The central panel presents a list of sections where users enter quantitative data or select an option from a dropdown menu (e.g., specifying the means of transport). The right panel contains hyperlinks to all sections within the current module.

Once the whole questionnaire is filled, the user will be able to display the results as numeric tables containing the impact of the process and the contribution of all the 7 steps. Two types of charts are also available: a stacked columns chart (Figure 4 as illustrative example) expressing the contribution of the modules to each midpoint impact category, and a pie chart summarizing the contribution of each impact category to the overall environmental impact (also called single score), calculated through normalization and weighting. The Eco-design tool has been tested by Assofond and 7 pioneer foundries who have proposed changes before the release of the final version of the Eco-design tool. The applicability of the tool has been significantly enhanced through the contributions of these foundries. Five new material types have been added, broadening the scope of analyses. Moreover, based on the feedback received, the tool now allows results to be exported in an editable Excel

format, making it easier to adapt and use the data from a managerial perspective. Additionally, the tool automatically generates a technical report, which managers can leverage to present results to customers or during internal meetings.

Figure 4 presents the results from a foundry that participated in the tool's development. The real-world case study presented here analyzes a process consisting of melting, casting, piece removal, finishing, and quality checks. Figure 4a shows that, for the most relevant impact categories discussed in Section 2, the melting phase is the primary environmental hotspot. Casting and finishing contribute similar levels of environmental impact (depending on the impact category considered), while quality checks are only significant for the indicator Marine eutrophication. Figure 4b highlights that Climate change is the most critical impact category overall. From an eco-design perspective, this indicates that improving resource efficiency in the melting phase offers the greatest potential for reducing the environmental impacts of the entire process. The tool's accuracy was validated by comparing its results with those generated by the professional LCA software SimaPro. The differences were found to be below 1%, confirming the tool's high level of accuracy. However, in order to guarantee high accuracy, high quality data entry from the user is also required.

We have seen that, while LCA is recognized as a robust methodology for evaluating the environmental impacts of industrial products (Monteleone et al., 2024; Nortgate et al., 2007), its practical adoption within the foundry sector - particularly among SMEs - remains limited. Many SMEs

FIGURE 3: Main dashboard of the Eco-design tool.

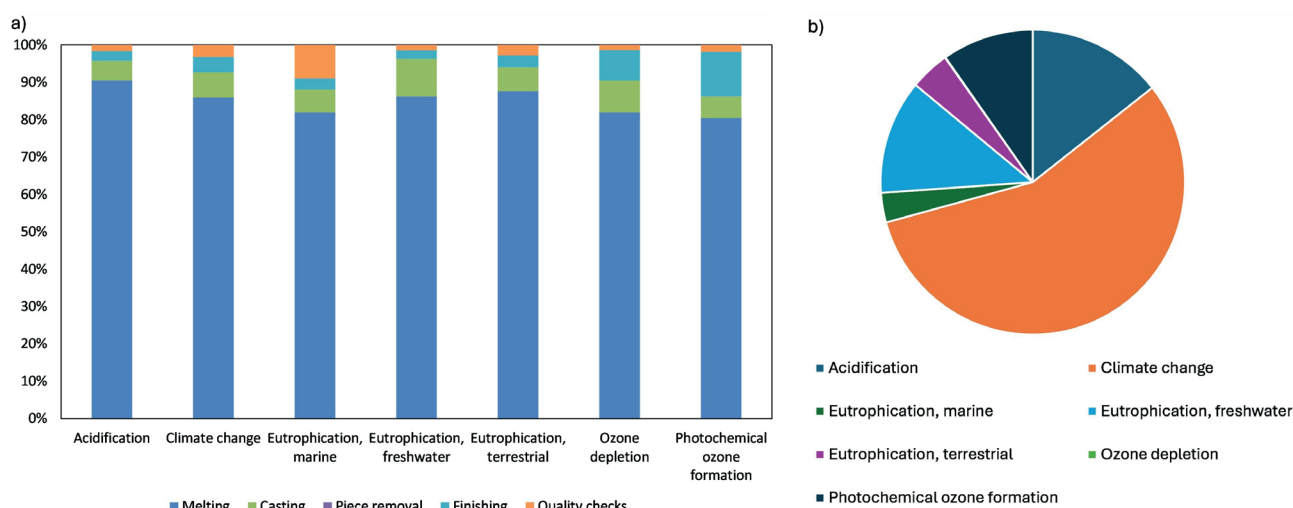


FIGURE 4: Results returned by the Eco-design tool.

face structural constraints such as a lack of trained personnel, insufficient access to specialized databases, and high costs associated with full LCA studies (Gómez-Garza et al., 2023; Ortiz et al., 2023). Nonetheless, these companies are increasingly asked by stakeholders - including clients and regulatory bodies - to disclose environmental performance data (Singh et al., 2021). Therefore, being able to monitor and improve their environmental footprint has become a strategic priority.

To bridge the gap between the complexity of traditional LCA approaches and the operational reality of SMEs, simplified tools are needed. As Gómez-Garza et al. (2023) argue, tailored methods are essential to support LCA uptake in resource-constrained contexts – such as the one of SMEs. In response to this need, the calculator developed in this study allows aluminum foundries to conduct a self-assessment of their environmental performance by entering site-specific data into a digital platform. This approach promotes a more agile and accessible form of environmental evaluation, supporting strategic decision-making and ultimately promoting wider participation in sustainability efforts and regulatory compliance.

This reinforces the crucial role of customized tools in overcoming traditional barriers to LCA adoption, paving the way for more inclusive and resilient sustainable development in the Italian aluminum sector. In fact, the customized calculator described in this paper aims to allow aluminum foundries not only to calculate their environmental performance, but also to do that in an agile and user-friendly way, inserting site-specific data on a digital platform that returns the eco-profile of their aluminum product (final or intermediate).

The developed tool has several practical implications, both for foundry managers and policymakers. In fact, while managers should cultivate a wider perspective, aligning their strategies with circular economy principles on a larger scale, policymakers can play key roles as both regulators and advocates for foundries, helping to create a supportive environment that enables their active participation in circular economy initiatives (Fellner et al., 2018).

From a managerial point of view, the calculator helps to develop an LCA of the aluminum product thus, allowing foundries to have critical data for external communication. Most of the Italian foundries belong to the supply chain of big companies which, under a normative point of view, need to collect data from the different actors of their supply chain. In this sense, foundries should be able to provide data as precise as possible (Feil et al., 2022). Moreover, LCA is crucial in supporting communication and marketing. Considered as a green marketing tool, besides a managerial one, LCA serves as the foundation for building a management strategy for emission reduction, which, if well communicated through appropriate indicators and Key Performance Indicators (KPIs), can evolve from being a purely technical tool to becoming a valuable element of strategic communication towards stakeholders.

An environmental assessment facilitates managers in the understanding of their production process, highlighting its strengths and weaknesses. Specifically, it could play a crucial role in identifying areas for improvement and establishing a strategy for continuous improvement. By providing a comprehensive assessment of environmental hotspots - such as energy consumption, emissions, and waste production - it enables managers to detect inefficiencies and prioritize actions for optimization (Finnveden et al., 2009). From a strategic perspective, the tool supports data-driven decision-making, allowing companies to set measurable sustainability goals, track progress over time, and adjust strategies accordingly. Moreover, by integrating eco-profile analysis into long-term planning, businesses can foster a culture of continuous improvement, enhance regulatory compliance, and strengthen their competitive advantage in an increasingly sustainability-focused market.

Ultimately, having a tool to calculate the eco-profile of a foundry can be highly beneficial for understanding its environmental performance, especially in the context of certifications like the MGI. The MGI is an environmental certification scheme promoted by the Italian Ministry of the Environment, based on the PEF methodology. It certifies the environmental performance of products made in

Italy, classifying them into three categories: A (better than the reference benchmark), B (in line with the reference benchmark), and C (worse than the benchmark; in this case, the logo is not granted). This classification is based on three threshold values based on a benchmark, which is the result of a sector-wide screening study conducted at the national level. Obtaining this certification requires companies to conduct a LCA of their products. However, if the assessment results place a product in Class C - indicating performance below the market benchmark - the company will not receive the certification. Conducting an LCA involves significant costs in terms of finances, expertise, and time, especially for small- or medium-size companies. Therefore, having the ability to self-evaluate a product's performance internally, without immediately commissioning an external LCA study, represents both an economic and strategic advantage (Singh et al., 2021). If a company finds that its product falls into Class C, it can implement an improvement plan and re-apply for certification once its environmental performance meets the required standards.

The tool also addresses a key issue in the Italian context: the lack of territorial representativeness in LCA studies available in the literature, which are often based on generic industry datasets (Brogaard et al., 2014; Liu and Müller, 2012). By enabling foundries to input localized, site-specific data, the calculator contributes to improving the accuracy and relevance of environmental assessments, which is crucial for ensuring reliable eco-profile evaluations (Rossi et al., 2025).

Last but not least, the developed tool could also be highly valuable for policymakers at the Italian level, particularly for defining PEFCR for aluminum foundries, building on the existing rules already developed for iron foundries under the MGI scheme. Since setting these rules requires data collection from foundries covering at least 51% of national production in terms of revenues - the threshold for representativeness in creating Product Category Rules

(PCR) - this tool can facilitate efficient and standardized data gathering across companies. Furthermore, the MGI scheme mandatorily requires that the proponents of PCR are at least 3 private companies, one of which shall be a small - or medium-size company. Encouraging broader participation through such a tool can support compliance with these procedural requirements. Achieving this level of representativeness requires coordination and data-sharing at scale. In this contest, the integration between the BRIEF and GRINS projects is strategic, as it leverages a broad network of participating companies.

A key barrier to overcome is to bridge the gap between the needs of industry and those of LCA practitioners. On one hand, LCA experts often lack access to primary data specifically collected for the geographical context under evaluation, which forces them to rely on industry-generic data in their analyses. On the other hand, foundries face barriers when advanced LCA modelling is required to assess the environmental impacts of their operations. As illustrated in Figure 5, the combined approach of the GRINS and BRIEF projects fosters mutual benefits: companies contribute data and, in return, receive simplified, tailored tools that enhance their capacity for self-assessment. At the same time, LCA experts are incentivized to support foundries in becoming more autonomous by providing tools adapted to their operational context. This collaborative model could help reach the data representativeness threshold required by MGI, laying the groundwork for the development of sector-specific PCRs for aluminum.

In our experience, the benefits of this virtuous cycle are evidenced by the increasing participation of foundries in the data collection campaign. Based on the data collected in GRINS, a first version of a calculator was developed within the BRIEF project. This tool has since been tested by some foundries that had not contributed with data during the GRINS data collection campaign. Two of them found the calculator poorly suited to their processes and are now

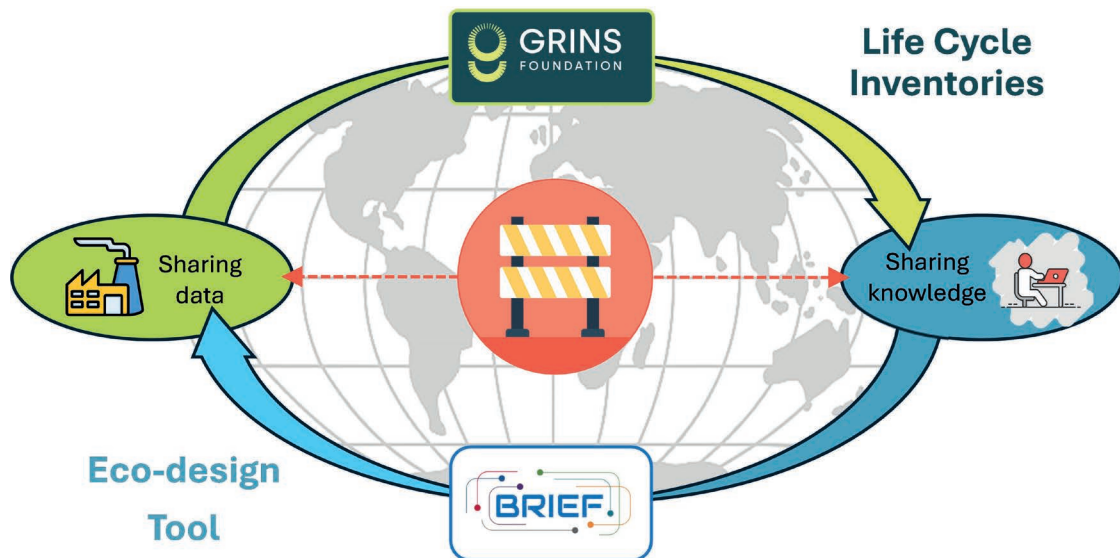


FIGURE 5: Sketch of the synergy between the projects GRINS (Growing Resilient, Inclusive and Sustainable) and BRIEF (Biorobotics Research and Innovation Engineering Facilities).

considering providing data to better tailor the tool to their specific context. We believe that this synergy between foundries and LCA experts - currently involving a few dozen facilities - has the potential to grow exponentially as the calculator is adopted on a larger scale. This is essential to achieving the goal of defining PCR for MGI or other labelling schemes that require extensive representativeness at a certain geographical scale (e.g., national level).

However, the ways in which the data collection campaign will be extended is currently under definition. Moreover, by encouraging a widespread adoption of this tool among foundries, the ultimate goal is to drive the national industrial development towards decarbonization and broader green transition objectives, supporting a more sustainable and competitive Italian manufacturing sector.

4. CONCLUSIONS

The goal of this study was to present the development and application of a user-friendly Eco-Design tool aimed at supporting Italian aluminum foundries - particularly SMEs - in conducting a preliminary self-assessment of their environmental performance. The tool was designed to fill a gap in the availability of site-specific, accessible LCA-based instruments tailored to the operational context of smaller foundries, which often lack the capacity to perform full LCA studies. It provides indicative environmental profiles based on real production data, helping companies identify environmental hotspots and align with circular economy principles.

The tool has already been tested by 7 foundries, and feedback suggests it is perceived as useful and comprehensive by those that participated in the initial data collection. Nevertheless, the tool has some limitations. First, the accuracy of the results depends heavily on the quality and completeness of the input data provided by each company. Although the tool includes guidance for users, manual data entry can introduce inconsistencies. Furthermore, the current version of the calculator is based on data collected from only 15 foundries, meaning that results cannot yet be generalized to represent the entire Italian aluminum sector. However, the applicability of the software has significantly improved after implementing the feedback from the foundries. A practical demonstration of the results that could be returned by the software are presented in this manuscript, highlighting how they could be interpreted using an eco-design perspective.

From a practical standpoint, the tool offers an accessible entry point for companies wishing to understand and improve their environmental performance. It can support internal decision-making and external communication - especially for SMEs required to provide environmental information to larger clients or regulatory bodies. Moreover, the tool supports companies in evaluating their eligibility for environmental certifications such as MGI. In this sense, we believe that a synergy between the GRINS and BRIEF projects has a high potential, that is currently unexplored at large scale, to extend the platform of foundries in a virtuous cycle towards the achievement of a strong representativeness of the data collected on the Italian territory.

Future work will focus on expanding the user base and increasing the territorial representativeness of the data collected, with the goal of reaching the 51% production threshold required for the development of sectoral PCR under the MGI scheme. Further developments may also include refining the user interface based on user feedback and enhancing modularity to include additional production configurations. At the company level, future developments could include integrating the tool with enterprise management systems. By automating data collection, this integration would allow the tool to self-update with real-time information, reducing the burden of manual data entry and improving the reliability of results. This would not only streamline operations, but also make the tool a more effective resource for monitoring and improving environmental performance over time, therefore helping to bridge the gap between LCA theory and LCA practice.

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Extra contents

COLUMNS AND SPECIAL CONTENTS

This section comprises columns and special contents not subjected to peer-review

Environmental Forensics, Law and Policy

The price of industrial influence in EU PFAS regulation

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1. INTRODUCTION

Per- and polyfluoroalkyl substances (PFAS) represent a family of over 10,000 synthetic chemicals characterized by strong carbon-fluorine bonds that make them extraordinarily persistent in the environment. These “forever chemicals” have been widely used since the 1950s across an expansive range of industrial and consumer applications—non-stick cookware, water-resistant textiles, firefighting foams, and semiconductor manufacturing, among many others (Glüge et al., 2020; Buck et al., 2011). What makes PFAS particularly concerning is not merely their ubiquity, but the combination of their environmental persistence with mounting evidence of bioaccumulation and links to serious health effects including cancer, immune system dysfunction, and reproductive disorders. The extreme resistance to degradation means that even if all PFAS uses were phased out today, environmental contamination would persist for generations (Dobrzyńska et al. 2025; Cousins et al., 2020).

Currently, the EU regulates PFAS one substance at a time. While three PFAS are banned under the POPs Regulation (Regulation (EU) 2022/2400) and several others are listed as substances of very high concern under the EU’s chemical regulation REACH (Registration, Evaluation, Authorisation and Restriction of Chemicals) (Regulation (EC) No 1907/2006), the vast majority of the 10,000+ PFAS remain unregulated. Globally, conflicts over PFAS cleanup liability are increasing—in courtrooms, in settlements, and in political negotiations over who bears the cost of remediation. Legal judgments, however, cannot undo environmental contamination. Without comprehensive restriction, PFAS will continue accumulating in nature faster than remediation efforts can address it.

In early 2023, five European countries—Denmark, Germany, the Netherlands, Norway, and Sweden—submitted a proposal to the European Chemicals Agency (ECHA) for a universal restriction of PFAS under REACH. This proposal seeks to ban the entire PFAS family, with limited derogations until alternatives can be developed (ECHA, 2025). The scope and ambition of this restriction are unprecedented in chemical regulation history. During ECHA’s public consultation period in 2023, the agency received over 5,600 submissions—a record number reflecting the profound societal and economic implications of regulating an entire chemical class.

Yet the formal consultation process tells only part of the story. The proposal has triggered an intensive lobbying campaign. The Forever Lobbying Project, a cross-border journalistic investigation involving 46 journalists across 16 countries, has documented this campaign, collecting over 14,000 documents related to industry lobbying efforts—constituting the world’s largest collection of PFAS industry documents to date (The Forever Lobbying Project, 2025).

2. DOCUMENTING THE LOBBYING CAMPAIGN

The lobbying effort against PFAS restriction has been particularly intensive in the fluoropolymer sector. Fluoropolymers, including well-known products like Teflon, represent a lucrative segment of PFAS manufacturing. Industry actors, led by Plastics Europe and its Fluoropolymer Product Group (FPG), have coordinated a systematic campaign to secure exemptions for fluoropolymers from any comprehensive ban (The Forever Lobbying Project, 2025).

These 14,000+ documents, obtained through extensive Freedom of Information requests across European institutions and member states, reveal how industry arguments have circulated through European policy networks—from corporate boardrooms to the European Commission, and onward to national governments in Germany, Sweden, Belgium, and beyond (The Forever Lobbying Project, 2025).

To assess the credibility of these arguments, the investigation developed a “stress test” methodology in collaboration with researchers at the University of Bristol specializing in corporate influence. Applied to 1,178 lobbying arguments from the plastics sector, this systematic evaluation revealed a troubling pattern: many of the industry’s key claims rest on questionable foundations (The Forever Lobbying Project, 2025).

Three pillars support the industry’s case for exemptions: scientific claims, assertions of technical necessity, and predictions of economic catastrophe. Each deserves scrutiny.

3. THREE PILLARS OF INDUSTRY ARGUMENTATION

3.1 Scientific Claims: The Case of Non-Existent Criteria

A cornerstone of the fluoropolymer industry’s scientific argumentation revolves around the concept of “Polymers



of Low Concern” (PLC)—the claim that fluoropolymers’ large molecular size prevents them from penetrating cell membranes and causing biological harm, thus rendering them safe despite their PFAS composition. In 997 instances across their lobbying documents, industry actors invoke “PLC criteria established by the OECD” to justify excluding fluoropolymers from the proposed ban (The Forever Lobbying Project, 2025). They cite two scientific articles as their foundation for this claim (Henry et al., 2018; Korzeniowski et al., 2023).

However, when the Forever Lobbying Project contacted the Organisation for Economic Co-operation and Development (OECD) directly, the response was unequivocal: “No agreed-upon set of criteria at the OECD level was finalized” (The Forever Lobbying Project, 2025). While OECD has convened expert meetings to discuss potential PLC criteria since 1993, these have received limited uptake in regulatory frameworks, and no harmonized international standard has been established (OECD, 2024). The PLC criteria, referenced nearly a thousand times in industry submissions, do not exist as an official OECD standard.

Further examination of the two cited scientific articles reveals significant conflicts of interest. All co-authors of both studies are either employees of or consultants to major fluoropolymer manufacturers including 3M, Chemours, DuPont, Arkema, and Solvay. The lead author of one study, Stephen H. Korzeniowski, worked for 37.5 years at DuPont before establishing Beachedge Consulting, which according to its website provides “advocacy work” for DuPont and its lobby organizations (The Forever Lobbying Project, 2025). Independent researchers have raised serious concerns about the scientific validity of treating fluoropolymers as low concern, pointing to pollution from production, emissions during use, and persistent contamination (Lohmann et al., 2020).

The pattern here follows a familiar playbook. Cite industry-funded research with conflicts of interest as though it represents independent science. What matters is not winning the scientific debate—that battle has already been lost. Rather, the strategy aims to manufacture just enough doubt to delay regulation for another few years, and then a few more after that, until companies have extracted every last profitable year of production while leaving society to bear the consequences.

3.2 Technical Necessity Claims: The “No Alternative” Narrative

The second pillar of industry argumentation centers on technical indispensability. Industry actors claim that fluoropolymers are irreplaceable for critical applications in the green transition, digitalization, medical technology, and Europe’s strategic autonomy. The Forever Lobbying Project collected 525 such “no alternative” claims from industry submissions. However, only 134 of these contained sufficient technical detail to permit verification. In collaboration with Stockholm University researchers, the investigation tested these claims against the Alternative Assessment Database developed within the EU-funded ZeroPM project. The results showed that for nearly two-thirds of the 134

verifiable cases, potential alternatives exist (The Forever Lobbying Project, 2025; Figuière et al., 2025).

Perhaps most revealing is the temporal dimension of this argument. The investigation uncovered documents showing that the US Fluoropolymer Manufacturers Group deployed virtually identical “critical applications” arguments as early as 2000, citing aerospace, automobiles, semiconductors, and medical devices as sectors requiring continued PFAS use to justify delaying regulation. Twenty-five years later, the same list appears in contemporary lobbying documents, raising questions about the industry’s investment in alternative development. As researcher Romain Figuière observes, “It seems they spend the money to slow down the process instead of spending [it] on investing in finding alternatives” (The Forever Lobbying Project, 2025).

The timeline reveals the scale of this delay. From the year 2000 to today spans a quarter-century. Factor in the regulatory process—another five to ten years before any restriction takes effect—followed by transition periods extending up to twelve years for applications like medical devices. The calculation is striking. The industry will have enjoyed nearly fifty years of continued production since it first deployed these arguments. This extended timeframe suggests that the strategy has been less about finding alternatives and more about prolonging the status quo for a chemical class whose risks were already apparent at the turn of the millennium.

3.3 Economic Impact Predictions: A History of Overestimation

The third pillar involves dire predictions of an economic catastrophe. According to industry warnings, PFAS restrictions will trigger sectoral collapse, frozen investments, supply chain disruption, and mass unemployment. Historical precedent suggests caution in accepting such predictions. During the REACH debates in the mid-2000s, the chemical industry predicted catastrophic economic losses of €20–30 billion. The actual costs proved to be €3 billion—barely above the Commission’s projections and a fraction of industry’s exaggerated forecasts (The Forever Lobbying Project, 2025; ChemSec, 2019).

The foundation of these economic warnings is shaky at best. Industry-commissioned impact assessments openly acknowledge that their figures rest on assumptions, incomplete data, and significant methodological limitations (Ricardo Energy & Environment, 2023). Even basic figures prove elusive. They failed to provide consistent employment numbers for fluoropolymer manufacturing, offering only a vague estimate of fewer than 5,000 workers across Europe (The Forever Lobbying Project, 2025).

What industry systematically ignores, however, are the economic benefits of restriction. Healthcare costs alone from PFAS exposure reach €52–84 billion annually across Europe, according to calculations by the Nordic Council of Ministers (Nordic Council of Ministers, 2019:14). The remediation bill—if contamination continues unchecked—will total €2 trillion over two decades, or €100 billion per year (The Forever Lobbying Project, 2025).

These costs do not appear in industry's economic models, and their absence represents a deliberate methodological choice rather than mere oversight. By excluding the costs borne by healthcare systems, communities, and future generations, industry presents a fiction: that restriction is expensive and inaction is free.

4. THE MECHANISM OF INFLUENCE: OWNING THE LANGUAGE

The effectiveness of lobbying is perhaps most visible not in the arguments themselves but in how they have penetrated official policy discourse. According to the Forever Lobbying Project's investigation, in April 2024, European Commission President Ursula von der Leyen wrote to industry representatives: "PFAS are currently needed for critical applications for the green and digital transitions and for the EU's strategic autonomy [...]" (cited at the Forever Lobbying Project, 2025). This language is not coincidental. "Green transition", "digital transition", "strategic autonomy", and "critical applications" appear both in industry lobbying arguments and in the Commission President's official correspondence—suggesting successful penetration of industry framing into policy discourse.

This phenomenon reflects what scholars of regulatory capture describe as "cultural capture"—the process by which regulators come to see the world through "shared but not explicitly stated understandings [...]" (Kwak, 2014:79) provided by regulated industries. Successful lobbying is not simply about financial influence or direct corruption; it involves shaping the terms of debate, defining what counts as "realistic," and determining which questions are considered relevant. Through these mechanisms, regulators become susceptible to pressures that emerge from the administrative process itself, ultimately producing regulatory outcomes that serve industry interests (Kwak, 2014:76-79; see also Chesterfield et al. 2025).

The tactics documented by the Forever Lobbying Project follow a well-established playbook. The plastics industry has adopted influence strategies previously deployed to defend tobacco, fossil fuels, and other controversial chemicals like Monsanto's glyphosate. The project calls this the work of "Merchants of Doubt", who have now polluted the public debate on PFAS (The Forever Lobbying Project, 2025). When von der Leyen's correspondence mirrors industry talking points so precisely, cultural capture has moved beyond theory into documented practice. The fundamental question—whether to ban these persistent pollutants—has been quietly replaced by a narrower debate about transition timelines and exemption criteria, as if the necessity of continued PFAS use were already settled fact rather than an industry assertion requiring scrutiny.

5. TOWARD EVIDENCE-BASED POLICY

The Forever Lobbying Project's investigation reveals a systematic gap between industry claims and verifiable evidence in three critical domains: the scientific basis for exemptions, the technical necessity of continued PFAS use, and the economic consequences of restriction. Address-

ing this gap requires structural changes to how evidence is evaluated in regulatory processes.

First, the burden of proof must rest squarely on industry. Claims about the safety, necessity, or irreplaceability of chemical substances should be supported by independent scientific research, not industry-funded studies with undisclosed conflicts of interest. When authoritative bodies like the OECD confirm that cited criteria do not exist, this should trigger immediate scrutiny of all arguments dependent on those criteria. ECHA should establish a mandatory pre-registration system requiring industry applicants seeking exemptions to submit all supporting studies for independent peer review before exemption applications are formally considered. This mechanism could build upon REACH's existing registration requirements by adding a verification stage where claimed scientific criteria must be validated against established databases and peer-reviewed literature before entering regulatory deliberations. Industry submissions citing non-peer-reviewed or proprietary research should be assigned lower evidentiary weight in ECHA's risk assessment committees unless the underlying data and methodologies are made publicly available for independent verification.

Second, transparency in policymaking must be enhanced. Freedom of Information requests should not be necessary to reveal that non-existent scientific criteria have dominated regulatory debates for years. All meetings between industry representatives and policymakers, all submitted documents, and all lobbying arguments should be proactively published and easily accessible to researchers, civil society, and affected communities. The European Commission should expand its Transparency Register to require real-time disclosure of all technical submissions and position papers submitted during REACH restriction processes, with a centralized public database modeled on the U.S. FDA's docket system. Member States should mandate that national authorities participating in REACH committees publish meeting minutes, stakeholder submissions, and voting records within a reasonable timeframe of each session.

Third, regulatory impact assessments must account for the full social costs of inaction. When industry calculates potential losses from regulation while ignoring €52-84 billion in annual healthcare costs and €2 trillion in long-term remediation expenses, the economic analysis is fundamentally incomplete. Comprehensive cost-benefit analysis must include environmental and health externalities currently borne by society rather than producers. The Commission's Better Regulation guidelines should be amended to require that all REACH restriction proposals include mandatory health impact assessments and environmental remediation cost projections prepared by independent economic consultants rather than industry-commissioned studies. These assessments should employ the "polluter pays" principle embedded in Article 191(2) TFEU by calculating the net present value of long-term contamination costs and assigning them to the regulatory scenario that permits continued use. ECHA's Socio-Economic Analysis Committee should be required to commission independent economic analyses as a standard practice for major re-

striction proposals, ensuring that regulatory decisions rest on balanced economic evidence rather than uncontested industry projections.

6. CONCLUDING REMARKS

PFAS regulation raises questions that extend far beyond chemicals policy, touching fundamental issues about how democratic societies evaluate evidence, allocate environmental burdens, and protect public health against well-resourced opposition. The Forever Lobbying Project has provided an unprecedented documentary foundation for understanding industry influence in this domain. Whether policymakers will use it to critically evaluate industry claims and resist cultural capture as the EU moves toward a final decision remains uncertain.

At stake is the question of who will bear the costs of PFAS contamination—the polluters or society at large. This is not simply a matter of distributing €2 trillion in long-term expenses. It is a test of whether evidence-based policy can withstand systematic campaigns of manufactured doubt when those campaigns are well-resourced and strategically sophisticated. The integrity of European policymaking depends on finding an answer that insulates regulatory decisions from the forms of influence this investigation has documented. What happens with PFAS will define not only the future of chemicals regulation but the resilience of evidence-based governance itself.

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DETRITUS & ART / A personal point of view on Environment and Art

by Rainer Stegmann

Artificial Intelligence (AI) is becoming an increasingly important part of our lives, and we do not know where it is already implemented or where it will go. Of course, we use it too in our scientific lives, and we have to take care not to get lazy in searching for the appropriate literature, lose part of our creativity, and become increasingly dependent.

Apropos creativity: I wanted to know how creative AI is in the area of Waste to Art. I asked my AI counterpart to create some artwork with this content. The results are mixed. When I used ChatGPT Brutus, the results were poor. Asking for a Waste to Art (WtoA) painting in the style of Rembrandt AI, just added some bottles and other items to an existing painting. AI also added used plastic bottles. Making AI aware of it –AI (how else can I call it?) (her?) him?) admitted its mistake. When I proposed other famous artists to AI, the results were similar, poor.

Then I used Dreamina AI Video Magic (dreamina.com), and the results were much better. Looking at the sculpture, Sayaka Ganz's style was not badly imitated, but, upon closer inspection, one can see it is far from substituting for the artist. Using the famous painting The Scream, created by Edvard Munch, as a basis for WtoA AI, made a mess out of it. For me, it looks more like a persiflage than a serious discussion of the given task. It actually looks funny to me. AI uses a similar style again, adding bottles and trash. The collage AI created from paper, textile, and plastic



Painting in style of Edvard Munch (1863-1944).

turned out, in my opinion, quite reasonable, except for the piece of plastic added more by chance, with no connection to the collage.



Sculpture made from plastic items in style of Sayaka Ganz (<https://sayakaganz.com>)



Collage using paper, textile and plastic.



What do we learn from this experiment? First of all, I am a complete layman, happy to use the basic functions. An expert can find better-suited programs. Reports indicate that AI can create paintings in the style of other painters, hardly distinguishable from originals. However, the examples presented here already show the potential of AI, and that frightens me.

AI will dramatically change our lives and the world. This development has just started, and considering the billions of dollars already invested and those that will be invested in this area.

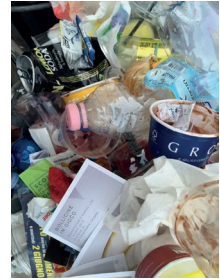
Of course, AI has many positive aspects and will make our lives easier and more comfortable, and make dull work redundant, but at what expense? It is our task to keep such expenses low.

Maybe in the future, AI will take over this DETRITUS Art Page; hopefully, we will increase our creativity so that it will take a while.

*In the next Issue I
will come back to
reality and I want to
open your eyes for
the beauty of trash
supported by a kind
of poem.*

"GARBAGE STORY"
Rainer Stegmann & Cristina v.d.
Westhuyzen

„Every item in this garbage bin has
a story to tell: plastic, paper and
tin. The remains of a busy day in a
city.
Water bottles, a fuzzy drink, an ice
cream cup smudged in melted
chocolate, an invitation to an
evening of music and wine, a
scrunched up ticket. Was it for a
museum?
The trail of leisurely time spent
strolling around in the warm sun.
The air is vibrant with sound. The
piazza is buzzing with many
people, colourful and confused like
the jumbled display in the bin.
Happy memories will be taken but
this will remain.“



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