



Bioconversion of Biomass Waste Drives Sustainable Development

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Abstract

Global population growth has created enormous amounts of biomass waste generated across agroforestry systems, animal husbandry, and municipal sectors. According to recent estimates, municipal solid waste has reached approximately 1.3 billion tons each year, alongside annual discharge volumes exceeding 170 million metric tons originating annually from pulp manufacturing sectors, necessitating urgent measures in waste conversion and resource recovery. Here, we review the classification and composition of biomass waste, with a focus on bioconversion technologies that include pretreatment, anaerobic digestion, fermentation, and composting. Analysis shows that certain microbial pretreatment strategies can achieve lignocellulose hydrolysis efficiency above 80, while anaerobic digestion can yield more than 300 mL methane/g.VS. It also highlighted the potential of insects such as black soldier fly larvae for both waste reduction and feed production. In addition, bibliometric results indicate that bioconversion has been a major research emphasis, with more than 470 occurrences in relevant literature from 2015 to 2024. This article concludes that bioconversion offers a sustainable path to address resource depletion, climate warming, and carbon neutrality, although improvements are still needed to enhance process efficiency and reduce overall costs.

Keywords: Bioconversion; Fermentation; Biofuels; Anaerobic digestion; Sustainable development.

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1. Introduction

UN demographic records indicate that human demographics have tripled since the mid-20th century.^[1] Rapid population growth has resulted in a surge in waste generation across sectors such as agriculture, industry, and households, posing significant waste management challenges and environmental hazards.^[2] Therefore, the transformation and utilization of biomass waste are essential to combat planetary heating and minimize climatic destabilisation while also providing raw materials for energy, chemicals, and materials to ensure the sustainability of human societies.^[3] Biomass constitutes a perpetually renewable carbon reservoir as its organic constituents originate from photosynthetic organisms and

heterotrophic fauna.^[4] Biomass waste can generally be categorized into agricultural and forestry waste, food waste, industrial organic waste, municipal solid waste, and livestock waste.^[5] Traditional biomass waste management often involves direct combustion or landfilling, which are not only inefficient but also create problematic issues including impacts on biodiversity, climate warming, and environmental pollution.^[6] The transformation of biomass waste, which has historically been regarded as valueless, into high-quality products for sustainable development has gained significant interest and attention.

Strategies for transforming biomass waste into value-added products are broadly categorized into two technological pathways, namely biotechnological processing and thermochemical valorisation.^[1] The latter uses heat to change the biomass waste into energy-dense substances.^[7,8] However, thermochemical processes often require additional chemicals, which may result in the formation of undesirable by-products that have the potential to be harmful to the environment. This presents challenges in utilizing these methods within sustainable waste management practices.^[9] As of October 16, 2024, a search of the Web of Science Core Collection was conducted using the terms '(biomass waste)' and/or

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Agriculture ranks as the second most significant contributor to greenhouse gas emissions after the energy sector while concurrently yielding substantial quantities of solid residues.^[18] The United Nations defines agricultural waste as waste generated from various agricultural operations.^[17] Agricultural biomass comprises three primary categories, namely crop waste, fruit or vegetable waste, and non-biomass hazardous fractions.^[19] Hazardous waste typically contains chemical constituents and is excluded from biomass classification. Crop residues refer to field-generated byproducts during cultivation,^[20] including leaves, cereal stovers (maize, rice, wheat), oat/barley straws, and seed pods. Current global production of crop residues reaches 28.02 million tonnes/year,^[21] with major contributors including cereal straws (wheat and rice) at 731 and 354 million tonnes annually, maize stover (204 Mt), and processing byproducts (rice husks and sugarcane bagasse) at 181 and 110 Mt respectively.^[22,23] The structural matrix of biomass predominantly contains cellulose, lignocellulosic architecture (lignin-hemicellulose complexes), and pectin.^[18] Fruit or vegetable processing wastes encompass peels, seeds, stems, roots, and tubers that are rich in bioactive compounds like dietary fibres, carotenoids, polyphenols, and polysaccharides.^[24,25]

Forestry waste refers to residues from forestry production and processing,^[26] such as logging residues, sawdust, discarded wood products and shavings.^[27] Globally, 460 million tonnes of wood biomass waste is produced annually, of which 20% is production loss, causing serious disposal problems and low utilization efficiency.^[6] Forest organic waste components contain significant amounts of lignocellulose and nutrients,^[28] and such losses can be converted into a fuel source.^[29] Industrial sludge effluent and black liquor from production processes are the main components of industrial biomass waste.^[6] Nearly 75% of the global industry uses sulphate-based pulping.^[30] This lignocellulosic fractionation process generates black liquor (BL) as a predominant byproduct,^[31] with annual global production exceeding 170 metric tons in the pulp and paper sector.^[32] BL has a solids content of about 65–85% and consists mainly of organic matter obtained from inorganic matter and lignocellulosic biomass from cooking chemicals such as NaOH, Na₂S and NaCO₃ in pulp and paper mill digesters.^[33] Sludge effluent is classified as a semi-solid residual stream. This byproduct predominantly contains extracellular polymeric substances (EPS), effluent-derived organic fractions, microbial cell aggregates, inorganic grit particulates, and adsorbed heavy metal ions.^[34] This compositional heterogeneity imposes significant constraints on conventional sludge management strategies, including landfilling, thermal oxidation, and aerobic bioconversion.^[35]

Every year, food systems generate around 1.3 billion metric tons of organic waste, costing over 1 trillion USD.^[15] These waste streams originate from multiple nodes across the food supply chain, including distribution hubs, industrial processing facilities, residential sectors, and both commercial and

domestic culinary operations. Conventional disposal methods such as burning waste with other materials for heat or energy are common but harmful to the environment.^[36] These methods pollute our water and soil and create unpleasant smells.^[37] However, food waste is a valuable resource as it is made up of lignocellulosic material, lipids, enzymes and nutrient-rich organic acids.^[38] Through specific processes such as anaerobic digestion or thermochemical cascades, these carbon-rich feedstocks can be turned into biofuels, compost, and platform chemicals.^[39]

Global municipal solid waste (MSW) generation currently stands at 1.3 billion tonnes annually, projected to surge to 2.2 billion tonnes/year by 2025 with organic matter constituting ~46%.^[40] MSW is sourced from commercial entities, institutions, offices, and households,^[41] with textiles and waste paper as its predominant constituents.^[6] Textile waste primarily includes discarded garments and household items like carpets, footwear, and linens.^[42] Waste paper encompasses three subtypes, namely printing sheets, cardboard, and sanitary paper,^[43] while other MSW comprises yard debris, plastic containers, furnishings, and rubber products.^[44] MSW represents a sustainable and cost-effective reservoir amenable to resource recovery via physical, chemical, and biological technologies.^[45]

The majority of animal waste is generated from fisheries, livestock waste, and meat and poultry processing waste,^[46-48] with fisheries waste including all potentially utilizable material removed from crustaceans, shellfish, and fish.^[49] Livestock production generates wastes in liquid and solid forms including urine, wastewater, disinfectants, manure, feed wastes and soiled bedding.^[23] Due to high levels of nitrogen (N) and phosphorus (P), if left untreated, it may lead to severe water pollution and eutrophication of surface waters.^[17] Abattoir waste consists mainly of rumen, faeces, blood, and other potential resources for energy production. Poultry solid waste consists mainly of feathers, bones, skin, tendons, feed and hatchery waste, whereas liquid wastes generated include faeces, urine, insecticides and others.^[50]

In conclusion, the wide variety of biomass wastes including agricultural residues, forestry by-products, industrial effluents, food system discards, municipal solid wastes, and by-products from animal husbandry, represents both a significant environmental challenge and a valuable renewable resource. These materials primarily composed of lignocellulosic structures, lipids, and nutrient-rich organic acids, require integrated valorisation strategies that go beyond conventional disposal methods. By strategically employing thermochemical processing, anaerobic digestion, and tailored biochemical conversion technologies, these carbon-rich substrates can be transformed into electricity, heat, biofuels, platform chemicals, and soil amendments. Future research should focus on optimizing conversion efficiencies, harmonizing the treatment of heterogeneous feedstocks, and conducting comprehensive life-cycle assessments to fully harness the sustainable potential of biomass wastes in the context of

circular bioeconomies.

3. Biological pretreatment

Pretreatment constitutes a pivotal methodological intervention for facilitating efficient bioconversion of biomass waste. The categories of pre-treatment methods commonly used are generally divided into biological, chemical and physical pretreatment. The objective of the various pretreatment methods is to deconstruct lignin, hemicellulose, and cellulose effectively. This process aims to convert these complex polymers into smaller fragments, facilitating easier digestion by enzymes and enhancing subsequent biorefinery processes. Ultimately, this approach allows the production of a range of valuable products. However, each pretreatment method has its pros and cons.^[51,52]

Both physical and chemical processes are expensive, whereas biological pretreatment is a low-energy, safe, green, and efficient process compared to traditional chemical and physical pretreatment methods.^[53,54] Biological pretreatment utilizes microorganisms (enzymes, fungi, and bacteria) to degrade the complex biopolymer structures (especially hemicellulose and lignin) in biomass wastes.^[55] Fungal-mediated biomass degradation predominantly involves three functional guilds: white-rot fungi, ascomycetous fungi, and brown-rot fungi.^[19] Among these, white-rot fungi are recognized as the predominant agents for pretreatment due to their enzymatic arsenal that synergistically disrupts lignin-cellulose matrices through targeted oxidative cleavage.^[56,57] Compared with fungi, bacteria possess the advantages of adaptability, rapid reproduction and small size. Hence, bacteria can be used in the digestion pretreatment process for hydrolysis of lignin and hemicellulose.^[58] The bacterial lignolytic mechanism involves two spatially segregated phases, namely extracellular depolymerization of lignin macromolecules followed by intracellular catabolism of aromatic intermediates.^[59] The lignin-degrading bacteria mainly include *Actinobacteria*, *α-Ascomycetes* and *γ-Ascomycetes*.^[60] Biological pretreatment exhibits inherent operational merits including energy-efficient processing, absence of chemical additives, and ambient reaction parameters. However, in most biological pretreatment processes, the treatment rates are very low and there is much room for optimization.^[56]

4. Bioconversion

Thermochemical and bioconversion methods are two fundamental approaches to bioenergy conversion derived from biomass and waste.^[61] These methods encompass a range of processes including liquefaction, combustion, pyrolysis, and gasification.^[62] While thermochemical processes allow a more extensive utilization of biomass, they tend to incur higher processing costs. Additionally, the resultant products are often complex mixtures that exhibit low concentrations and are highly dependent on the characteristics of lignocellulosic feedstocks.^[63] In contrast, the development of value-added

products through microbial processes is recognized as one of the most environmentally sustainable technologies. Microbial cell factories are engineered for robust conversion selectivity and adaptability, necessitating relatively straightforward facilities and enabling operations tailored to specific objectives.^[5] Bioconversion specifically refers to the biological transformation of carbonaceous feedstocks found in organic waste streams into bioenergy carriers and valuable biochemicals through biocatalytic processes. This transformative process is supported by the involvement of microorganisms, insects and invertebrates.^[64-66]

4.1 Anaerobic digestion

Anaerobic digestion (AD) represents a highly studied bioconversion process, functioning as a critical pathway for decarbonization efforts aimed at achieving net-zero targets. This process facilitates the valorization of various biowaste materials, including wastewater, urban organic solid waste, agricultural residues, and biomass derived from livestock.^[13,62,67] Furthermore, it encompasses the conversion of waste oils and animal fats into valuable products, such as biomethane and biogenic carbon dioxide.^[67,68] Notably, AD offers several significant advantages over traditional aerobic treatment methods, including reduced energy requirements and lower sludge production.^[69]

The anaerobic digestion process occurs in a heated, sealed, airless vessel, which creates ideal conditions for bacterial fermentation of organic matter under anaerobic conditions.^[70] The reaction systems used in anaerobic digestion can be categorized according to the stage of fermentation (two-stage or one-stage digestion), the mode of feed (continuous, semi-continuous or batch digestion) and the reaction rate of substrate loading (high or low).^[71,72] Hydrolysis, the initial phase of transforming biomass waste within AD systems, is inhibited under substrate recalcitrance, especially when treating waste with high solid content.^[73] Acidogenic microbiota metabolize hydrolyzed substrates to generate volatile fatty acids (VFAs) along with hydrogen derivatives, NH₃, H₂, CO₂, lactate, and alcohols.^[74] While VFA accumulation enhances methane yield, exceeding system tolerance limits causes pH reduction and microbiota dysregulation, destabilizing AD processes.^[75,76] The third phase involves acidogenic microbiota converting acidogenic byproducts into CO₂/H₂/acetate,^[71] while methanogenic archaea mediate the final AD step through anaerobic biomass conversion to biomethane.^[77]

In conclusion, the primary challenges associated with anaerobic digestion at present include its relatively low overall efficiency in degrading organic matter and the extended residence time required for the process.^[68] Hydrolysis serves as the rate-limiting step within this biochemical mechanism. To address these issues, various biological, chemical, and physical pretreatment methods have been suggested to facilitate the release of extracellular polymeric substances (EPS) into the aqueous phase, thereby aiding in the disruption of flocs.^[78]

Subsequently, the process progresses to cell division and the release of intracellular components into the soluble phase, which aims to enhance degradation efficiency.^[79] These limitations may be alleviated through the co-digestion of diverse feedstocks and the adjustment of appropriate temperature, pH, and reactor parameters.^[80,81] Table 1 presents the efficacy of diverse pretreatment protocols on the methane productivity of different biomass wastes.

The enhanced metabolic activity and non-oxidative bioenergy generation depend on the substrate properties as well as the selection of appropriate bioreactor designs.^[82] High-rate anaerobic systems are effective in reducing the duration of waste treatment in the reactor and have well-designed heating and agitation systems.^[72] A small-scale biodigester implementation in Peru's Chillon Valley demonstrated 25% efficiency gains through optimized free volume ratios and agricultural waste-cattle manure co-digestion, validating these optimization principles in rural settings.^[83] Recently, there has been a growing abundance of optimization techniques for anaerobic fermentation, such as the addition of specific microorganisms (bioaugmentation), bioelectrochemical reactors, light and laser irradiation, and the addition of biochar.^[75,84-86] The main biotransformation mechanisms of action of the various improvements in the AD process are illustrated in Fig. 2. Pretreatment is crucial as it enhances cellulose accessibility, facilitating efficient hydrolysis and is applicable across diverse conversion technologies (Fig. 2A). Contemporary research prioritizes single- or multi-stage methods that target the removal of hemicellulose and lignin, as this delignification directly affects the bioavailability of cellulose for microbial degradation.^[87] Fig. 2B contrasts non-biochar reactors (low methane yield, long lag phase, instability from ammonium/heavy metals) with biochar-amended reactors. The latter's enhanced AD efficiency arises from three key mechanisms under controlled pyrolysis/dosing: Direct Interspecies Electron Transfer facilitation, inhibitor adsorption, and microbial support. Fig. 2C presents a

schematic diagram of an integrated Microbial Electrolysis Cell coupled with Anaerobic Digestion (AD-MEC). The cylindrical reactor contains two vertical electrodes (anode and cathode) embedded within a layer of active carbon granules. A low external voltage applied via the top-mounted power source drives electron transfer between electrodes. Simultaneously, biogas is collected from the gas outlet while liquid parameters are monitored through a sampling port connected to a peristaltic pump. This system leverages active carbon granules to adsorb inhibitors and provide microbial colonization sites, thereby enhancing methane yield and stabilizing operational performance. Fig. 2D illustrates the catalytic role of zero valent iron (ZVI) in anaerobic digestion through a cyclic iron redox process. Within the triangular reaction zone, ZVI corrosion releases ferrous ions and electrons, while metal-reducing bacteria drive $\text{Fe}^{2+}/\text{Fe}^{3+}$ interconversion. This process reduces oxidation-reduction potential (ORP), stabilizes pH, and stimulates enzymatic activity in methanogens, collectively enhancing biogas production and wastewater purification as annotated by upward arrows. In a sequential processing scheme, biodigested biosolids undergo dewatering, with resultant digestate and moisture-reduced residual solids being valorized for composting and soil amendment production.^[13] These products facilitate plant growth and enhance disease resistance while also mitigating agricultural surface pollution and contributing to food security.^[88] Recent advances demonstrate the viability of converting anaerobic digestion-derived methane into high-value nutritional products as an alternative to conventional energy generation, leveraging innovative bioreactor configurations where methane serves as a feedstock for microbial protein synthesis; notably, a gas-delivery membrane bioreactor utilizing hollow fiber membranes achieves near-complete substrate utilization with 100% CH_4 and NH_4^+ conversion while enabling suspended growth of inter-species methanotrophic consortia, yielding single-cell protein (SCP) at 1.36 g/g CH_4 with 67% protein content and over 42% essential

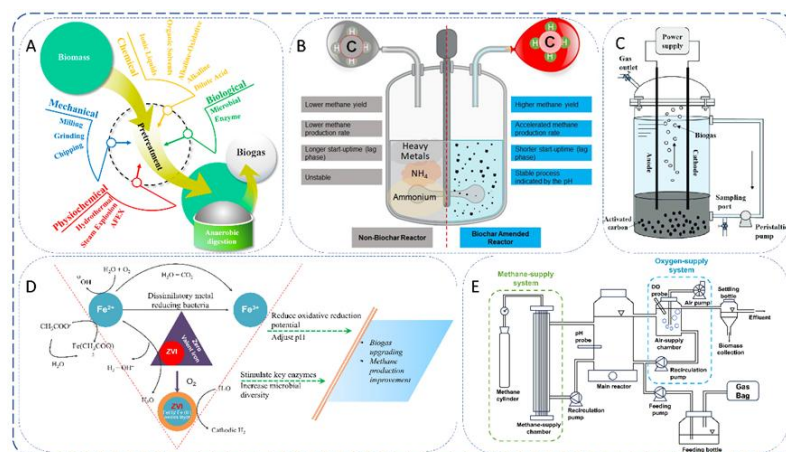


Fig. 2: Mechanisms of action of technologies optimised for anaerobic fermentation (A) Schematic diagram of enhanced methane production by pretreatment of lignocellulosic wastes. Reproduced from^[90] (B) Addition of biochar. Reproduced from^[91] (C) Electro-generation of methane in the AD-MEC system. Reproduced from^[92] (D) Mechanism of nanoparticles to enhance anaerobic digestion. Reproduced from^[93] (E) Bioreactor for producing SCP. Reproduced from^[89]

Table 1: The impact of pretreatment on biogas and/or methane yield.

Substrates	Pretreatment	Methane yield	Biogas yield	Ref.
Corn stover	5%NaOH (w/w)	NA	372.4 L/kg	[94]
Date palm waste	1-ethyl-3-methylimidazolium acetate	321.67 mL CH ₄ /g TS	NA	[95]
Fallen leaves	3.5%NaOH (w/w)	81.8 L/kg VS	NA	[96]
Air-dried wheat straw	0.6 g/g VS CaO and 0.4 g/g VS ZVI	154.1 ± 7.5 mL/g VS	NA	[97]
Waste activated sludge	Cation exchange resin (CER)	123.7 ± 0.8 mL CH ₄ /g VS	NA	[98]
Sludge	Stirred and heated at 70 °C for 9 h	248 mL /g VSS _{in}	NA	[99]
Sludge	pH 9.5 for 24 h	282.5 ± 14.1 mL/g VSS	NA	[100]
Sludge and vegetable wastes	Grinding substrate	141 mL/g VS	NA	[101]
Food waste and cattle manure	NA	317 mL/g VS	NA	[102]
Food waste	High voltage pulse discharge	315 mL/g	NA	[103]
Lipid waste	Ultrasound	927.97 mL/g TVS	NA	[104]
Paper waste	Microbial consortium for 4 days	404 mL/g VS	221 mL/g VS	[105]
MSW	Hydrothermal liquefaction	284.351 mL/g VS	394 mL/g VS	[106]
Cardboard and chicken manure	NA	319.62 mLCH ₄ /g VS	NA	[107]
MSW and orange peel waste	NA	432.39 m/g VS	NA	[108]
Chicken manure	Extraction with water	NA	527.8 m ³ /Mg VS	[109]
Raw cattle manure	6% NaOH at 55°C	About 170 mL CH ₄ /g VS	NA	[110]
Dairy manure and food waste	NA	311 L/kg VS	531 L/kg VS	[111]
Fish waste and waste-activated sludge	NA	683.8 mL CH ₄ /g VS	NA	[112]

amino acids, which surpasses most reported values for methane-based SCP and exemplifies a dual-value strategy that enhances waste valorization by transforming low-energy-density biogas into nutritionally dense biomass and concurrently addresses global food insecurity through land-independent protein production.^[89] The reactor configuration enabling this conversion features a partitioned gas-delivery system and suspended-growth design: as illustrated in Fig. 2E, methane is pressurized in hollow fiber membranes of 280 μm diameter, facilitating diffusion without bubble formation to maximize gas-liquid transfer efficiency with over 95% CH₄ utilization, while oxygen-saturated medium is separately supplied to the main reactor to support optimal methanotrophic consortia growth in suspended form, ensuring facile biomass harvesting.^[89]

4.2 Fermentation

4.2.1 Bioethanol fermentation

Biomass waste is conventionally converted into bioethanol through two principal bioconversion pathways, namely enzymatic saccharification and microbial fermentation.^[113] Current research emphases centre on bioethanol production from cellulose and lignin hydrolysates,^[114] utilizing diverse substrates including forestry residues (leaves, bark, sawdust, harvest remnants), agricultural byproducts such as cereal straw and sugarcane bagasse, municipal organic waste streams,^[115] and pulp processing residues.^[116] The inherent recalcitrance of lignocellulosic biomass originates from its heterogeneous macromolecular architecture and complex chemical matrices.^[117] Advanced pretreatment strategies enhance cellulose/hemicellulose exposure to hydrolytic enzymes by modifying substrate porosity and crystallinity.^[118] A representative study employing tetrahydrofuran-water co-

solvent pretreatment achieved 83.2–100% cellulose conversion in crop residues through selective lignin extraction, with enzymatic hydrolysis generating glucose yields of 234.9–274.0 mg/g feedstock.^[119]

The effectiveness of pretreatment parameters exerts dominant control over hemicellulose hydrolysis, facilitating near-quantitative conversion to monomeric sugars. Simultaneously, a minor proportion of cellulose undergoes glucose generation. However, a significant carbohydrate fraction persists in non-hydrolyzed forms, necessitating iterative saccharification for complete conversion.^[115] Chemical-based hydrolysis faces diminishing competitiveness due to high reagent costs and byproduct formation (e.g., furfural), which often requires additional purification.^[120] In contrast, enzymatic hydrolysis exhibits superior operational merits including minimal toxicity, cost-effectiveness, absence of process inhibitors, and reduced equipment corrosion relative to acid/alkali approaches.^[121] Critical enzymatic components comprise cellulases specifically endo-β-1,4-glucanases, exoglucanases, and β-glucosidases that synergistically depolymerize cellulose into glucose,^[122] coupled with xylanases that randomly cleave β-1,4-glycosidic bonds in xylan matrix of hemicellulose to release xylose and xylobiose.^[123]

After pretreatment and hydrolysis, cellulose and hemicellulose depolymerize to produce monosaccharides.^[115] Yeasts (e.g. *Saccharomyces cerevisiae* and *Picrospermum*), fungi (e.g. *Rhizoctonia*, *Trichoderma* and *Trichoderma rezigogenes*) and bacteria (e.g. *Escherichia coli* and *Aeromonas fermentans*) have been reported to be used in the fermentation of hydrolysates.^[120] Monosaccharides are fermented by these microorganisms and then distilled to produce alcohol and carbon dioxide.^[122,124] Currently, bioethanol production faces

challenges of economic cost, conversion efficiency, inhibitors and toxic chemicals.^[125] Based on these challenges, a number of optimization techniques have been developed, including advanced pretreatment techniques, the addition of catalysts, genetic engineering and production process optimization.^[126-128] Fig. 3 illustrates the mechanism of action of some of the optimization techniques. The bioethanol production process (Fig. 3A) encompasses four configurations, namely consolidated bioprocessing (CBP), combined saccharification and co-fermentation (SSCF), integrated saccharification-fermentation (SSF), and separate hydrolysis and fermentation (SHF).^[129] Given the limitations of SHF including high operational costs, contamination risks, and inhibition challenges,^[130] the integrated approaches of SSF/SSCF/CBP have been adopted for industrial-scale bioethanol synthesis owing to reduced capital expenditure, enhanced productivity, and operational simplicity.^[131] In addition, the use of additives is also a means of enhancing bioethanol production. It was found that the gas-liquid interface in the enzymatic reactor leads to cellulase inactivation due to shear force and cellulase unfolding (Fig. 3B). The incorporation of surfactant PEG4600 is noteworthy due to its structural attributes. At the two-phase interface, PEG4600 poly exhibits a competitive interaction with cellulase, effectively reducing the amount of cellulase present at the interface. This mechanism consequently diminishes the inactivation of cellulase, enhancing its operational efficacy.^[132] Chitosan-functionalized magnetic nanoparticles applied to pretreated corn cob substrates achieved a glucose output of 21.84 g/L (64.45% conversion rate), validating their saccharification capacity.^[133] Specifically, Fig. 3C details the bioethanol production enhancement process using magnetic nanoparticle (MNP) immobilized enzymes. The sequence initiates with lignocellulosic biomass progressing to enzymes immobilized

on MNPs. Subsequent mixing with biomass triggers hydrolysis where cellulose is enzymatically degraded into fermentable sugars. A magnetic field is then applied to separate immobilized enzymes from hydrolysate, simultaneously enabling enzyme recovery and ethanol output. This approach exploits MNPs' superparamagnetism for rapid enzyme retrieval, substantially lowering operational costs in lignocellulosic bioethanol processing. Additionally, Fig. 3D illustrates Systems Metabolic Engineering integrating four key approaches: Traditional Metabolic Engineering displays the glucose-to-ethanol metabolic pathway with intermediates Glucose-6-P, PYRUVATE, and Acetyl-CoA alongside energy carriers ATP/NADH; Adaptive Laboratory Evolution shows comparative growth curves of wild-type versus adapted strains over time; Synthetic Biology features a circular genome structure highlighting genetic modification elements; Systems Biology and Computational Modeling integrates computer analysis with molecular network diagrams. These components collectively enable targeted bioengineering of microbial systems, alleviating ethanol stress in microorganisms to enhance bioproduction efficiency.^[134]

4.2.2 Bio-hydrogen fermentation

Biohydrogen generation via biomass fermentation demonstrates superior efficacy compared to traditional H₂ synthesis methods including steam methane reforming and electrolytic water splitting.^[138] Consequently, biohydrogen provides a viable means of supplying low-pollution, high-efficiency, sustainable hydrogen and is therefore considered a promising methods of hydrogen production.^[139] This bioprocess has been extensively implemented to valorize lignocellulosic feedstocks alongside food industry byproducts and urban organic waste streams.^[140,141]

Dark fermentative (DF) production of biohydrogen is done

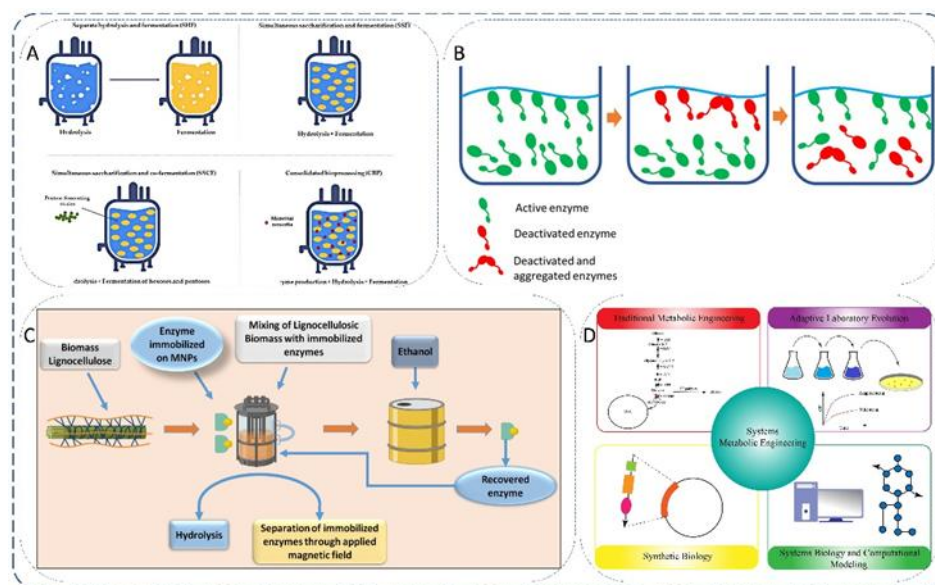


Fig. 3: Techniques and mechanisms to optimize the bioethanol conversion process (A) Fermentation strategies used to optimize the process. Reproduced from^[125] (B) Enzyme inactivation. Reproduced from^[135] (C) Enzyme immobilization on magnetic nanomaterials. Reproduced from^[136] (D) Various technologies applied to improve ethanol tolerance in yeast cells. Reproduced from^[137]

by dark fermenting bacteria, which can be classified as either specialized anaerobes or facultative anaerobes based on their ability to grow in the presence of oxygen.^[140] *Clostridium*, *Ethanolbacter* and *Vibrio desulfuricans* are referred to as specialized anaerobes, while *Bacillus*, *Escherichia coli*, *Klebsiella*, *Citrobacter* and *Enterobacter* are referred to as parthenogenetic anaerobes. Almost 70% of hydrogen production is studied by anaerobic bacteria of the genus *Clostridium*.^[142] After pretreatment, microorganisms utilize starch, glucose, and lignocellulose as substrates to synthesize pyruvate through the glycolytic pathway. Pyruvate subsequently enters the acid-producing pathway, facilitating the production of biohydrogen. While dark fermentation (DF) represents an efficient method for hydrogen production, the metabolic activity of fermentative microorganisms in DF systems can be adversely affected by various factors. These include the presence of additional substrates and nutrients, acidic pH levels, elevated temperatures, metal ions, competing microorganisms, toxic substances derived from substrates, and undissociated organic acids.^[143] Nanotechnology is now widely applied to enhance hydrogen generation in DF. The inherent characteristics of nanoparticles make them effective in this process, improving the performance of microbes that produce hydrogen.^[144] A 41% enhancement in H₂ production capacity was observed in DF systems incorporating 200 mg/L iron oxide nanoparticles synthesized from olive leaf extracts.^[145] Supplementation with metallic cations enhances biohydrogen synthesis during DF. Taherdanak *et al.* demonstrated that Ni²⁺ ions induced a statistically significant ($p < 0.01$) elevation in H₂ yield, achieving a 55% productivity surge.^[146]

The process of photo-fermentative biohydrogen production has been shown to generate hydrogen from small molecules of fatty acids and alcohols through the action of nitrogen-fixing enzymes of photosynthetic bacteria such as *Rhodobacter sphaeroides* and *Pseudomonas sphaeroides*.^[147,148] From an

economic perspective, hydrogen production through deep fermentation is more advantageous and profitable than the photo-fermentation process because it produces hydrogen continuously and is not dependent on the energy provided by sunlight.^[149] To further enhance efficiency, adding nanoparticles in the biohydrogen-production method (Fig. 4A) enhances hydrogen generation by activating hydrogenase activity and accelerating electron transfer, thereby optimizing metabolic flux toward H₂ formation. Researchers utilized rice straw as the substrate and supplemented the process with iron (Fe) and molybdenum (Mo) as biocatalysts to facilitate electron transfer. This approach resulted in a notable increase in hydrogen production, reaching up to 2.15 mM/h.^[150] Research indicates that integrating DF effluents into photo-fermentative processes represents a viable strategy to amplify biohydrogen output. As the integrated bioreactor (Fig. 4B) employs a horizontally-elongated rectangular container design. A membrane separation medium divides the system into the dark chamber positioned on the left and the photo chamber on the right. The dark chamber monitors initial hydrogen production via a left-mounted gas flow meter, while the final gas output is quantified by a right-positioned gas flow meter in the photo chamber.

The photo chamber features an external light source atop to supply photosynthetic energy, with a heating magnetic stirrer at the base maintaining constant temperature and homogeneous mixing. Metabolic complementarity is enhanced by increasing the photo chamber volume ratio. Metabolites generated during DF can serve as feedstocks for subsequent photobiological conversion, establishing a cascading biorefinery model.^[142] For instance, Özgür *et al.* achieved sucrose-to-hydrogen conversion efficiencies of 4.2 mol H₂/mol sucrose via DF, which escalated to 13.7 mol H₂/mol sucrose through sequential DF-photofermentation integration.^[151]

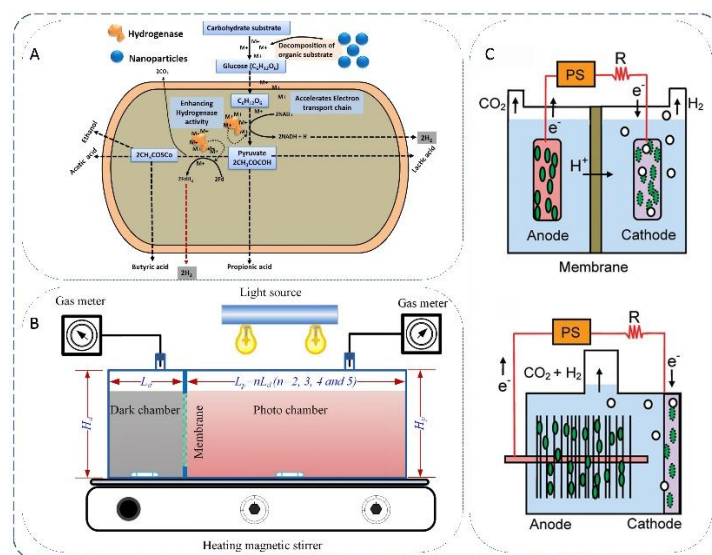


Fig. 4: Technological means to enhance biohydrogen fermentation (A) Nanoparticles to promote biohydrogen production. Reproduced from^[155] (B) Integration of dark and photo-fermentation. Reproduced from^[156] (C) Schematic diagrams of dual-chamber (planar anode) and single-chamber membrane-free (brushed anode) MECs. Reproduced from^[157]

Biohydrogen production extends beyond fermentative methods to include biophotolysis (direct and indirect) and microbial electrolysis.^[152] Bio-photolysis refers to the process by which photosynthetic organisms decompose water in the presence of light. A significant advantage of this process is that it does not necessitate any additional nutrients, as sunlight, carbon dioxide, and water serve as the fundamental raw materials required.^[153] Alternatively, Hydrogen production can be achieved through the process of electrohydrogenation within a microbial electrolysis cell (MEC). As shown in Fig. 4C, two electrochemical systems are vertically arranged. In the upper section, CO₂ oxidation at the anode releases electrons flowing rightward to the cathode for H₂/O₂ production, while protons migrate through the membrane. The lower section demonstrates reversed electron flow from power source oxidation to reduce resistor, maintaining proton transmembrane movement. This method demonstrates a higher yield compared to traditional fermentation methods and offers improved energy efficiency relative to water electrolysis.^[154]

4.2.3 Compost

Composting serves to stabilize biomass waste while enhancing the bioavailability of heavy metals and reducing pathogen populations. Humus-enriched compost, once stabilized, proves to be a highly effective phytostimulant agent.^[13] Depending on the methodology employed in the recycling of organic solid waste, composting can be categorized into various technologies, including aerobic composting and vermicomposting.^[158] Organic waste suitable for use as fertilizers can be classified into several categories, namely municipal waste, food waste and plant-based compost, as well as animal organic waste.^[159,160]

Aerobic composting represents a biochemical process designed to stabilize organic waste and render it harmless.^[161] This process unfolds through three distinct stages: a primary thermophilic phase, followed by a secondary thermophilic interval, and concluding with a stabilization phase. Each stage is governed by specific thermal ranges that facilitate transitions between these phases. Critical to the success of this process are microbial communities, including bacteria, actinomycetes, and fungi, which are prevalent in natural ecosystems.^[162] These microbial communities operate effectively under varying temperature conditions to break down biomass waste into nutrient-rich compounds. The resulting mature compost forms a stabilized humic matrix that significantly enhances soil fertility by improving its physicochemical properties.^[163] Compost quality is influenced by ambient temperature, feedstock type, CN ratio, oxygen concentration, and additives.^[164] The current problems that need to be urgently addressed in aerobic composting technology are char loss,^[165] long composting time and odour gas emission.^[166] Effective means of improvement have been developed in commerce, such as the addition of microbial inoculum, aeration (forced aeration, natural convection and physical mixing of substances) and the addition of biochar.^[167,168]

Bioaugmentation represents a strategic approach to regulating the functional pathways of decomposer communities within the composting process.^[169,170] This method has been demonstrated to mitigate unpleasant odours significantly, elevate temperatures, shorten processing duration, and enhance humification efficiency. Through the inoculation of beneficial microorganisms, it is possible to establish an advantageous microbiota, improve microbial quality, increase overall microbial populations, and develop specialized enzymatic systems.^[171] A notable study revealed that the co-inoculation of maize stover, chicken manure, and mushroom residues with *Xylaria* spp. and *Bacillus* spp. resulted in a greater abundance of *Mycobacterium* spp. and facilitated an earlier maturation of compost by seven days.^[172]

For decomposition, mineralization and value addition to biomass waste, vermicomposting is the preferred management method. This technology uses the activity of earthworms and micro-organisms in the decomposition process to produce nutrient-rich, enzyme- and micro-organism-rich organic fertilizers.^[173] Earthworms serve in processing diverse organic residues, including horticultural and vegetable by-products, mushroom residues, digestate, rice husks, municipal solid waste, and mixtures of various biomass wastes.^[174] The types of earthworms used in vermicomposting can be classified by their habitat, namely epigeic, endogeic and anecic. *E. fetida* and *E. andrei* represent the dominant species in vermiculture, globally distributed with exceptional environmental adaptability.^[163] For optimal valorization of organic substrates into humus-rich amendments, integrated phases encompassing microbial decomposition, earthworm-mediated bioturbation, feedstock assimilation, and maturation require systematic implementation.^[175] Following microbial initiation of enzymatic degradation within the matrix, earthworms exert synergistic effects on the physical-biochemical attributes of the soil via specialized physiological processes. As they tunnel and burrow, these creatures not only extrude the substrate but also significantly increase its specific surface area and oxygen levels. This alteration in the environment creates a more favorable habitat for a diverse range of microbial communities, facilitating their succession. Additionally, earthworm-microbe synergism enhances organic waste conversion via gut microbial decomposition (Fig. 5A), where extracellular enzymes break down macromolecules. Excreted nutrient-rich vermicompost, characterized by porous structure and bioactivity, delivers carbon/nitrogen/trace elements to environmental microorganisms, establishing a closed loop. This collaboration accelerates mineralization, reduces heavy metal bioavailability, and mitigates greenhouse gas emissions, enabling efficient resource recovery.^[176-178]

In the process of vermicomposting, earthworms play a crucial role not only in shaping the microbial community but also in producing mucus that contains carbohydrates and proteins. This mucus contributes to the moisture retention associated with earthworm movement while also exerting regulatory effects on the decomposition of organic matter and

limited, this presents both a challenge and a significant opportunity for advancing composting technology in the future.

4.2.4 Oleaginous microorganisms fermentation

Microbial-derived lipids (unicellular oil bodies) can be biosynthesized by oleaginous microorganisms utilizing diverse organic substrates, including glycerol, volatile fatty acids (VFAs), and agro-residue-derived saccharines.^[193] Oleaginous species are defined as microorganisms that possess the ability to store intracellular lipids, constituting 20% or more of their dry biomass weight. This category includes microalgae, bacteria, fungi, and yeasts. The lipid reserves within these organisms predominantly consist of triacylglycerols (TAGs), which are essential as primary substrates for the advanced synthesis of biofuels.^[194] Oleaginous yeasts and bacteria exhibit superior lipid productivity compared to algae. Current taxonomic records identify over 160 yeast species with lipid-accumulating capacity, including industrially relevant strains such as *Y. lipolytica*, *T. oleaginosus*, *R. toruloides*, *L. starkeyi*, and *R. glutinis*.^[195]

The key technological processes involved in the synthesis of biobased diesel through microbial lipids consist of several sequential stages. These include the harvesting of oil-rich microorganisms, the extraction of intracellular lipids, the catalyzed conversion of these lipids, and the subsequent refinement of the resulting crude biodiesel.^[196] Biomass wastes used for lipid production can be classified into two categories, hydrophilic and hydrophobic substrates, based on the way oleaginous micro-organisms synthesize lipids. Hydrophobic matrices consist of industrial fats and hydrophobic food wastes, while hydrophilic organic wastes consist of plant wastes, fruit wastes, molasses, sewage sludge, glycerol and paper mill wastes from biodiesel production and soap manufacturing.^[193] The structural characteristics of the two substrates define distinct lipogenic pathways. Oleaginous microorganisms possess the capability to absorb hydrophobic substrates, such as triglycerides, oils, alkanes, and fatty acids derived from a variety of sources. Following this absorption, these microorganisms synthesize lipid molecules through a diverse array of metabolic pathways. Empirical studies have demonstrated the successful conversion of dairy effluent by *Yarrowia lipolytica*, resulting in a lipid productivity of 32% (13.53 g/L) and yielding 61% biodiesel through catalytic transesterification.^[197] Secondly, oleaginous microorganisms utilize hydrophilic substrates (alcohols, saccharide compounds) as metabolic precursors for lipogenesis.^[193] In one study, crude glycerol and lignocellulosic biomass were used to prepare single-cell oils via *Cutaneotrichosporon oleaginosum*, yielding 75.7% lipid content and 61.1 g/L titer.^[198]

A range of process conditions and external factors influences lipid production in oleaginous microorganisms. Notably, when hydrophobic substrates are utilized, lipid accumulation appears to be unaffected by nitrogen

exhaustion.^[199] However, when hydrophilic substrates are used under nitrogen-deficient culture conditions, oil-containing microorganisms accumulate lipids in large quantities.^[200] Synergistic cultivation strategies have been shown to significantly enhance lipid biosynthesis, as evidenced by the use of microalgal yeast consortia, specifically *Chlorella vulgaris* var. in conjunction with *R. glutinis* TISTR 5159. This cross-kingdom interaction yielded a noteworthy biomass productivity of 4.63 ± 0.15 g/L and lipid yields reaching 2.88 ± 0.16 g/L.^[201] The genetic manipulation of *T. dermatis* was achieved through a combination of chemical mutagenesis and sequential plasma treatment, resulting in the development of the hyperproductive mutant strain L7. This strain has been optimized for lignocellulosic conversion utilizing sugarcane bagasse. Experimental validation has demonstrated that this mutant significantly enhances fermentation kinetics, reducing the process time by 24 h, while also increasing both lipid titer and biomass accumulation.^[202] The industrial-scale application of microbial lipid technology necessitates not only innovations in genetic engineering and fermentation kinetics but also robust policy frameworks and cross-sector collaboration. For instance, Mexico City's Quintuple Helix Model integrates government, academia, and industry to deploy patented IPN-GBD-1000 technology, efficiently converting waste cooking oil into biodiesel. This approach significantly reduces urban waste and enhances energy security. Concurrently, the United States leverages Renewable Fuel Standard (RFS) and Low Carbon Fuel Standard (LCFS) policies to mandate biofuel blending, prioritizing low-carbon feedstocks like used cooking oil. This finding validates the synergistic dynamics of policy frameworks in driving the scaled production of microbial lipids, thereby reinforcing their pivotal nexus within sustainable energy transitions.^[203,204]

5. Utilization of biomass waste bioconversion products

Biomass waste is converted to generate diverse value-added commodities advancing ecological preservation, economic viability, and societal needs, categorized into fertilizers, soil amendments, feeds, and renewable energy carriers. The extended application of synthetic fertilizers containing potassium, phosphorus, and nitrogen has triggered substantial ecological disturbances due to their global agricultural prevalence.^[160] Bio-composting delivers an eco-sustainable methodology for transforming waste streams into humic-rich amendments via indigenous microbial consortia and enzymatic catalysis.^[169,205] Digestate and vermicompost liquors effectively degrade organic pesticides and pharmaceutical residues in contaminated soils,^[175] functioning as pedological enhancers while augmenting fertility. Biomass resources present significant potential for addressing the increasing demand for alternative sources of animal protein. The conversion of these bioresources into entomoprotein provides scalable solutions for food and feed applications. Additionally, the utilization of residual insect frass as organic amendments contributes to sustainable agricultural practices.^[28] Black

soldier fly-mediated bioconversion yields pupae containing $\approx 40\%$ protein ($\approx 40\%$) and lipids ($\approx 30\%$), demonstrating viability as swine and aquaculture nutritional supplements to replace conventional aquatic-derived protein sources.^[206]

Biofuels come in three different states: solid (biochar), liquid and gas.^[207] Among them, liquid and gaseous forms of biofuels can be derived from bioconversion, including biodiesel, bioethanol, biohydrogen and biogas.^[208] Biodiesel is recognized as a sustainable combustion fuel and serves as a viable alternative to petroleum-based diesel, requiring no modifications to existing engines for implementation. The utilization of biodiesel has been shown to significantly reduce emissions of CO₂, CO, hydrocarbons, and particulates when compared to conventional diesel.^[208] Lipids derived from biomass waste facilitate a variety of applications, including the production of oleochemicals, cosmetics, textiles, coatings, lubricants, and bio-polyurethane foams.^[193] Ethanol produced through the bioconversion of biomass waste results in lower carbon monoxide emissions, estimated at 12–15%, making it an environmentally sustainable option.^[209] With a composition that includes 34.7% oxygen, which contrasts with the oxygen-free nature of gasoline, bioethanol improves oxidative combustion efficiency by approximately 15%, thereby reducing both particulate matter and nitrogen oxide (NO_x) emissions. Furthermore, its minimal detectable sulfur content permits the implementation of blending protocols that significantly lower fuel sulfur levels. This in turn plays a crucial role in mitigating the impacts of acid rain by minimising sulfur oxide emissions.^[210]

Biohydrogen is a promising renewable fuel with the capacity to replace fossil fuels.^[153] As an efficient energy carrier, it supports energy storage from variable renewable sources, significantly reduces greenhouse gas emissions, and serves as a valuable chemical feedstock. Additionally, biohydrogen can be used both as a direct fuel in internal combustion engines and for powering fuel cells.^[211,212] It is a key driver in transitioning from fossil fuels to a bio-based economy. Biogas serves as a sustainable alternative to liquefied petroleum gas (LPG) and compressed natural gas (CNG), providing 30% efficiency in power generation when compressed, while its slurry byproduct acts as an effective nutrient-rich fertilizer.^[183]

In conclusion, the conversion of biomass waste into value-added products presents a strategic approach to harmonise environmental sustainability with economic and social goals. By converting residual organic materials into humic-rich soil amendments, bio-fertilizers, and entomoproteins, as well as utilizing lipid- and carbohydrate-derived intermediates for biofuels, oleochemicals, and speciality materials, these processes effectively close nutrient loops and reduce reliance on fossil-derived inputs in agriculture, aquaculture, and energy sectors. Importantly, integrated approaches that combine anaerobic digestion, insect-based valorization, and thermochemical upgrading can optimize resource recovery while simultaneously decreasing greenhouse gas emissions

and pollutant loads. To fully exploit this potential, future efforts should concentrate on enhancing microbial and enzymatic catalysts, developing modular biorefinery platforms that can process diverse feedstocks, and conducting thorough techno-economic and life-cycle assessments. Advancements in these areas will be crucial for the large-scale implementation of biomass waste bioconversion within circular bioeconomies.

6. Conclusion and outlook

This review examines the role of microorganisms and animals through anaerobic digestion, fermentation, and composting in converting biomass wastes from agriculture, forestry, and industry into products of higher value for the use and development of human society and the natural environment. The above description shows that bioconversion is a greener way of converting biomass with a lower economic burden and more sustainable development. Nevertheless, the field confronts multidimensional challenges: efficiency bottlenecks (slow pretreatment kinetics, hydrolytic delays), process instability (VFA/ammonia inhibition, enzyme deactivation), product defects (sulfur-contaminated biogas, low-titer ethanol), environmental risks (carbon loss, heavy metal enrichment), and systemic contradictions (technology integration barriers, feedstock heterogeneity). The elevated costs and inhibitory residues of physicochemical pretreatment not only contradict bioconversion's economic advantages but also exacerbate fermentation constraints, while feedstock variability coupled with economic viability limitations fundamentally impedes scaling. Addressing these requires dual-track advancement: intensifying biological pretreatment research to enhance efficacy without cost inflation, and expanding diversity in catalyst/microbial/zoological inoculant development to boost high-value product synthesis capacity—though current additive formulations lack diversity, necessitating deeper scalability investigations. Ultimately, integrated physico-chemical-biological technologies and multi-substrate systems will unlock versatile valorization pathways, complemented by strategic innovations including AI-driven process control, waste-derived nanocatalysts, gene-edited strains, and modular circular biorefineries—all synchronized with policy incentives to catalyze a paradigm shift from linear waste disposal to circular bioeconomy.

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Conflict of Interest

The authors declare that they have no conflict of interest.

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