

1 General Introduction

1.1 Bioeconomy

Unlimited growth is incompatible with the fundamental laws of nature.¹

The current linear economy strictly depends on coal, oil, and natural gas for the production of goods, the generation of energy, and transportation. The exploitation of these reserves has led to great improvements in the quality of life for billions of individuals since the industrial revolution. Behind the current form of resource utilization, there is an “take, make, use, dispose” mindset, where disposal is the end of life of every product.² However, the usage of fossil resources causes massive emission of CO₂ and other greenhouse gases (GHGs). This has given rise to the climate crisis, with all its disastrous consequences on the environment and human health, including extreme weather events, insecurities in food supply, and land shortage.³⁻⁷ Additionally to greenhouse gas emissions and the resulting climate crisis, the linear economy massively endangers the biosphere, referred to as the “biosphere crisis”. The ongoing destruction of key ecosystems, for example by deforestation, and the resulting loss in biodiversity, also endangers human survival.^{8, 9}

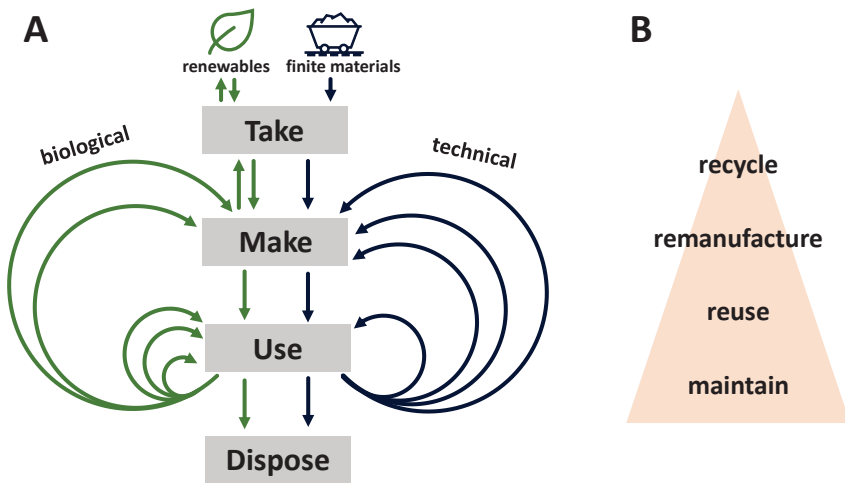


Figure 1 Visualization of the circular economy. (A) Technical and biological utilization of renewables and finite materials in the circular economy and the linear economy. In the linear economy, materials undergo the “take, make, use, dispose” lifeline, while in a circular economy, materials fluctuate between “make” and “use” by maintaining, reusing, remanufacturing, and recycling. This scheme is an adapted version of the iconic “Butterfly Diagram” of the Ellen MacArthur Foundation.¹⁰ (B) Hierarchy of the options to incorporate materials and goods in a circular economy. Maintaining a material usually requires the least energy, thus, it is the preferred option. If maintenance is not possible, the material should be reused, then remanufactured. Recycling of a material should only be done if it is unavoidable.¹¹

To limit the effects of the climate crisis and the biodiversity crisis, it is evident that humanity must transition from the current linear economy to a circular economy. Accordingly, we have to phase out fossil fuels, and treat waste as a valuable resource. The basis of the circular economy is closed material loops, which implies the minimization of waste generation and the preservation of the value of products, materials, and resources (**Figure 1A**). Notably, recycling a material usually requires more energy than maintaining or reusing it. Thus, recycling is not the preferred option for the preservation of materials in a circular economy (**Figure 1B**). Still, recycling is often necessary and a valid method in this context.^{12, 13}

One key player in the circular economy is the so called bioeconomy, where lithospheric inputs such as fossils are substituted with biogenic inputs such as biomass in economic activities.^{14, 15} In the envisaged bioeconomy, renewable carbon reserves such as agricultural biomass, organic waste, and industrial side streams (e.g. CO₂, plastics), are used to generate material, chemicals, fuels, and energy (**Figure 2**).^{16, 17}

Circular (bio)economies are not a new concept; in fact, circular practices are deeply rooted in human history. A mindset of reusing, remanufacturing, and recycling of materials was found to be the foundation of, e.g., ancient Indian cultures, making the circularity concept one of the oldest one known to humanity.^{18, 19}

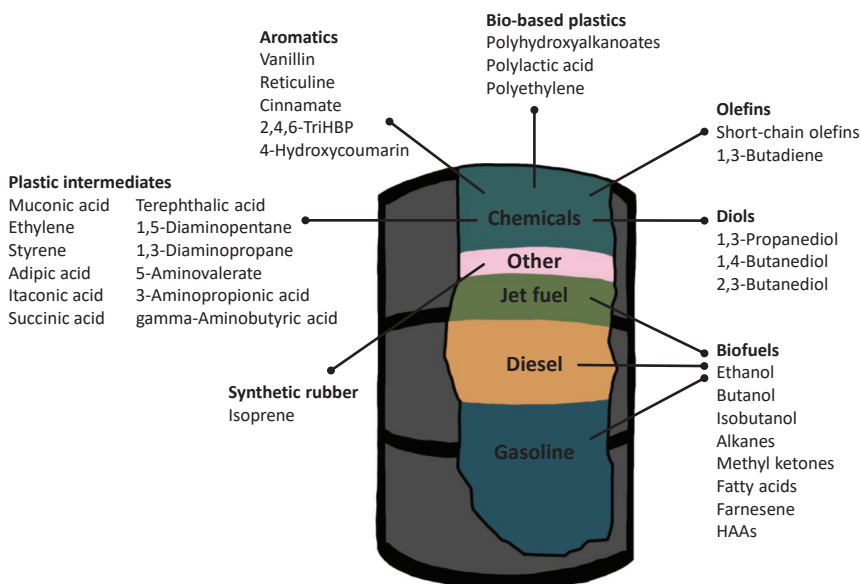


Figure 2 Replacing the oil barrel. Possibilities of a more sustainable production for various chemicals, with different degrees of commercialization,²⁰ 2,4,6-TriHBP = 2,4,6-trihydroxybenzophenone; HAAs = 3-(3-hydroxy-alkanoyloxy)alkanoates.^{21, 22}

It should also be noted that a bioeconomy is not inherently sustainable or “circular”. The bio-based production of goods does not necessarily imply its re-use or recycling and the general avoidance of over-consumption.^{23, 24} Also an increased pressure on water supply and natural ecosystems for the generation of biomass are part of the risks of a bioeconomy.^{14, 25}

However, with the techniques of the life sciences and biotechnology, modern or “knowledge based” bioeconomy can be transformed into one of the most advanced and sustainable economic concepts, securing the prosperity of humanity.²⁶

1.1.1 Biotechnology within a sustainable bioeconomy

The 21st century was claimed to be the “Century of Biology”.²⁷ The application of biological methods in the production of goods is referred to as industrial biotechnology, and it plays a key role in the bioeconomy. In industrial biotechnology, biological systems such as microorganisms or enzymes are used as catalysts for the generation of value-added goods, preferably from renewable carbon sources. The application of living microorganisms in fermentation processes plays a key role in industrial biotechnology.^{28, 29} One remarkable feature of microorganisms is their capability to transform a broad variety of carbon sources, including those obtained from industrial side streams, biomass, and CO₂, into various products (Figure 3).³⁰

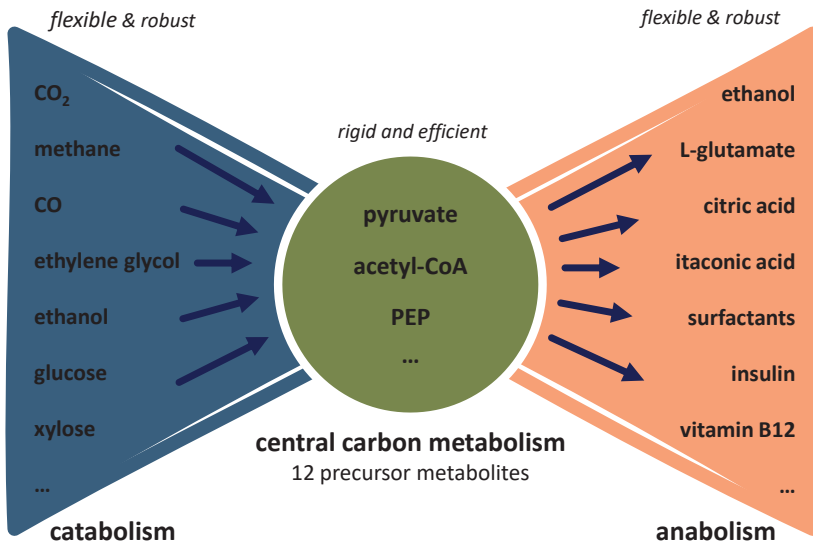


Figure 3 The bow-tie structure of microbial metabolism. Various substrates can be utilized in catabolic enzymatic reactions (left) to form the building blocks of the central carbon metabolism (center). Those central precursor metabolites can be converted to value-added goods in anabolic reactions (right), partly enabled by genetic engineering. The products are shown in declining order of their market size. Both catabolism and anabolism have a high variation and flexibility within species, while the central carbon metabolism is mostly conserved for efficiency. PEP = phosphoenolpyruvate.³¹⁻³⁴

The utilization of the metabolic networks in living cells has the benefit of highly stereoselective product formation and a high product purity. Additional advantages of industrial biotechnology include a lowered energy consumption, the use of non-toxic aqueous solvents, and decreased generation of waste, compared to purely chemical production.^{35, 36}

In ancient times, the fermentation of *e.g.* food products with non-adapted microbes was practiced as an inherently biotechnological process. Since the 70s, pioneering DNA-based technologies such as polymerase chain reaction (PCR), DNA sequencing, and, more recently, CRISPR Cas9, have been developed that turn microorganisms into “programmable biocatalysts”.³⁷ Currently, mRNA technologies and the utilization of artificial intelligence are being researched and open up new possibilities in the production of advanced chemicals and pharmaceuticals.³⁸⁻⁴¹ The possibility to tailor a cell’s genetic repertoire to specific applications transformed the entire field of industrial biotechnology and generally accelerated bioprocess development.^{42, 43} Using those optimized microorganisms as biocatalysts can benefit both the sustainability and the efficiency of existing processes, *e.g.* by allowing for higher product yields, and facilitate the development of new, revolutionary products, such as mRNA vaccines.^{20, 44}

Further examples of successful industrial implementation of biotechnological processes for fine and bulk chemicals include the production of citric acid, glycerol, lysine, xanthan, and insulin, to name a few.^{28, 45-47} Common microbial workhorses in these processes include well-known model organisms such as *Escherichia coli* and *Saccharomyces cerevisiae* as well as novel microorganisms such as *Ogataea polymorpha* or *Paracoccus pantotrophus* that can be exploited for their advanced properties.^{48, 49} Some features, *e.g.* a high robustness towards fluctuating conditions and a high genomic stability are critical for the application of microorganisms as cell factories in industrial processes and biorefineries.^{50, 51}

1.1.2 Biorefineries for biofuels and bulk chemicals

Refineries are facilities where different unit operations are combined to convert a raw material into fuels, electricity, heat, and chemicals. Traditionally, petrochemical refineries split crude oil into different fractions that are further processed into an immense variety of products that are the foundation of our society, fueling the linear economy.⁵² The unit operations of petrochemical refineries have been optimized for decades, so that most processes operate at the upper limit of capacity and efficiency.⁵³⁻⁵⁵ The product range of a petrochemical refinery includes platform chemicals and fuels, which form the basis of chemical and pharmaceutical industries.^{56, 57}

Just as a petrochemical refinery is the foundation of the linear economy, biorefineries are their counterpart in a circular bioeconomy. Biomass serves as the primary carbon source for biorefineries and results from the fixation of CO₂ from the atmosphere during photosynthesis, which creates a circular approach by nature.⁵⁸ First generation biorefineries are based on the edible parts of crops rich in starch, sugars, or oils (wheat, sugar cane, oil palm etc.). While these have the potential to reduce GHG emissions,

first generation biorefineries compete with food production.⁵⁹ Second generation biorefineries work with non-edible feedstocks such as lignocellulosic plant material or industrial side streams. By using these feedstocks, products can be produced sustainably, while waste accumulation can be reduced.

Lignocellulose in particular is an abundant resource that has the potential to replace fossils. It has a high carbohydrate content and is, unlike fossil resources, available fairly consistently throughout the world.⁶⁰ Additionally, the cultivation of dedicated energy crops with high biomass yields on marginal soils holds great potential.⁶¹

In a lignocellulosic refinery, biomass first undergoes pre-treatment to obtain the major components cellulose, hemicellulose, and lignin in an accessible form (**Figure 4**). These are subjected to different bio- and chemocatalytic conversion steps for the generation of a broad variety of products.⁶² While this concept holds great potential for a more sustainable future and some sites are already operating, the large-scale economic feasibility has to be improved.⁶³ The technologies for processing crude oil were developed since the 1860s, while bio-refineries still lack standardized methods and a uniform processing approach.⁶⁴ For example, it is unclear which pre-treatment method is best during the processing of different forms of lignocellulosic material.⁶⁵⁻⁶⁷ It is widely recognized that industrial biotechnology will play a key role in the conversion of lignocellulosic building blocks to fine and bulk products.^{20, 68} However, bioprocessing is currently often not as effective as chemical processing and often has low product titers, resulting in higher costs for products of renewable origin.^{69, 70}

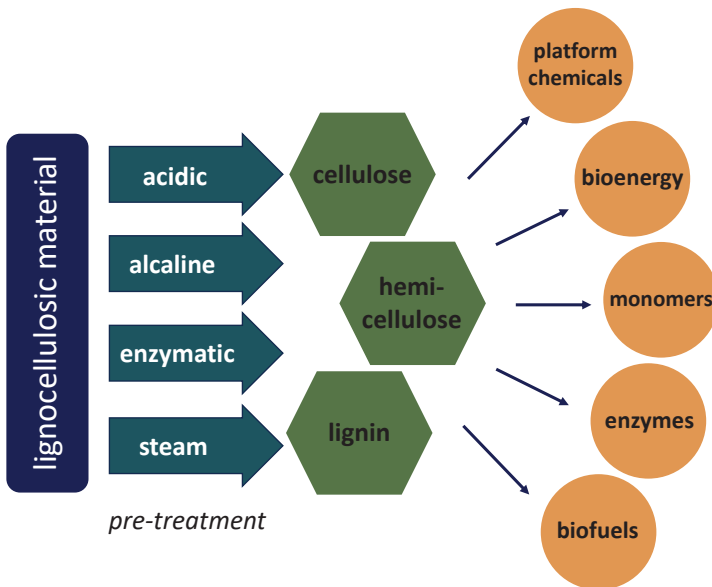


Figure 4 Scheme of a lignocellulosic biorefinery.⁶²