

Microbial Alchemy: Waste valorization for yeast oil production for a circular future

Monalisa Ghara, Sakshi Kumari, Rashi Jain, Harshita Mahar, Tanya Khanna, Christine Jeyaseelan, and Debarati Paul*

Amity University Uttar Pradesh, Noida, Sec 125, Uttar Pradesh, India

*Corresponding author: dpaul@amity.edu

Abstract

The growing demand for sustainable solutions has driven researchers to explore microbial solutions for oil production using waste feedstock, enabling its valorization via oleaginous yeast strains. Depending on species and cultivation conditions these microbes accumulate over 50% lipids of their total cellular biomass, by using various carbon sources from organic waste. This microbial-driven process not only reduces environmental burdens associated with waste disposal but also presents an eco-friendly alternative to conventional palm oil. By harnessing these microbes, industries can integrate circular economy principles, converting agricultural, industrial, and food waste into renewable lipid sources for biofuels, cosmetics, nutraceutical and food applications. This study explores various waste utilization approaches, the applications and challenges of yeast oils, and future prospects, contributing to a more resource-efficient and sustainable future.

Keywords: Oleaginous yeast, waste valorization, yeast oils, circular economy, sustainable future

1 Introduction

Yeasts are eukaryotic microorganisms found in diverse ecological niches such as water, soil, air, and particularly on plant and fruit surfaces where they play a direct role in the decomposition and natural fermentation. Nutritionally, yeasts are less demanding than other microorganisms, e.g., lactic acid bacteria, typically require only basic compounds such as fermentable sugars, amino acids, vitamins, minerals, and oxygen

for growth, whereas yeast use a wide range of carbon resources [1,2]. Morphologically, they exhibit a wide range of forms, commonly round, oval, or ellipsoidal. Microscopic examination is typically the first step in their identification, followed by microbiological and biochemical tests. Further classification involves more detailed assays like sugar fermentation and amino acid assimilation. Yeast species also vary in their production of lipid, ethanol, organic acids, which helps in identification and classification [1].

Metabolically, yeasts can ferment a wide variety of sugars, including glucose, fructose, sucrose, maltose, and maltotriose, commonly found in ripe fruits and processed grains. They also tolerate acidic conditions, withstanding pH levels as low as 3.5 [1,2]. For industrial use, yeasts are broadly categorized into *Saccharomyces* and non-*Saccharomyces* groups. *Saccharomyces cerevisiae*, in particular, is the most studied and widely used species in wine and beer production due to its robust fermentation capacity, fast growth, and adaptability [1, 3]. The oleaginous (oil accumulating) yeast strains are non-*Saccharomyces* strains, of immense industrial interest, because of their oil producing capability that is associated with their unique ability of metabolizing a wide range of carbon sources available in waste hydrolysates.

The global economy is currently dealing with two challenges that oleaginous yeast can overcome (i) The need for alternate renewable resource to suffice the needs of the increasing population because the energy obtained from fossil fuels, oil crops etc. cannot realistically satisfy even a small fraction of the existing demand. Exploring renewable energy



Graphical abstract

resources which do not compete with arable land has therefore become inevitable. (ii) With the ever-increasing global population and consumption patterns, effective waste management has become a complex and pressing challenge. Oleaginous yeast strains circumvent the above challenges by their remarkable ability to accumulate lipid globules and produce industrially important by-products, by feeding on a diverse range of waste feedstock, when cultivated in a biorefinery mode. Consequently, they have attracted interest in the biofuel, food, nutraceutical as well as the cosmetic industry amidst several challenges posed by cheaper conventional oil sources, and technical challenges owing to seasonal/regional variation in waste residues [1-3].

This review provides an overview of advances in cultivation of oleaginous yeast strains for oil production and consequent waste management. It also presents the

challenges faced by yeast oil production, their probable mitigation and future perspectives.

2. Yeast oil: industrial interest and challenges

Oleaginous yeasts accumulate lipids amounting to over 50% of their dry cell weight (DCW) in the form of intracellular lipid globules; certain strains such as *Yarrowia lipolytica*, *Rhodospiridium toruloides*, *Lipomyces starkeyi*, etc, achieve lipid concentrations as high as 60–70% when cultivated under carbon-rich and nutrient-deficient conditions [2-5]. Among other microbial oil producers, oleaginous yeasts stand out due to their ability to utilize a wide range of carbon sources from sugars and agro-industrial waste hydrolysates to alternative carbon sources such as glycerol, a by-product generated in large quantities during biodiesel production, or molasses produced by sugar industry [1-6] [Figure 1]. The yeast lipid is mainly composed of triacylglycerols (TAGs), which share a structural resemblance with plant-derived oils, making them well-suited for biodiesel production. TAGs can be efficiently transformed into fatty acid methyl esters (FAMES), the key constituents of biodiesel, through the conventional transesterification process [2, 5]. Microbial oil has garnered considerable interest as a promising feedstock to produce biofuels, oleochemicals, and food products as a replacement for palm oil [2-6]. The challenges to sustainable yeast oil production from waste feedstock are also many and need careful investigation, focussed research initiatives and industry initiatives.

The natural compositional variability of mixed waste residues poses a key challenge for scaling up lipid production processes. These waste materials, originating from diverse plant sources and varying across seasons, exhibit fluctuating nutrient profiles—such as differences in sugar levels, mineral content, and the presence of inhibitory compounds. This inconsistency can hinder the reproducibility and reliability of fermentation results in industrial settings [6, 7, 8]. However, this variability may also serve as an advantage. The heterogeneous composition of such waste

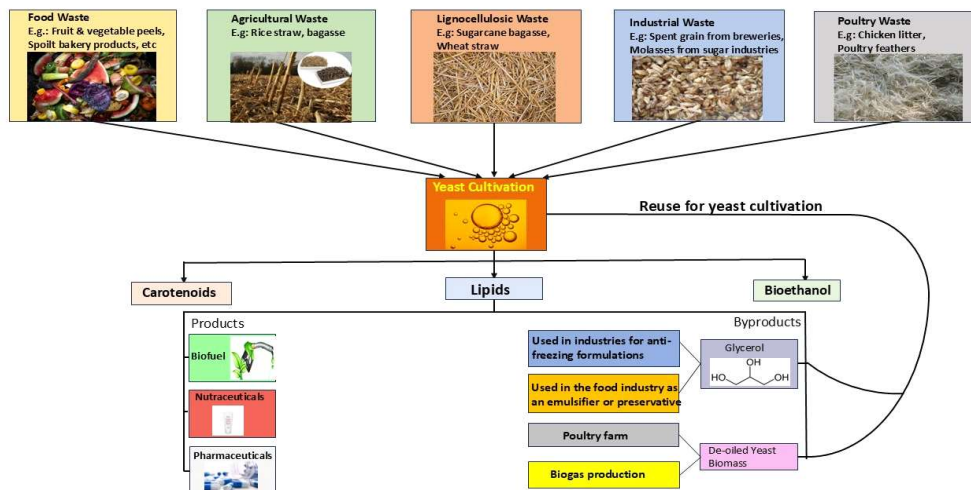


Fig. 1: Oleaginous yeast efficiently converts various waste streams into valuable lipids, promoting circular economy practices and consequently reducing environmental impacts due to unplanned waste generation.

hydrolysates can introduce a wider range of carbon sources and micronutrients, potentially boosting the metabolic adaptability of oleaginous yeasts such as *R. toruloides*. The metabolic flexibility enables the yeast to efficiently divert carbon into lipid synthesis, even when working with inconsistent or suboptimal feedstocks. As a result, *R. toruloides* stands out as a strong candidate for bioconversion applications using low-cost, variable waste streams [9-11] [Figure 2].

Numerous research groups have explored lipid production using oleaginous yeasts cultivated on hydrolysates obtained from lignocellulosic biomass [12]. However, these yeasts typically prioritize glucose consumption over xylose, with xylose uptake beginning only after glucose is exhausted, a phenomenon known as glucose repression [13, 14]. This sequential sugar utilization extends the overall conversion time and reduces the economic feasibility of the process. To address this, efforts have focused on promoting the simultaneous conversion of glucose and xylose into lipids. Despite these attempts, glucose repression remains a major

hurdle, and optimal co-conversion has yet to be realized. Therefore, identifying oleaginous yeasts that can metabolize both glucose and xylose concurrently is crucial for enhancing lipid production efficiency. Notably, *Lipomyces starkeyi* AS 2.1560 is one such yeast capable of co-utilizing glucose and xylose for lipid synthesis [15]. The ability to simultaneously utilize multiple sugars to accumulate lipids is of great importance when planning lipid production from lignocellulosic hydrolysates.

3. Oleaginous yeast strains: Waste valorization

Oleaginous yeast strains are increasingly being recognized for their ability to valorize waste, i.e., converting low-value or waste materials into high-value products, particularly lipids (fats and oils), which can be further used for sustainable production of biofuels, cosmetics, food additives, nutraceuticals, feed, etc [Table 1] [Figure 1].

3.1 Agricultural waste

Agricultural waste includes all organic residues generated from farming activities, such as crop residues. This can be segregated

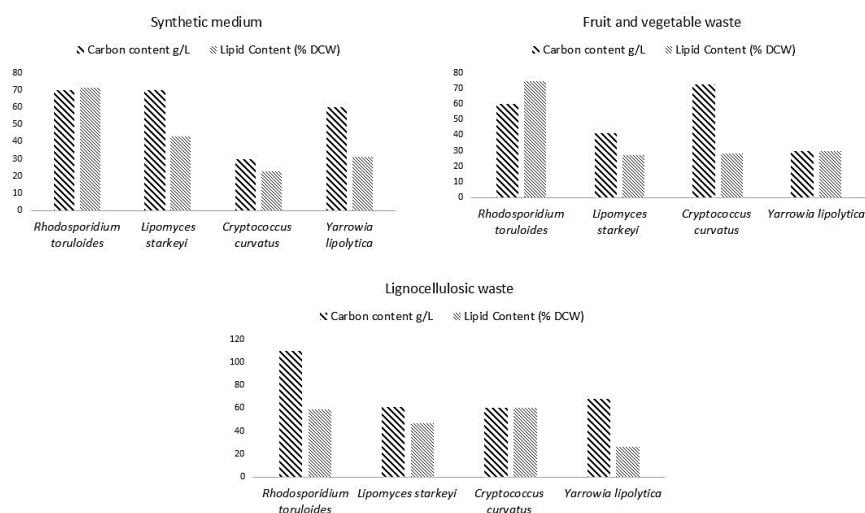


Fig. 2: *R. toruloides* stands out as a strong candidate for microbial alchemy, i.e. bioconversion of low-cost, variable waste streams to high value products.

as fruit and vegetable waste and lignocellulosic waste. The compositions of these two substrate categories differ in carbon content and recalcitrance to pretreatment processes.

3.1.1 Fruit and vegetable waste

Organic refuse from vegetable markets, including peels, pulp, and spoiled produce has been investigated as a promising substrate for yeast-based fermentation because of its rich carbohydrate content such as glucose, xylose, sucrose, and widespread availability in urban regions [13, 34, 35]. This type of waste contains a higher proportion of water, sugars, organic acids, and dietary fibers, making it well-suited for aerobic and anaerobic digestion to produce value added products. Thereby they can be degraded via steaming or hydrolyzing [13, 16]. Many studies highlight the potential of yeast strains, including *Y. lipolytica*, *R. toruloides*, *C. curvatus*, etc in bioconversion process (Table 1). Advancements in biotechnology have enhanced yeast-based waste processing, improving efficiency and economic feasibility [34-36]. After appropriate pretreatment and

hydrolysis, this waste has been effectively used to promote yeast growth and lipid production, providing the combined advantage of sustainable waste disposal and biodiesel generation [37-38]. Utilizing agricultural and vegetable market waste as substrates for fermentation offers a cost-effective solution for biodiesel production while promoting sustainable waste management and advancing the principles of a circular bioeconomy. This strategy helps alleviate the food-versus-fuel dilemma, transforms low-value organic residues into useful resources, and encourages the adoption of eco-friendly biofuel technologies.

3.1.2 Lignocellulosic waste

This category comprises of agricultural residues, such as wheat straw, rice husks, sugarcane bagasse, and corn stover, that are menacing for environmentalists due to inadequate disposal, leading to huge financial burden on states. These lignocellulosic wastes are easily available, and upon hydrolysis, release fermentable sugars that oleaginous yeasts can efficiently convert

Table 1: Various yeast strains that can utilize waste as feedstock for production of lipids and other industrially important by-products

Yeast strain	Waste substrate	Product	References
<i>Rhodosporidium toruloides</i>	Fruit and vegetable waste	Lipid	[13]
<i>Rhodosporidium toruloides</i>	Mandi waste	Carotenoid	[16]
<i>Cryptococcus curvatus</i>	Vegetable waste	Lipid	[17]
<i>Rhodotorulaglutinis</i>	Pulp and paper waste	Lipid	[18]
<i>Rhodotorulaglutinis</i>	Molasses	Lipid	[13]
<i>Rhodosporidium paludigenum</i>	Rice residue	Lipid	[19]
<i>Cryptococcus curvatus</i>	Corn stover	Lipid	[20]
<i>Rhodosporidium toruloides</i>	Corn stover	Lipid	[21]
<i>Rhodosporidium toruloides</i>	Wood hydrolysate	Lipid	[22]
<i>Yarrowia lipolytica</i>	Waste bread	Food and animal diet	[23]
<i>Lipomyces starkey</i>	Oil palm empty fruit bunch	Lipid	[24]
<i>Rhodosporidium toruloides</i>	Tea waste	Lipid	[25]
<i>Yarrowia lipolytica</i>	Glycerol	Lipid	[26]
<i>Rhodosporidium diobovatum</i>	Crude glycerol	Lipid	[27]
<i>Metschnikowia pulcherrima</i>	Glycerol	Lipid	[28]
<i>Yarrowia lipolytica</i>	Deproteinized whey	Lipid	[29]
<i>Yarrowia lipolytica</i>	Fish waste	Flavoring compounds	[30]
<i>Candida utilis</i>	Poultry manure	Single-cell protein	[31]
<i>Cryptococcus curvatus</i>	Rice waste & shrimp shells	Lipid	[32]
<i>Rhodosporidium toruloides</i>	Waste water	Lipid	[33]

into lipids [16, 19-22]. But compared to fruit and vegetable waste lignocellulosic waste is more resistant to degradation and requires rigorous pretreatment processes. Post pre-treatment, the released cellulose and hemicellulose must be saccharified to simple sugars utilizable by microorganisms, by chemical or enzymatic means, which leads to increased processing costs. This problem may be ameliorated using crude microbial enzymes or application of simultaneous hydrolysis and saccharification or solid state fermentation[36]. The metabolic versatility makes oleaginous yeast strains particularly suitable for fermenting lignocellulosic waste, which contains a mixture of sugars derived

from cellulose and hemicellulose. Notably, *R. toruloides* IFO0880 can efficiently utilize these sugars, even in the presence of inhibitory compounds, enhancing its applicability in bioconversion processes [39]. Dai et al. (2019) cultivated *Rhodosporidium toruloides* AS 2.1389 on enzymatically hydrolyzed corn stover followed by simultaneous saccharification and lipid production [21]. This approach achieved a lipid yield of 0.080 g/g dry pretreated biomass (DryPB) corn stover. The fatty acid profile of the produced lipids was similar to that of conventional vegetable oils, indicating their suitability for biodiesel production. *R. toruloides*-1588 and *R. toruloides*-7191 proved to be promising strains for lipid production

using C5 and C6 sugar from wood hydrolysate [22]. Agricultural wastes has the future potential to serve as feedstock for producing biofuels that are cost-wise comparable and competitive with fossil fuels.

3.2 Food and kitchen waste

Food waste, encompassing items such as expired bakery products, and kitchen residues, represents an abundant and underutilized resource for microbial lipid production. Rich in carbohydrates, proteins, and lipids, food waste can be hydrolyzed enzymatically or chemically to release fermentable sugars, providing a cost-effective substrate for oleaginous yeasts. Waste bread (WB) has been effectively utilized as a substrate for microbial lipid production. In various studies, the oleaginous yeast *Yarrowia lipolytica* strain K57 was cultivated on WB, showing that it can achieve a biomass concentration exceeding 62 g/L and a biomass yield of 0.45 g/g. This demonstrates the potential of WB as a feedstock for biomass and lipid production in a circular economy context [23,40]. Potato peel waste (PPW), a byproduct of the potato processing industry, has been explored for microbial lipid production [41]. Although specific studies on lipid production from PPW are limited, its rich carbohydrate content makes it a promising substrate for fermentation processes. Industrial food processing wastes, comprising a mix of various food residues, have been investigated for lipid production. Qi et al. (2020) demonstrated that *Rhodospiridium toruloides* M11, M14, and M18 could be

cultured in tea waste, achieving significant lipid production [25]. This study highlights the potential of utilizing various waste for lipid production. Niehus et al., (2018) explored the use of food waste hydrolysate as a substrate for cultivating *Yarrowia lipolytica* W29 (ATCC20460)[42]. The yeast achieved a lipid yield of 0.25 g/g substrate, demonstrating the viability of using food waste for microbial lipid production.

3.3 Industrial waste

Yeast fermentation has been widely explored for the valorization of industrial waste. Compared to agricultural or domestic one, industrial waste is considered much more complicated in terms of pollutants nature (heavy metals, recalcitrant substances, pesticides, etc.) and their abundance [43]. Thus, their treatment is a significant environmental challenge. Innumerable electro and thermochemical conversion techniques have been assessed to tackle this but oleaginous yeast have the ability to metabolize structurally diverse waste compounds and route them towards a single product such as lipids, something that purely chemical processes struggle to achieve efficiently[43]. For instance, *Yarrowia lipolytica* has been shown to utilize glycerol, a biodiesel byproduct for growth and accumulate lipids up to 40% of its dry cell weight (DCW) [26]. Likewise, *Cryptococcus curvatus* has demonstrated efficient lipid accumulation when cultivated on organic waste from food processing and textile industry waste, including glucose, xylose, and acetate[44]. In a study waste from industrial food processing, as pumpkin peels and syrup from candied fruits manufacture, were used for

Table 2: Yeast oil based companies worldwide.

Company Name	Location	Product	Feed stock
COLIPI	Germany	Essential oil	Turning CO ₂ to value
ÄIO	Estonia	Red oil, encapsulated oil	Upcycle low-value by-products into lipid rich biomass
Blinkcosmetics	-	Syfer Natural Lip Oil VC & Yeast Oil	-
The goodbye company	US	Essential oil serum	-
Insempra	Germany	Natural superior lipid	-
CleanFood.group	UK	CLEANOilCell	Food waste

cultivation of yeast such as *Rhodospiridiobolus azoricus* and *Cutaneotrichosporon oleaginosum* containing 24 g/L of lipid which can be used for biodiesel production, cosmetics, and other high-value applications [36].

3.4 Fish and poultry waste

Fish waste, including skin, bones, and viscera, can be processed using yeast fermentation to produce biofuels, enzymes, collagen, gelatin, and omega-3 oils. Similarly, poultry waste, such as feathers and blood, can be converted into protein-rich feed, bio-fertilizers, and industrial enzymes through microbial processing. *Yarrowia lipolytica*, known for its hydrocarbon metabolism, processes fish waste to generate flavoring compounds that improve the taste and aroma of seafood-based products [30]. *Candida utilis*, widely used yeast in biotechnology, transforms poultry manure into single-cell protein (SCP), offering a sustainable alternative to traditional animal feed protein sources [31]. A study investigated the possibility of using yeast fermentation to transform shrimp waste to chitin for further application [45]. When *Cryptococcus curvatus* ATCC 20509 was cultured in the blends of rice hydrolysates and shrimp shells hydrolysate, lipid content and yield were 52.25 % and 0.204 g/g. The fatty acid compositions of lipid were similar to those of typical vegetable oils [32].

4. Yeast oil and its applications

The last decade has seen significant increase in research and development in production of lipid from yeast as a promising alternative to palm and soybean oil for producing biofuels and biodiesels. Moreover, oleaginous yeast is being commercialized for human consumption along with industrial applications like paints, coatings, detergents and cosmetics [Table 2].

4.1 Biodiesel Production using yeast lipid

Since, oleaginous yeasts can accumulate high levels of lipid, more than 50% of its biomass; they can potentially be used for production of biodiesel. Yeast lipids have the

potential to help meet the growing demand for oil by partially replacing or supplementing conventional vegetable oils and fats. Biodiesel is the most commonly suggested application, followed by uses in food and dietary supplements (such as cocoa butter equivalents, palm oil substitutes, and PUFAs), as well as in oleochemicals and animal feed [46]. Notably, more than 75% of published studies have identified biofuel as a key application for yeast lipids [47-49]. The lipid profile of *R. toruloides* cultivated under the given conditions demonstrates excellent suitability for biodiesel production, particularly for markets where fuel stability and long shelf life are prioritized over low-temperature operability [13, 9].

The biodiesel derived from this microbial lipid source would exhibit high cetane number, superior oxidative stability, and good storage characteristics, making it particularly well-suited for tropical or temperate environments where cold flow is of lesser concern. However, the relatively low unsaturation also implies that cold flow properties may be suboptimal for colder climates, and thus blending with FAMES rich in monounsaturated fatty acids (e.g., from canola or soybean oil) may be necessary to achieve seasonal performance standards. In *R. rhodochrous*, over 80% lipid was accumulated with respect to its dry biomass, using glucose as a carbon source; this lipid was transesterified to produce biodiesel [51]. Palm oil, extracted from the mesocarp of oil palm fruits, is rich in saturated fatty acids—particularly about 44% palmitic acid which gives it a semi-solid consistency at room temperature. With over 69 million tonnes produced annually, it represents roughly one-third of global vegetable oil production [49]. Owing to the high oil yield of palm trees around four tonnes per hectare palm oil is also the most inexpensive vegetable oil. However, rising demand has resulted in significant deforestation, especially in tropical rainforests of Asia and South America. Yeast oil serves as a potential alternative to palm oil because of its composition and high productivity. Certain

wild-type oleaginous yeasts, such as *Lipomyces lipofer*, *L. starkeyi*, *Macalpinomyces permophorus*, *M. pulcherrima*, and *R. glutinis*, have been shown to produce oils similar in composition to palm oil [52, 53]. This yeast could potentially be used directly as cooking oil or further refined for other applications. Despite offering advantages, e.g. customizability, organic production, and being free from deforestation, impacts commercial adoption which has remained limited due to the easy availability of palm oil and its low market price. Lowering production costs through the use of inexpensive substrates and advanced processing technologies is essential. Despite several challenges the industrial sector is still interested in sustainable alternatives for palm oil and this has resulted in an upsurge in research and development [49].

4.2 Nutritional value of yeast oil

Beyond their lipid content, oleaginous yeasts offer additional nutritional value, such as a balanced amino acid profile, high levels of essential amino acids, and antimicrobial properties. Their protein content is especially rich in lysine, and the methionine content is comparable to that of soybean meal, making them suitable for feed supplementation. Using the entire yeast biomass helps retain valuable nutraceuticals within the cells. Furthermore, antimicrobial compounds produced by yeasts may enhance feed preservation and potentially improve disease resistance in animals. Noteworthy oleaginous yeasts with known antimicrobial activity include *Metschnikowia pulcherrima* and *Wickerhamomyces anomalus*[49].

Oleaginous yeasts can serve as a valuable component of animal and aquaculture feed, offering a rich source of protein, vitamins, antioxidants, and fatty acids. Supplementing animal feed with long-chain polyunsaturated fatty acids (PUFAs) has shown benefits across various types of livestock, including poultry, pigs, and dairy animals. One notable example is Verlasso, a venture based on DuPont's genetically

modified *Yarrowia lipolytica* strain, which was developed to enhance the levels of EPA and DHA in salmon diets[53].

4.3 Ornamental value of yeast oil

The cosmetics industry is vast and highly profitable, but it often faces concerns regarding the safety and effectiveness of its products. As a result, introducing new ingredients requires careful consideration and thorough testing. Yeast oil is a well-recognized ingredient in cosmetics, known for its biologically beneficial components like amino acids, polysaccharides, which moisturize skin, promote cell regeneration, delay signs of aging, and accelerate wound healing as well. It is typically formulated with other agents like vitamins, moisturizers, and antioxidants to enhance its cosmetic effects. For instance, an oral tablet combining yeast extract with vitamins has been developed to treat sunburn by reducing skin cell damage and lipid peroxidation, highlighting its potential in preventing photoaging and oxidative stress [50].

Nonetheless, any new ingredient or formulation must undergo stringent safety testing. The effect of yeast extract on skin irritation was tested along common skincare ingredients such as vitamins A, C, and E on the skin of healthy individuals. After several days of use, no irritation was observed, and all formulations improved skin smoothness and increased hydration in the stratum corneum. While these results are promising, additional studies are needed to validate the safety profile of such products [50].

Yeast oil also demonstrates the ability to break down and eliminate melanin in the skin, making it useful in skin-brightening products. A comparison between a conventional skin-lightening agent and a formulation containing natural ingredients like yeast extract and salicylic acid showed similar efficacy in reducing dark spots and evening out pigmentation [50].

However, the natural formulation offered advantages such as abundant raw material availability and lower production costs. Overall, natural ingredient-based formulations,

including those with yeast extract, tend to be better tolerated over long-term use than those with synthetic chemicals, offering greater promise for future innovation. Despite its potential, the use of yeast extract in skincare is still relatively limited and underexplored in scientific research, suggesting a significant opportunity for new product development and applications [51, 52].

5. Circular economy and environmental impact

5.1 Circular economy

Yeast plays a vital role in the circular bioeconomy by transforming waste into valuable resources, particularly in waste management and microbial oil production. Through waste valorization, yeast can convert organic waste, such as food scraps and agricultural residues, into useful products, reducing landfill waste and contributing to sustainability [46]. After utilizing the waste the remaining residue can be used as bio-compost. In microbial oil production, yeast, especially strains like *Yarrowia lipolytica*, *Rhodospiridium toruloides* efficiently utilize low-cost substrates such as lignocellulosic waste and industrial by-products to generate single-cell oils (SCOs), which serve as sustainable alternatives to petroleum-based fuels and vegetable oils [55]. The de-oiled yeast biomass can be further used as nitrogen source for yeast cultivation or poultry feed. Additionally, yeast exhibits potential in plastic waste up cycling [35]. These processes help reduce pollution and lower carbon emissions, aligning with global sustainability goals. By repurposing waste and supporting biofuel production, yeast-driven innovations enhance environmental conservation and industrial efficiency, making them an integral part of the circular bioeconomy [Figure 1].

5.2 Environmental benefits

5.2.1 Converting Waste to Resources

Yeasts are capable of converting agricultural and industrial waste materials into

valuable lipids, helping reduce the volume of waste sent to landfills and lowering methane emissions. This supports waste-to-value initiatives and sustainable resource management [56].

5.2.2 Renewable Lipid Sources

Single-cell oils (SCOs) produced by yeasts offer a promising alternative to conventional oils, including fossil-derived and palm-based oils. Their use in biodiesel production can reduce deforestation, lead to the waste management and build reliance on non-renewable resources [57,58].

5.3 Environmental Drawbacks

5.3.1 High Energy Consumption

Producing and processing yeast lipids—especially under aerobic conditions—requires significant amounts of energy. This can result in a considerable carbon footprint unless offset by renewable energy sources [58].

5.3.2 Wastewater Management Challenges

The fermentation process generates nutrient-rich wastewater, particularly high in nitrogen and phosphorus. If not properly treated, this effluent can lead to water pollution and eutrophication, harming aquatic ecosystems[59]. However, this wastewater may be used for irrigation, because it is loaded with micro nutrients for plants.

6. Yeast oil: challenges, mitigation and future prospects

6.1 Inconsistent raw materials

Organic feedstock e.g. fruit peels, molasses, lignocellulosic biomass, etc often fluctuates in composition based on seasonal and regional factors. As mentioned in section 2, the variability of substrates presents a significant challenge for microorganisms involved in biofuel fermentation [36]. While addressing the seasonal availability of these substrates may remain difficult until large-scale, year-round production of food, fruits, and vegetables is

achieved, alternative strategies could help mitigate this issue. One promising approach is the development of optimized starter cultures and microbial consortia, incorporating novel isolates and genetically modified microorganisms [2]. These could be engineered to hydrolyze and metabolize diverse agricultural wastes based on their specific compositions, marking a crucial advancement in overcoming this challenge [36]. Additionally, the hydrolysate may contain microbial growth inhibitors, such as levulinic acid, acetic acid, and other weak organic acids, which can adversely affect yeast metabolism. To overcome this, either a pretreatment step to detoxify the hydrolysate or the development of inhibitor-tolerant yeast strains may be required, as emphasized in previous studies [2, 36].

6.2 *Difficulty in scaling up*

Lipid production in yeasts is highly sensitive to environmental and nutritional parameters like pH, temperature, carbon-to-nitrogen (C/N) ratio, and oxygen availability. While these can be tightly controlled in laboratory setups, reproducing the same precision in industrial bioreactors is much more complex and cost-intensive [60, 61].

6.3 *Complex lipid extraction*

Extracting lipids from yeast cells typically requires high energy input and often relies on toxic solvents such as hexane and chloroform. Although eco-friendly alternatives such as supercritical CO₂ extraction are emerging, they involve higher expenses and are technically demanding for industrial use [50].

6.4 *Concerns about genetic modification*

To increase lipid yields or expand substrate range, many yeast strains are genetically engineered. However, the use of genetically modified organisms (GMOs) raises public concern and regulatory barriers, particularly in regard to biosafety and environmental impact [2]. Finally, the

downstream conversion of lipid to biodiesel remains an energy-intensive phase of the production pipeline. Traditional transesterification processes involve multiple steps, significant energy input, and post-reaction purification, which contribute to overall inefficiencies. Integrating advanced bioprocess strategies such as *in situ* transesterification or direct fermentation-to-biodiesel methods (i.e., consolidated bioprocessing) may offer more streamlined alternatives. Such approaches combine lipid production and conversion into a single operational step, minimizing handling and reducing energy demand, as demonstrated in studies by Zhang et al [63]. Addressing these challenges through both biological and process engineering interventions will be essential for the sustainable industrial deployment of microbial biodiesel platforms [2, 64].

6.6 *Future prospects*

The successful demonstration of lipid accumulation by yeast when cultivated on waste highlights the growing promise of microbial oils as sustainable and renewable alternatives to conventional plant-based biodiesel feedstocks. Unlike crop-derived oils, microbial lipids can be produced on non-arable land, using agro-industrial residues, thereby avoiding food-versus-fuel conflicts and contributing to waste valorization. This aligns well with the principles of a circular bioeconomy, where resource efficiency and environmental sustainability are prioritized [2, 65, 66]. Furthermore, the energy requirements for steps such as biomass drying, lipid extraction, and solvent evaporation are substantial and must be accounted for in life cycle assessments (LCA). Strategies like adopting low-energy drying methods (e.g., solar or vacuum drying) and implementing energy-efficient process designs could significantly decrease overall energy consumption and environmental footprint [67, 68]. The development of green solvent-based extraction methods (ethanol, toluene, and

ethyl acetate etc.) offers a promising path toward sustainable lipid recovery, reducing reliance on toxic solvents [9]. Future advancements should focus on optimizing biodegradable and renewable solvents to enhance efficiency and minimize environmental impact. Enhancing lipid yield either through metabolic engineering or process optimization would also reduce the biomass requirement per unit of lipid, further improving the process's economic and environmental performance [2].

7. Conclusion

Microbial lipid production using waste substrates presents a sustainable and cost-effective platform, but its success at an industrial scale will depend on addressing key technical limitations. With continued advancements in green technology and strain development, this approach could become a competitive and eco-friendly alternative in the biofuel and oleo-chemical sectors. Even after dynamic research on sustainable oil production from oleaginous yeast strains, this product has not been widely upscaled by industry and only few industries have accepted the challenges. Public awareness about accepting yeast oil as a healthier and environment-friendly alternative to conventional plant oils holds the key to its successful commercialization. Extensive outreach programs must propagate yeast oil applications to promote more initiatives and industries in this area.

8. Acknowledgment

The authors acknowledge DST-ANRF (CRG/2022/003411) for funding and Amity University for support and research facilities. The insightful discussion with Prof. Jean-Marc Nicaud (AgroParisTech, Université, Paris-Saclay, Jouy-en-Josas, France) and Dr Dipanwita Biswas is greatly acknowledged.

9. Conflicts of interest

The authors declare that there is no conflict of interests.

10. References

- 1 Mota MN, Múgica P, Sá-Correia I. Exploring Yeast Diversity to Produce Lipid-Based Biofuels from Agro-Forestry and Industrial Organic Residues. *Journal of Fungi*. 2022; 8(7):687.
- 2 Kumar KK, Deeba F, Pandey AK, Islam A, Paul D, Gaur NA. Sustainable lipid production by oleaginous yeasts: Current outlook and challenges. *Bioresour Technol*. 2025;421:132205.
- 3 Ageitos, Jose Manuel et al. Oily yeasts as oleaginous cell factories. *Applied microbiology and biotechnology*. 2011;90(4): 1219-27.
- 4 Němcová A, Szotkowski M, Samek O, Cagánová L, Sipiczki M, Márová I. Use of Waste Substrates for the Lipid Production by Yeasts of the Genus *Metschnikowia*-Screening Study. *Microorganisms*. 2021 Nov 4;9(11):2295.
- 5 Warke MA, Pawar PP, Kothari et al. Two-stage approach for microbial oil production using *Yarrowia lipolytica* NCIM 3590. *Advances in Biotechnology & Microbiology*. 2017;5(1):555652.
- 6 Banerjee S, Dien BS, Kristen K et al. Pilot-scale processing of *Miscanthus x giganteus* for recovery of anthocyanins integrated with production of microbial lipids and lignin-rich residue. *Chemical Engineering Journal*. 2024;485:150117.
- 7 Daniel Pleissner, Tsz Him Kwan, Carol Sze Ki Lin. Fungal hydrolysis in submerged fermentation for food waste treatment and fermentation feedstock preparation. *Bioresource Technology*. 2014;158:48-54.
- 8 Rakicka M, Lazar Z, Dulerio T et al. Lipid production by the oleaginous yeast *Yarrowia lipolytica* using industrial by-products under different culture conditions. *Biotechnol Biofuels*. 2015;8:104.
- 9 Zainuddin MF, Fai CK, Ariff AB, Rios-Solis L, Halim M. Current Pretreatment/Cell Disruption and Extraction Methods Used to Improve Intracellular Lipid Recovery from Oleaginous Yeasts. *Microorganisms*. 2021; 9(2):251.

- 10 Brandenburg, Jule et al. Oleaginous yeasts respond differently to carbon sources present in lignocellulose hydrolysate. *Biotechnology for biofuels*. 2021;14(1):124 .
- 11 Yonghong Li, Zongbao (Kent) Zhao, Fengwu Bai. High-density cultivation of oleaginous yeast *Rhodospiridium toruloides* Y4 in fed-batch culture. *Enzyme and Microbial Technology*. 2007; 41 (3,2007):312-317.
- 12 Brandenburg J, Blomqvist J, Shapaval V et al. Oleaginous yeasts respond differently to carbon sources present in lignocellulose hydrolysate. *Biotechnol Biofuels*. 2021;14: 124.
- 13 Singh G, Sinha S, Bandyopadhyay KK et al. Triauxic growth of an oleaginous red yeast *Rhodospiridium toruloides* on waste 'extract' for enhanced and concomitant lipid and β -carotene production. *Microb Cell Fact*. 2018;17:182.
- 14 Tanimura A, Sugita T, Endoh R, Ohkuma M, Kishino S, Ogawa J, Shima J, Takashima M. Lipid production via simultaneous conversion of glucose and xylose by a novel yeast, *Cystobasidium iriomote nse*. *PLoS One*. 2018;13(9): e0202164.
- 15 Zhiwei Gong, Qian Wang, Hongwei Shen, Cuimin Hu, Guojie Jin, Zongbao K Zhao. Co-fermentation of cellobiose and xylose by *Lipomyces starkeyi* for lipid production. *Bioresource Technology*. 2012;117:20-24.
- 16 Sinha S, Singh G, Arora A et al. Carotenoid Production by Red Yeast Isolates Grown in Agricultural and "Mandi" Waste. *Waste Biomass*. 2021; 12:3939–3949.
- 17 Chatterjee SS, Mohan V. Microbial lipid production by *Cryptococcus curvatus* from vegetable waste hydrolysate, *Bioresource Technology*. 2018;254:284-289.
- 18 Amirsadeghi, Marta, Shields-Menard, Sara French, Todd & Hernandez, Rafael. Lipid Production by *Rhodotorulaglutinis* from Pulp and Paper Wastewater for Biodiesel Production. *Journal of Sustainable Bioenergy Systems*. 2015;5:114-125.
- 19 Chaiyaso, Thanongsak & Techapun, Charin & Seesuriyachan, Phisit & Watanabe, Masanori & Srisuwan, Wimada. Screening of Oleaginous Yeast for Lipid Production Using Rice Residue from Food Waste as a Carbon Source. *KKU Res.j*. 2016;22:116-126.
- 20 Gong Z, Shen H, Yang X, Wang Q, Xie H, Zhao ZK. Lipid production from corn stover by the oleaginous yeast *Cryptococcus curvatus*. *Biotechnol Biofuels*. 2014;7(1):158.
- 21 Dai X, Shen H, Li Q, Rasool K, Wang Q, Yu X, Wang L, Bao J, Yu D, Zhao ZK. Microbial Lipid Production from Corn Stover by the Oleaginous Yeast *Rhodospiridium toruloides* Using the PreSSLP Process. *Energies*. 2019;12(6):1053.
- 22 Carlos S Osorio-González, Krishnamoorthy Hegde, Pedro Ferreira, et al. Lipid production in *Rhodospiridium toruloides* using C-6 and C-5 wood hydrolysate: A comparative study. *Biomass and Bioenergy*. 2019;130:105355.
- 23 Carsanba E, Agirman B, Papanikolaou S, Fickers P and Erten H. Valorisation of Waste Bread for the Production of Yeast Biomass by *Yarrowia lipolytica* Bioreactor Fermentation. *Fermentation*. 2023;9(7):687.
- 24 Thanapimmetha, Anusith & Peawsuphon, Noppan & Chisti, Yusuf & Saisriyoot, Maythee & Srinophakun, Penjit. Lipid production by the yeast *Lipomyces starkeyi* grown on sugars and oil palm empty fruit bunch hydrolysate. *Biomass Conversion and Biorefinery*. 2021.
- 25 Qi F, Shen P, Hu R. et al. Carotenoids and lipid production from *Rhodospiridium toruloides* cultured in tea waste hydrolysate. *Biotechnol Biofuels*. 2020;13:74.
- 26 Papanikolaou S, Aggelis G. Lipids of oleaginous yeasts. Part II: Technology and potential applications. *European Journal of Lipid Science and Technology*. 2011;113(8):1052–1073.
- 27 Munch, Garret, Sestric, Ryan, Sparling, Richard, Levin, David B, Cicek, Nazim. Lipid Production in the under-characterized oleaginous yeasts, *Rhodospiridium Babjevae* and *Rhodospiridium diobovatum*, from

biodiesel-derived waste glycerol. *Bioresour. Technol.* 2015;185:49–55.

28 Santamauro F, Whiffin FM, Scott RJ, Chuck CJ. Low-cost production by oleaginous yeast cultured in non-sterile conditions using waste resources. *Biotechnol Biofuel.* 2014;7:34–44.

29 Taskin M, Saghafian A, Aydogan MN, Arslan NP. Microbial lipid production by cold-adapted oleaginous yeast *Yarrowia lipolytica* B9 in non-sterile whey medium. *Biofuel Bioprod Bioref.* 2015;5:595–605.

30 Gottardi D, Ciccone M, Siroli L, Lanciotti R, Patrignani F. Use of *Yarrowia lipolytica* to Obtain Fish Waste Functional Hydrolysates Rich in Flavoring Compounds. *Fermentation.* 2022;8(12):708.

31 Kargi F, Shuler ML. A mixed yeast-bacteria process for the aerobic conversion of poultry waste into single-cell protein. *Biotechnol Lett.* 1981;3:409–414.

32 Jia-xuan Zhang, Xiao-le Liu, Li Wang, Zhen Fang. Two-stage process production of microbial lipid by co-fermentation of glucose and N-acetylglucosamine from food wastes with *Cryptococcus curvatus*. *Bioresource Technology.* 2023;387:129685.

33 Ling J, Nip S, Shim H. Enhancement of lipid productivity of *Rhodospiridium toruloides* in distillery wastewater by increasing cell density. *Bioresour Technol.* 2013;146:301–9.

34 Râpă M. Darie-Niță, R.N Coman, G. Valorization of Fruit and Vegetable Waste into Sustainable and Value-Added Materials. *Waste.* 2024;2(3):258-278.

35 Mújdeci GN, Khosravi-Darani K. Valorization of Fruit and Vegetable Waste: Yeast Fermentation. Springer. 2022;315-342.

36 Donzella S, Serra I, Fumagalli A et al. Recycling industrial food wastes for lipid production by oleaginous yeasts *Rhodospiridiobolusazoricus* and *Cutaneotrichosporonoleaginosum*. *Biotechnol Biofuels.* 2022;15:51.

37 Ravindran R. and Jaiswal AK. Exploitation of Food Industry Waste for

High-Value Products. *Trends in Biotechnology.* 2016;34(1):58–69.

38 Angerbauer C, Siebenhofer M, Mittelbach, Guebitz, GM. Conversion of sewage sludge into lipids by *Lipomyces starkeyi* for biodiesel production. *Bioresource Technology.* 2008; 99 (8): 3051–3056.

39 Kim J, Coradetti, ST Kim, YM Gao, et al. Multi-Omics Driven Metabolic Network Reconstruction and Analysis of Lignocellulosic Carbon Utilization in *Rhodospiridium toruloides*. *Frontiers in Bioengineering and Biotechnology.* 2021;8:612832 .

40 Carsanba E, Papanikolaou S, Fickers P, Agirman B, Erten H. Citric Acid Production by *Yarrowia lipolytica*. In: *Non-conventional Yeasts: from Basic Research to Application.* Springer. 2019:91-117.

41 Javed A, Ahmad A, Tahir A, Shabbir U, Nouman M, Hameed A. Potato peel waste-its nutraceutical, industrial and biotechnological applications. *AIMS Agriculture and Food.* 2019;4(3):807–823.

42 Niehus X, Crutz-Le Coq, AM, Sandoval G, Nicaud JM, Ledesma-Amaro R. Engineering *Yarrowia lipolytica* to enhance lipid production from lignocellulosic materials. *Biotechnology for Biofuels.* 2018;11(1):11.

43 Sartaj K, Prasad R, Matsakas L, Patel A. Review of Transforming recalcitrant wastes into biodiesel by oleaginous yeast: An insight into the metabolic pathways and multi-omics landscape. *Chemical Engineering Journal.* 2023; 474.

44 Ryu BG, Kim J, Kim K, Choi YE, Han JI, Yang JW. High-cell-density cultivation of oleaginous yeast *Cryptococcus curvatus* for biodiesel production using organic waste from the brewery industry. *Bioresource Technology.* 2013;135:357-364.

45 T M N Ta et al. Investigation of chitin recovery from shrimp waste by yeast fermentation. *IOP Conf. Ser.: Earth Environ. Sci.* 2023;1155:012012.

- 46 Abein F, Chuck CJ. The history, state of the art and future prospects for oleaginous yeast research. *Microb Cell Fact.* 2021;20(1):221.
- 47 AE Atabani, TMI Mahlia, HH Masjuki, et al. A comparative evaluation of physical and chemical properties of biodiesel synthesized from edible and non-edible oils and study on the effect of biodiesel blending. *Energy.* 2013;58:296-304.
- 48 Demirbaş Ayhan. Biodiesel: A Realistic Fuel Alternative for Diesel Engines. 2008:111-160
- 49 Zuleta E C, Baena L, Rios L A, Calderón JA. The oxidative stability of biodiesel and its impact on the deterioration of metallic and polymeric materials: A review. *Journal of the Brazilian Chemical Society.* 2012;23:2159–2175.
- 50 Rakicka M et al. Evaluation of different extraction techniques for lipid recovery from *Yarrowia lipolytica*. *Journal of Industrial Microbiology & Biotechnology.* 2015; 42(5):729–738.
- 51 Shields-Menard SA, Amirsadeghi M, Sukhbaatar B, Revellame E, Hernandez R, Donaldson JR, French WT. Lipid Accumulation by *Rhodococcus rhodochrous* Grown on Glucose. *Journal of Industrial Microbiology and Biotechnology.* 2015;42:693-699.
- 52 Sara A Shields-Menard, Marta Amirsadeghi, W Todd French, Raj Boopathy. A review on microbial lipids as a potential biofuel. *Bioresource Technology.* 2018; 259:451-460.
- 53 Athenstaedt K, Jolivet P, Boulard C, Zivy M, Negroni L, Nicaud JM, Chardot T. Lipid particle composition of the yeast *Yarrowia lipolytica* depends on the carbon source. *Proteomics.* 2006;6(5):1450-9.
- 54 Tao Z, Yuan H, Liu M, Liu Q, Zhang S, Liu H, Jiang Y, Huang D, Wang T. Yeast Extract: Characteristics, Production, Applications and Future Perspectives. *J Microbiol Biotechnol.* 2023;33(2):151-166.
- 55 Odaneth, AA, Pawar, PP, Kothari SD, Warke MA, Lali AM. Simultaneous lipid biosynthesis and recovery for oleaginous yeast *Yarrowia lipolytica*. *Biotechnology for Biofuels.* 2019;12:237.
- 56 Saranraj P, et al. Microbial degradation of agricultural wastes. *International Journal of Microbiological Research.* 2013;4(3):235–245.
- 57 Beopoulos A, et al. *Yarrowia lipolytica* as a model for bio-oil production. *Trends in Biotechnology.* 2009;27(10):494–500.
- 58 Kumar S, Shukla A, Singh P. Environmental impact of biofuel production. *Renewable Energy.* 2020;147:2033-2043.
- 59 Li Y, Jin Y, Li J, & Li H. *Treatment of distillery wastewater using anaerobic digestion and short-cut nitrification–denitrification process.* *Bioresource Technology,* 2013;132, 190–198.
- 60 Li Y, et al. Biofuels from Microalgae. *Biotechnology Progress.* 2008; 24(4): 815–820.
- 61 Papanikolaou S, et al. Lipid production by oleaginous yeast growing on industrial glycerol. *Bioresource Technology.* 2002;82(1):43–49.
- 62 Groenewald M, et al. The road to industrial biotechnology: a case study of yeast development. *FEMS Yeast Research.* 2014;14(6):822–834.
- 63 Zhang X, Yan S, Tyagi RD, Surampalli RY, Valéro JR. Ultrasonication aided in-situ transesterification of microbial lipids to biodiesel. *Bioresour Technol.* 2014;169:175–180. doi:10.1016/j.biortech.2014.06.108.
- 64 Yellapu SK, Kaur R, Kumar LR, Tiwari B, Zhang X, Tyagi RD. Recent developments of downstream processing for microbial lipids and conversion to biodiesel. *Bioresour Technol.* 2018;256:515–528. doi:10.1016/j.biortech.2018.01.129.
- 65 Zeng Y, Bian D, Xie, Y, Jiang X, Li X, Li P, Zhang Y, Xie T. Utilization of food waste

hydrolysate for microbial lipid and protein production by *Rhodospiridium toruloides* Y2. Journal of Chemical Technology & Biotechnology. 2016;92(3):666–673.

66 Carmona-Cabello M, García IL, Papadaki A, Tsouko E, Koutinas A, Dorado MP. Biodiesel production using microbial lipids derived from food waste discarded by

catering services. Bioresource Technology. 2021;323:124597.

67 Sander K, Murthy GS. Life cycle analysis of algae biodiesel. The International Journal of Life Cycle Assessment, 2010;15(7):704–714.

68 Belessiotis, V, Delyannis E. Solar drying. Solar Energy. 2011;85(8):1665–1691.