

Use of Agro-Industrial Waste as Substrates in Citric Acid Production: A Review

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Abstract

Due to its antioxidant, acidifying, and flavor-enhancing properties, citric acid is one of the most versatile organic acids, with high demand in the food and beverage industry. Although the conventional system utilizes the fungus *Aspergillus niger* in media based on sugarcane molasses, the need for economic and environmental optimization has directed research toward alternative substrates and microorganisms. This review aims to conduct a literature survey of the potential use of agro-industrial waste as carbon and nitrogen sources in citric acid production. A structured literature search was conducted in the ScienceDirect, PubMed, SciELO and Google Scholar databases, covering studies published between 1977 and 2026, from which 65 production records were compiled. Data analysis shows that the diversity of available waste, combined with new fermentation strategies, allows not only for the reduction of operational costs but also for the mitigation of environmental impacts. A critical analysis further shows that yield variability within a single substrate class often exceeds the variability between different classes, indicating that inducer supplementation, pretreatment and nutrient balance are stronger determinants of performance than substrate identity alone.

1. Introduction

Citric acid (2-hydroxypropane-1,2,3-tricarboxylic acid) is an organic acid widely distributed in nature. Its concentration is significantly high in citrus fruits, where it is primarily responsible for the acidic sensory profile and characteristic flavor of lemons and oranges. Physiologically, this compound is involved in various metabolic processes in living beings, including the citric acid cycle (Krebs cycle), which is responsible for cellular respiration in aerobic cells (McMurry 2016; Nangare et al. 2021; Kirimura and Yoshioka 2019; Show et al. 2015).

In addition to being extremely important for the metabolism of living organisms, citric acid is also widely used in various industrial sectors because it is biodegradable, water-soluble, non-toxic, antioxidant, acidifying, and flavoring; it can also act as a preservative and buffering agent (Nangare et al. 2021; Yu et al. 2017; Zhang et al. 2020). Due to its broad applicability, global citric acid production has shown continuous growth. In 2025, the produced volume of citric acid exceeded 3 million tons, with an economic turnover higher than US\$ 3 billion (Książek 2023; Behera et al. 2021; Mordor Intelligence 2025; Grand View Research 2023; IMARC 2025).

At the beginning of its production history, citric acid was isolated from lemons (Berovic and Legisa 2007; Sackett 2024; Vargas and Medina 2012). However, currently, its industrial production is predominantly carried out via the fermentative pathway using the filamentous fungus *Aspergillus niger* and carbon and nitrogen sources such as sugarcane or beet molasses, corn starch, and starch hydrolysate (Kirimura and Yoshioka 2019).

Other microorganisms such as *Candida lipolytica*, *Penicillium citrinum*, and *Yarrowia lipolytica* are also capable of accumulating citric acid, but *Aspergillus niger* remains the most widely used producer owing to its high productivity and its ability to grow on diverse substrates (Dhillon et al. 2011; Kudzai et al.

2016; Ozdal and Basara 2019; Rzechonek et al. 2019). The present review has three objectives. First, it compiles, in a uniform format, production data from agro-industrial residues across both submerged and solid-state fermentation, allowing direct comparison of substrate performance. Second, it analyses the variability observed within each residue class and identifies the process variables that account for it. Third, it situates this body of evidence within the context of Brazilian agro-industry, one of the largest generators of such residues worldwide, and identifies the research gaps that currently separate laboratory demonstration from industrial adoption.

2. Review methodology

This review followed a structured literature search conducted between 2024 and 2026 in the ScienceDirect, PubMed, SciELO and Google Scholar databases, complemented by the Periódicos CAPES portal and ResearchGate for access to full texts. The search combined the terms “citric acid production” with “agro-industrial waste”, “agro-industrial residue”, “by-product”, “submerged fermentation”, “solid-state fermentation”, “*Aspergillus niger*” and “*Yarrowia lipolytica*”, in English and Portuguese, without restriction on publication year. The earliest primary study retained dates from 1977 and the most recent from 2026.

Studies were included when they (i) reported experimental citric acid production by a microorganism, (ii) used an agro-industrial residue or by-product as the main carbon source, or a conventional or synthetic medium serving as a comparative baseline, and (iii) reported a quantitative maximum titre or yield. Studies were excluded when the substrate was not specified, when production was reported only qualitatively, or when the full text could not be obtained. Review articles were consulted for context but were not entered in the production tables.

For each retained study, the following variables were extracted: producing microorganism, substrate, fermentation mode, fermentation time and maximum production. Values were transcribed in the units reported by the original authors (g L^{-1} for submerged fermentation; g kg^{-1} of dry substrate for solid-state fermentation) and were not converted, since substrate loading was frequently unreported. This resulted in 39 production records for submerged fermentation (Table 2) and 26 for solid-state fermentation (Table 3), in addition to a survey of industrial applications (Table 1).

3. Applications of citric acid

Citric acid is a widely used food additive (INS 330), employed as an acidulant, flavorant, antioxidant, preservative, or buffering agent in the production of soft drinks, juices, ice cream, and candies. With high demand, the food industry consumes approximately 75% of global citric acid production (Kirimura and Yoshioka 2019; IHS Markit 2015; Muñoz-Villa et al. 2014; Qiu et al. 2014; Sawant et al. 2018). Citric acid is also used in fruit preservation, acting in the control of microbial diseases—such as in apples and cashews, which are highly susceptible to post-harvest infection by *Rhizopus*, *Colletotrichum*, and

Penicillium—and in the inhibition of enzymes that cause fruit browning (Duangmal et al. 2017; Goyeneche et al. 2014; Sanchís et al. 2016; Yahia 2011).

In the chemical sector, about 15% of global production is used for industrial applications, including metallurgical and textile processes and cleaning product formulations (Kirimura and Yoshioka 2019; Show et al. 2015; Angumeenal and Venkappayya 2013). In detergents, citric acid has replaced phosphate compounds due to its lower eutrophic impact on aquatic ecosystems. Additionally, several studies demonstrate its effectiveness in treating effluents containing heavy metals, attributed to its high complexation capacity with metal ions such as iron and copper (IHS Markit 2015; Muñoz-Villa et al. 2014; Di Palma and Mecozzi 2007).

The remaining 10% is destined for the pharmaceutical and cosmetic industries, where citric acid can be used as a flavoring agent, antioxidant, anticoagulant, or buffering agent (Nangare et al. 2021; Kirimura and Yoshioka 2019; Show et al. 2015; Książek 2023; Soccol et al. 2006). Furthermore, citric acid has been used as an additive in animal nutrition, with studies demonstrating relevant antimicrobial, anti-inflammatory, and nutritional effects (Dai et al. 2018; Ferreira et al. 2020; Khajepour and Hosseini 2012; Swaggerty et al. 2020; Zhao et al. 2019).

As observed in Table 1, the use of citric acid covers a wide range of industrial sectors. The growing demand for citric acid stimulates the study of new production routes, seeking increased yield and reduced process costs.

Table 1
Industrial and biotechnological applications of citric acid reported in the literature.

Sector	Application	Reference
Animal Nutrition	Nutritional supplement for piglet growth.	Ferreira et al. (2020)
	Anti-inflammatory dietary supplement for turbot fish.	Zhao et al. (2019)
	Nutritional supplement to improve the immune system of chicks	Swaggerty et al. (2020)
	Nutritional supplement in fishmeal for belugas	Khajepour and Hosseini (2012)
Food	Microbial control in raw oysters	Mahmoud (2014)
	Quality improvement of sea bass fillets	Qiu et al. (2014)
	Enzymatic inhibition for prevention of fruit browning	Duangmal et al. (2017); Goyeneche et al. (2014)
	Enzymatic inhibition for browning prevention and microbial control in persimmons	Sanchís et al. (2016)
Effluent treatment	Eco-friendly calcium phosphate scale inhibitor	Zhang et al. (2016)
	Leaching of lithium batteries for recovery of Li, Co, Ni, and Mn	Meng et al. (2020)
	Chelating agent in heavy metal extraction from industrial and municipal sludge	Wang et al. (2015)
	Chelating agent in the mobilization of heavy metals in marine sediments	Di Palma and Mecozzi (2007)
Medicine	Synthesis of nanoparticles for medical applications	Nataraj et al. (2020)
	Retarding agent for bone degeneration	Wang et al. (2019)
	Protective agent against liver injury	Kim et al. (2019)
	Promotion of immune function by strengthening the intestinal barrier and modulating inflammatory response	Hu et al. (2024)
Industry	Crosslinking agent for improving mechanical properties and reducing water absorption in banana and wheat fiber	Nataraj et al. (2018)
	Homogeneous catalyst for sugarcane bagasse pretreatment for better sugar conversion	Gomes et al. (2020)
	Crosslinking agent for improving packaging produced with bovine collagen	Andonegi et al. (2020)

Sector	Application	Reference
	Complexing agent and carbonaceous precursor in the synthesis of electrochemical materials for sodium-ion batteries	Huang et al. (2022)
	Leaching agent for iron removal in white porcelain	Katsuki et al. (2026)

Source: prepared by the authors, 2026.

4. Citric acid production

Historically, citric acid was first isolated from lemon juice in 1784 by Karl Wilhelm Scheele. The commercialization of this compound only occurred in 1826 in England by John and Edmund Sturge; lemons were also used for its production, as about 9% of a lemon's mass is composed of citric acid. In 1893, Carl Wehmer studied the accumulation of citric acid through the fermentation of the fungus *Penicillium glaucum* in a culture medium containing sucrose. Microbiological production only reached an industrial scale in the United States in 1923, following James N. Currie's discovery of the production potential of the fungus *Aspergillus niger* in 1917 (Kirimura and Yoshioka 2019; Berovic and Legisa 2007; Grewal and Kalra 1995; Max et al. 2010; Currie 1917).

Currently, the traditional extraction method from citrus fruits is still used in some countries like Greece and Mexico; however, its production is not significant. Citric acid can also be obtained through chemical synthesis, but fermentation using microorganisms has become the most economical production route, accounting for about 99% of global production (Berovic and Legisa 2007; Muñoz-Villa et al. 2014; Copetti 2019; Iqbal et al. 2015).

Microorganisms such as *Aspergillus wentii*, *Aspergillus aculeatus*, *Aspergillus awamori*, *Aspergillus carboneires*, *Aspergillus foetidus*, *Candida tropicalis*, *Candida lipolytica*, *Penicillium citrinum*, and *Yarrowia lipolytica* are capable of accumulating citric acid. However, *Aspergillus niger* is the most used microorganism in its production due to its ease of handling and high productivity across various raw materials (Muñoz-Villa et al. 2014; Angumeenal and Venkappayya 2013; Copetti 2019; Papanikolaou et al. 2002; Troiano et al. 2020).

Regarding the substrates used in citric acid production, sugarcane molasses is the most widely used industrial substrate (Kirimura and Yoshioka 2019). Sugarcane molasses is a byproduct of the sugar industry widely used in fermentative processes as a carbon source due to its composition rich in sugars such as sucrose, glucose, and fructose (Melo 2003; Feltrin et al. 2000).

Agribusiness is extremely important for the Brazilian economy, accounting for 23.5% of the country's GDP in 2024. Brazil is a pioneer in the production and export of products such as soybeans, sugar, coffee, and orange juice (Government of Brazil 2026). In this scenario, Brazil also emerges as a potential producer of agro-industrial waste. One of today's major environmental challenges is the proper

management of waste generated in different industrial processes. Consequently, several studies have evaluated the potential of using agro-industrial waste as substrates for citric acid production.

Citric acid production can occur through submerged or solid-state fermentation. Tables 2 and 3 shows the literature survey of citric acid production using filamentous fungi and yeasts with different substrates.

Table 2
Citric acid production by submerged fermentation by different microorganisms

Group	Microorganism	Substrate	Time (h)	Maximum production (g L ⁻¹)	Reference
Filamentous Fungi	A. foetidus	Brewery waste	96	12.3	Hang et al. (1977)
	A. niger	Brewery waste	336	19	Roukas and Kotzekidou (1986)
	A. niger	Sugarcane molasses	168	72	Islam et al. (1986)
	A. niger	Sugarcane molasses	168	96.1	Ikram-ul et al. (2004)
	A. niger	Sugarcane molasses	152	98.08	Lotfy et al. (2007)
	A. niger	Beet molasses	152	113.5	Lotfy et al. (2007)
	A. niger	Sugarcane molasses and pumpkin	312	14.86	Majumder et al. (2010)
	A. niger	Apple pomace sludge	96	9	Dhillon et al. (2011)
	A. niger	Whey	96	6.5	Dhillon et al. (2011)
	A. niger	Synthetic Prescott & Dunn enriched medium	24	62.9	Pastore et al. (2011)
	A. niger	Date syrup	144	56.7	Mostafa and Alamri (2012)
	A. niger	Treated sugarcane molasses	168	14.17	Iqbal et al. (2015)
	A. niger	Cassava peel	84	88.73	Adeoye et al. (2015)
	A. niger	Cassava and citric acid wastewater	60	142.4	Xu et al. (2016)
	A. niger	Rice extract	120	14.4	Kudzai et al. (2016)
	A. niger	Potato extract	96	1.47	Kudzai et al. (2016)

Group	Microorganism	Substrate	Time (h)	Maximum production (g L ⁻¹)	Reference
	<i>A. niger</i>	Corn stover	192	100.04	Zhou et al. (2017)
	<i>A. niger</i>	Potato peel and soybean meal	144	0.40453	Makut and Ekeleme (2018)
	<i>T. viride</i>	Potato peel and soybean meal	-	0.2976	Makut and Ekeleme (2018)
	<i>T. viride</i>	Glucose and soybean meal	144	15.17	Ekeleme et al. (2018)
	<i>A. niger</i>	Beet molasses and chicken feather peptone	168	68.8	Ozdal and Basara (2019)
	<i>A. niger</i>	Cheese whey	240	2.379	Magalhães et al. (2019)
	<i>A. niger</i>	Cassava peel	48	1.82	Ajala et al. (2020)
	<i>A. niger</i>	Sweet potato peel	168	3.22	Aboyeji et al. (2020)
	<i>A. niger</i>	Cashew juice	240	92.8	Adeoye and Lateef (2021)
	<i>A. niger</i>	Sugarcane molasses	384	28.25	Amin et al. (2025)
Yeast	<i>Y. lipolytica</i>	Glycerol	110	35	Papanikolaou et al. (2002)
	<i>Y. lipolytica</i>	Glycerol	550	97.8	Rywińska et al. (2011)
	<i>C. parapsilosis</i>	Crude glycerol from soybean biodiesel	168	11.3	West (2013)
	<i>C. guilliermondii</i>	Crude glycerol from soybean biodiesel	168	10.4	West (2013)
	<i>Y. lipolytica</i>	Synthetic medium and glucose	144	85	Kamzolova and Morgunov (2017)
	<i>S. cerevisiae</i>	Glucose	168	0.105	Afolabi et al. (2018)
	<i>C. tropicalis</i>	Synthetic medium and sodium nitrate	-	0.1322	Afolabi et al. (2018)

Group	Microorganism	Substrate	Time (h)	Maximum production (g L ⁻¹)	Reference
	<i>P. guilliermondii</i>	-	-	0.115	Afolabi et al. (2018)
	<i>Y. lipolytica</i>	Glycerol	144	75.9	Rzechonek et al. (2019)
	<i>Y. lipolytica</i>	Corn milling waste	552	38	Cavallo et al. (2020)
	<i>Y. lipolytica</i>	Synthetic glucose, fructose, sucrose medium	144	129	Żywicka et al. (2020)
	<i>Y. lipolytica</i>	Expired commercial glucose	136	82	Diamantopoulou et al. (2023)
	<i>Y. lipolytica</i>	Corn straw hydrolysate	216	83.6	Lu et al. (2025)

Source: prepared by the authors, 2026.

Table 3
Citric acid production by solid-state fermentation by different microorganisms

Group	Microorganism	Substrate	Time (h)	Maximum production (g Kg ⁻¹)	Reference
Filamentous Fungi	A. niger	Apple pomace with 4% methanol	120	883	Hang and Woodams (1984)
	A. niger	Apple pomace	120	90	Hang and Woodams (1986)
	A. niger	Kiwi peel	96	100	Hang et al. (1987)
	A. niger	Soybean residue	264	51	Khare et al. (1995)
	A. niger	Sugarcane bagasse	120	48.7	Vandenberghe et al. (2000)
	A. niger	Cassava bagasse	120	88	Vandenberghe et al. (2000)
	A. niger	Coffee husk	120	12.7	Vandenberghe et al. (2000)
	A. niger	Apple pomace	96	124	Shojaosadati and Babaeipour (2002)
	A. niger	Cassava bagasse	144	269	Prado et al. (2005)
	A. niger	Treated sugarcane bagasse	120	137.6	Khosravi-Darani and Zoghi (2008)
	A. niger	Oil palm empty fruit bunches	168	337.94	Bari et al. (2009)
	A. niger	Pineapple waste	120	60.61	Kareem et al. (2010)
	A. niger	Apple pomace	120	46	Kumar et al. (2010)
	A. niger	Oil palm empty fruit bunches	168	367.40	Alam et al. (2011)
	A. niger	Orange peel	85	193.2	Torrado et al. (2011)
	A. niger	Apple pomace	72	128	Dhillon et al. (2011)

Group	Microorganism	Substrate	Time (h)	Maximum production (g Kg ⁻¹)	Reference
	A. niger	Brewery waste	72	14	Dhillon et al. (2011)
	A. niger	Peat	72	31.6	Dhillon et al. (2011)
	A. niger	Tung cake with glycerol	168	350	Schneider et al. (2014)
	A. niger	Cassava peel	192	32.7	Nwosu and Kareem (2015)
	A. niger	Lemon peel	192	34.4	Nwosu and Kareem (2015)
	A. niger	Dried pomegranate peel	192	351.5	Roukas and Kotzekidou (2020)
	A. niger	Banana peel	48	124	Chysirichote (2020)
	A. niger	Treated wheat straw	120	25.6	Egbe et al. (2022)
	A. niger	Cocoa pod husks	72	978.52	Oliveira et al. (2022)
Yeast	Y. lipolytica	Pineapple waste	144	202.35	Imandi et al. (2008)

Source: prepared by the authors, 2026.

The literature survey highlights the wide diversity of studies investigating the use of agro-industrial waste as substrates for citric acid production. Generally, waste from sugarcane, apple, brewery, soybean, biodiesel, and cassava chains stands out due to high availability and application potential, especially in countries with a strong agro-industrial base, such as Brazil.

Comparative data analysis indicates that these substrates can achieve significant productivities, in some cases comparable to or higher than those obtained with conventional substrates, particularly when used in optimized processes or associated with pretreatments. It is also observed that using these residues contributes not only to process cost reduction but also to environmental impact mitigation by promoting byproduct utilization and integrating citric acid production into a circular economy perspective.

4.1. Sugarcane Residues

Among the evaluated substrates, sugarcane-derived residues stand out as one of the primary raw materials for citric acid production in both submerged and solid-state fermentation. This prominence is directly related to the high availability of these residues in countries with strong sugar-alcohol production, as well as their composition rich in sugars and organic compounds that can be metabolically converted by producing microorganisms.

According to the literature survey, sugarcane molasses has been widely used in studies focused on optimizing citric acid production by *Aspergillus niger*, mainly through obtaining mutant strains. Strategies such as gamma radiation mutagenesis (Islam et al. 1986; Majumder et al. 2010) and ultraviolet radiation (Iqbal et al. 2015; Ikram-ul et al. 2004; Lotfy et al. 2007) have been employed to increase the metabolic flux directed toward citric acid biosynthesis. The predominance of these approaches is directly related to the fact that the *A. niger*–sugarcane molasses system represents the consolidated industrial standard, so studies focus on improving established processes.

Among the studies using sugarcane molasses as a substrate in submerged fermentation, Ikram-UL, Ali, et al. and Lotfy, Ghanem, and Elhelow stood out with high yields of 96.1 g L⁻¹ and 98.08 g L⁻¹, respectively (Ikram-ul et al. 2004; Lotfy et al. 2007). Meanwhile, the studies by Majumder, Khalil et al. and Iqbal, Haq, et al. achieved lower results of 14.86 g L⁻¹ and 14.17 g L⁻¹, respectively (Majumder et al. 2010; Iqbal et al. 2015). However, the study by Majumder, Khalil, et al. stands out by showing that joining substrates can favor the process, as sugarcane molasses used as a single substrate provided a production of 4.88 g L⁻¹, while a productivity of 14.86 g L⁻¹ was obtained from the substrate mixture (Iqbal et al. 2015; Majumder et al. 2010).

The study by Amin et al. (2025), which does not employ mutation techniques, presented a productivity of 28.25 g L⁻¹, an intermediate value compared to systems with mutant strains. This result indicates that while mutagenesis is an effective strategy for maximizing production, approaches based on optimizing cultivation conditions and medium formulation can also result in relevant performances. Thus, improvements in citric acid production from sugarcane molasses can be achieved through different strategies, with the choice depending on the balance between operational complexity, cost, and productivity gain (Amin et al. 2025).

Table 3 shows the literature survey of citric acid production by solid-state fermentation (SSF), where sugarcane bagasse was used as a substrate in the studies by Vandenberghe et al. and Khosravi-Darani and Zoghi (Vandenberghe et al. 2000; Khosravi-Darani and Zoghi 2008).

Vandenberghe et al. compared the citric acid production potential of three agro-industrial residues—sugarcane bagasse, cassava bagasse, and coffee husk—reaching maximum productions of 48.7 g Kg⁻¹, 88 g Kg⁻¹, and 12.7 g Kg⁻¹, respectively, with the fungus *A. niger* (Vandenberghe et al. 2000). The high productivity of cassava bagasse in this study compared to the other substrates can be explained by the presence of components such as calcium, phosphorus, and vitamin B2 Vandenberghe et al. (2000).

Khosravi-Darani and Zoghi also used *A. niger* as the producing microorganism. Their study compared citric acid production using pretreated sugarcane bagasse (acid, alkaline, or urea) with untreated bagasse. Urea pretreatment proved to be the most effective option, obtaining a production of 137.6 g Kg⁻¹ at the laboratory scale. Once optimal production conditions were obtained, the experiment was repeated at scale using a 20L bioreactor; under these conditions, the maximum citric acid production was 82.38 g Kg⁻¹.

Consequently, sugarcane residues exhibit high versatility as substrates, being widely applicable in different process configurations. While molasses is consolidated as a high-efficiency substrate in submerged systems, bagasse and other solid residues represent promising alternatives in solid-state fermentation processes, especially when integrated into waste valorization and circular economy strategies (West 2023).

4.2. Brewery Waste

Eighty percent of the waste generated in breweries comes from the extraction of malt grains for wort production. Its composition is rich in fibers and proteins (Skend et al. 2018; Mathias et al. 2014). In biotechnology, residual malt can be used in the production of the medicinal fungus *Herichium erinaceus* (Wolters et al. 2016), lactic acid (Liguori et al. 2015), and lipids (Patel et al. 2018).

In citric acid production, brewery waste has been used in both types of fermentation, primarily using *Aspergillus niger* as the producing microorganism. In solid-state fermentation, Dhillon, Brar et al. (2011) reported a productivity of 14.0 g kg⁻¹, while in submerged fermentation Roukas and Kotzekidou (1986) obtained 19.0 g L⁻¹ using the same substrate. Additionally, Hang, Splittstoesser et al. (1977) investigated the use of this residue in submerged fermentation with *Aspergillus foetidus*, reaching a production of 12.3 g L⁻¹ (Dhillon et al. 2011; Hang et al. 1977; Roukas and Kotzekidou 1986).

In the study by Pathania et al. (2018), citric acid production from brewery residue via solid-state fermentation showed maximum values of approximately 0.23% (w/w). The results indicate that despite process viability, the low concentration of fermentable sugars in the substrate significantly limits productivity compared to conventional substrates, highlighting the importance of medium supplementation with nitrogen sources and potassium dihydrogen phosphate to increase yield (Pathania et al. 2018).

The results indicate that although brewery waste shows potential as a substrate for citric acid production, the observed productivities are lower than those obtained with substrates richer in fermentable sugars, such as molasses or starch hydrolysates. This performance may be associated with the lower availability of easily assimilable carbohydrates and the presence of structural fractions that limit the microorganism's metabolic access, indicating the need for pretreatments or medium supplementation for process optimization.

4.3. Soybean Waste

Residues derived from the soybean industry, such as soybean cake and okara, show potential as substrates in biotechnological processes due to their composition rich in proteins, amino acids, and carbohydrates. These materials have been explored in the production of different bioproducts, including organic acids and enzymes, through fermentative processes (Li et al. 2012; Andrade et al. 2014; Uzun et al. 2006).

In citric acid production, soybean cake was explored in the study by Ekeleme, Makut et al., where it was used along with glucose to obtain a citric acid production of 15.17 mg/mL through submerged fermentation using the fungus *Trichoderma viride*. This study showed that high sugar concentrations can decrease citric acid production by generating an unwanted amount of residual sugars (Ekeleme et al. 2018).

Makut and Ekeleme used potato peels as a substrate and soybean cake as a supplementary nitrogen source. *Aspergillus niger* and *Trichoderma viride* were used as producing microorganisms in SmF, obtaining a maximum citric acid production rate of 0.40453 g L⁻¹ with *Aspergillus niger* and 0.2976 g L⁻¹ with *Trichoderma viride*, indicating that process efficiency depends heavily on medium formulation and the interaction between the substrates used (Ekeleme et al. 2018; Makut and Ekeleme 2018).

Soybean biodiesel was used along with glycerol as a substrate in SmF in the study by West (2013). This was the only study found using the yeast *Candida parapsilosis* as the producing microorganism. The maximum citric acid production obtained was 11.3 g L⁻¹ West (2013).

Khare, Jha, and Gandhi (1995) analyzed the potential use of Okara residue, a byproduct of the soy milk and tofu industry, as a substrate. Again, *Aspergillus niger* was used in SSF, achieving a production efficiency of 51 g kg⁻¹ (Khare et al. 1995).

Soybean residues show potential for application in fermentative processes; however, yields vary significantly depending on substrate composition, the need for nutritional supplementation, and the microorganism employed, highlighting the importance of optimizing cultivation conditions to enable industrial-scale application. Additionally, there is a limitation of recent studies specifically focused on using these residues for citric acid production, indicating a need for further investigation to explore their potential in this context more deeply.

4.4. Biodiesel Production Waste

Glycerol (C₃H₈O₃), also known as glycerin, is a byproduct generated on a large scale during biodiesel production. Some studies identify glycerol as a carbon source for filamentous fungi species, such as *A. niger* and *B. ribis* EC-01, in the production of enzymes and oxalic acid. Among the surveyed studies, glycerol proved to be an effective and widely used substrate for fermentation by the yeast *Yarrowia lipolytica*.

Conventional citric acid production, which utilizes *Aspergillus niger* and sugarcane molasses, presents environmental impacts related to the volume and characteristics of the generated effluent. This effluent

contains unfermented substrate fractions, residual sugars, fungal mycelium, metabolic byproducts (such as oxalic acid), and residues from purification steps, including activated carbon, hexacyanoferrate compounds, and heavy metals. Additionally, production with *A. niger* is associated with the emission of potentially allergenic spores, posing environmental and occupational risks (Kamzolova 2023; Cavallo et al. 2017; Latif et al. 2023; Sales 2009; Igliński et al. 2022).

In this context, the yeast *Yarrowia lipolytica* has emerged as a promising alternative for citric acid production, offering advantages such as higher tolerance to different substrates (including industrial waste like glycerol) and lower sensitivity to impurities. Furthermore, *Y. lipolytica* does not form spores, reducing occupational health risks, and may require fewer pretreatment steps. Another relevant aspect is its ability to operate under less restrictive cultivation conditions, contributing to the development of more sustainable processes (Cavallo et al. 2017; Kamzolova 2023; Papanikolaou and Aggelis 2010; Carsanba et al. 2019).

Studies by Papanikolaou et al., Rywińska et al., and Rzechonek et al. used *Y. lipolytica* in submerged fermentation (SmF) and achieved maximum productions of 35 g/L, 97.8 g/L, and 75.9 g/L, respectively (Papanikolaou et al. 2002; Rywińska et al. 2011; Rzechonek et al. 2019). Also in submerged fermentation, glycerol was utilized by West using various *Candida* species. The maximum citric acid production obtained was 11.3 g/L using *C. parapsilosis* and 10.4 g/L using *C. guilliermondii* (West 2013).

In solid-state fermentation (SSF), glycerol was used as an additional carbon source for tung cake (Schneider et al. 2014). The addition of 20% glycerol to the tung cake yielded 350 g/L. This study also indicates that substrate excess can hinder fungal metabolism, as lower production rates were obtained with 40% glycerol (Schneider et al. 2014). This negative effect caused by glycerol excess was also observed in studies by Chatzifragkou et al. and Cremonese (Chatzifragkou et al. 2011; Cremonese 2012). Cremonese (2012) reported an increase in citric acid production yield in solid-state fermentation after adding different proportions (10%, 20%, and 40%) of glycerol to castor and tung cake. The optimal yield of 35% was obtained with the addition of 20% glycerol to castor cake. Lipase production was also positively affected by glycerol addition (Cremonese 2012). Studies by Legiša and Matthey relate glycerol production by the fungus *Aspergillus niger* to its capacity to accumulate citric acid (Legiša and Matthey 1986a; Legiša and Matthey 1986b).

4.5. Cassava Waste

The peel and bagasse residues generated during the cassava starch extraction process possess a composition rich in fiber and carbon, making them suitable as substrates for citric acid production (Vandenberghe et al. 2000; Pondja et al. 2017).

Analyzed studies indicate that cassava residues are effective substrates in both solid-state fermentation (SSF) and submerged fermentation (SmF). They often exhibit yields superior to those observed for sugarcane residues; for example, Vandenberghe et al. (2000) utilized the microorganism *A. niger* to

obtain a productivity of 48.7 g/kg using sugarcane bagasse, compared to 88 g/kg using cassava bagasse.

In submerged fermentation, a study by Adeoye, Lateef, and Gueguim-Kana showed that cassava peel can be equated to sugarcane molasses for citric acid production. In this study, citric acid production by *A. niger* strains mutated by ultraviolet radiation reached 88.7 g/L , a result similar to other studies using molasses as a substrate (Adeoye et al. 2015).

Xu et al. proposed an integrated cleaner production approach based on the reuse of treated effluents from citric acid production itself. After anaerobic digestion and electrodialysis treatment, the effluent was reused in the process, resulting in an average production of 142.4 g/L —a value similar to that obtained with conventional water (141.6 g/L). These results represent some of the highest reported values for submerged fermentation among the analyzed studies (Xu et al. 2016).

Conversely, studies such as Ajala et al. (2020) demonstrate that process efficiency can vary significantly, with yields of 1.82 g/L after 48 hours, highlighting the influence of operational conditions.

In solid-state fermentation, Prado et al. (2005) evaluated citric acid production using *Aspergillus niger* at a semi-pilot scale, comparing different bioreactor configurations. The horizontal drum reactor showed a production of 269 g/kg in 144 hours, while the tray reactor reached 263 g/kg in 120 hours, the latter being considered more advantageous regarding energy consumption and operational cost. Furthermore, Nwosu et al. (2015) investigated the viability of cassava and lemon peels as substrates, reaching maximum productions of 32.7 g/kg for cassava peel and 34.4 g/kg for lemon peel.

Overall, studies show that cassava residues stand out as highly promising substrates, presenting higher productivities than other agro-industrial wastes. This performance is linked to a composition rich in easily assimilable carbohydrates, favoring microbial metabolism and product accumulation.

4.6. Apple Waste

Apple residues, particularly the bagasse resulting from juice and cider production, stand out as promising substrates due to their high total sugar content (including glucose, fructose, and sucrose) and organic compounds that favor microbial growth (Vendruscolo et al. 2008; Shalini and Gupta 2010).

This literature survey observed that apple residues are more frequently associated with solid-state fermentation than submerged fermentation. For submerged fermentation, only the study by Dhillon et al. (2011) was found, which achieved a production of 9 g/L using apple wash effluents and the fungus *A. niger*. This limitation in liquid media is largely related to sugar dilution during aqueous extraction and the presence of potentially inhibitory trace elements such as iron and manganese. In contrast, SSF performs better by simulating the natural habitat of filamentous fungi.

Studies by Hang and Woodams (Hang and Woodams 1984; Hang and Woodams 1986) demonstrate a strong influence of media supplementation on productivity. While in natura apple bagasse resulted in a production of 90 g/kg , the addition of 4% methanol raised this value to 883 g/kg one of the highest values observed in this survey. This effect is associated with altered cell permeability and metabolic modulation.

However, productivity can be optimized even without chemical stimulants through rigorous control of physical parameters. Shojaosadati and Babaeipour (2002) demonstrated that by adjusting moisture content to 75% and temperature to 30°C , a yield of 124 g/kg could be achieved using *A. niger* in SSF. This underscores the importance of Response Surface Methodology to identify operational conditions that minimize metabolic stress.

4.7. Synthetic Media

Synthetic media are also used in production optimization studies. Pastore, Hasan, and Zempulski (2011) supplemented Prescott and Dunn's synthetic medium with nitrogen sources (urea, peptone, and ammonium sulfate) and sucrose, showing that the association of nitrogen sources increases fermentation yield.

Kamzolova and Morgunov (2017) also studied nutrient availability for the yeast *Y. lipolytica*. Unlike the previously mentioned study, nitrogen deficiency in *Y. lipolytica* cultivation favored citric acid accumulation. Additionally, Żywicka et al. (2020) used synthetic media to show that yeasts immobilized in bacterial cellulose had a higher production potential than free yeasts.

Diamantopoulou et al. (2023) evaluated the use of expired commercial glucose as a substrate, with the strain *Y. lipolytica* ACA-DC 50109 reaching 82 g/L in a fed-batch bioreactor. This highlights the viability of transforming waste into a high-performance carbon source.

The lowest production efficiency found in this survey for SmF was in the study by Afolabi, Adeyemo, and Balogun (2018), which obtained 0.105 g/L using *Saccharomyces cerevisiae* on synthetic glucose medium, indicating that certain microorganisms are simply not effective for citric acid accumulation.

4.8. Other Residues

In addition to the substrates discussed individually, the literature review highlights the use of various other agro-industrial residues in citric acid production, albeit with lower frequency or a smaller volume of available data. Notable among these are beet residue (Ozdal and Basara 2019; Lotfy et al. 2007), pumpkin (Majumder et al. 2010), dairy (Dhillon et al. 2011; Magalhães et al. 2019), dates (Mostafa and Alamri 2012), rice (Kudzai et al. 2016), potato (Kudzai et al. 2016; Ekeleme et al. 2018; Aboyeji et al. 2020; Makut and Ekeleme 2018), corn (Meng et al. 2020; Lu et al. 2025; Zhou et al. 2017; Cavallo et al. 2020), poultry waste (Ozdal and Basara 2019), coffee (Vandenberghe et al. 2000), oil palm (Bari et al. 2009; Alam et al. 2011), pineapple (Imandi et al. 2008; Kareem et al. 2010), orange (Torrado et al. 2011),

peat (Dhillon et al. 2011), pomegranate (Roukas and Kotzekidou 2020), kiwi (Hang et al. 1987), banana (Chysirichote 2020), cashew (Adeoye and Lateef 2021), wheat (Egbe et al. 2022), and cocoa (Oliveira et al. 2022), the latter achieving the highest production value found among all submerged fermentation studies at 978.52 g/kg.

The literature survey conducted shows that these materials possess potential for application in fermentative processes, especially when used as complementary substrates or in conjunction with nutritional supplementation strategies.

4.9. Evolution of Trends and Future Perspectives

The analysis of the temporal trajectory of the research compiled in this review reveals a shift in the studied strategies for citric acid production. In the pioneering studies of the 1970s and 1980s, research focused primarily on demonstrating the technical feasibility of producing citric acid from simple, widely available residues. Filamentous fungi, specifically *Aspergillus niger*, were predominant. Studies such as those by Hang et al. (1977) and Hang and Woodams (Hang and Woodams 1984; Hang and Woodams 1986) evaluated the technical viability of residues like apple bagasse and brewery spent grain, utilizing *Aspergillus niger* and chemical stimulants (such as methanol) to prove that production from waste was viable.

In the 1990s and early 2000s, the literature began to incorporate a broader range of agro-industrial residues and explored them comparatively. During this period, residues such as okara, sugarcane bagasse, cassava bagasse, kiwi peel, and coffee husks were evaluated. Solid-state fermentation (SSF) also gained ground as a promising alternative. Additionally, a sophistication in optimization tools was observed; the study by Vandenberghe et al. (2000) marked a phase of systematic residue comparison, while Shojaosadati and Babaeipour (2002) utilized Response Surface Methodology to extract the intrinsic potential of substrates without the need for toxic additives.

Between the mid-2000s and the 2010s, a phase of technological intensification occurred. During this interval, research explored not only the viability of residues but also yield-enhancement strategies, such as ultraviolet or gamma radiation mutagenesis, nutritional supplementation, the use of methanol as a metabolic stimulant, chemical pretreatments, and comparisons between different types of bioreactors.

In the last decade, the focus has shifted toward sustainability, operational safety, and the diversification of the producing microorganism. The rise of the yeast *Yarrowia lipolytica* reflects the demand for microorganisms that do not form spores and possess greater resilience to impure substrates, such as crude glycerol and expired glucose. The most recent investigations also point toward the total integration of processes within a circular economy. Studies such as those by Xu et al. (2016), involving effluent reuse, and Amin et al. (2025), focusing on molasses optimization without mutagenesis, indicate that the future of citric acid production depends not only on raw yield but on the capacity to operate sustainable processes.

From a chronological perspective, it is also possible to observe that certain substrates remain recurrent over time, while others have emerged more recently due to industrial availability and biotechnological advancements. Sugarcane remains a historical and industrial benchmark; cassava and apple have established themselves as high-performance residues; glycerol emerges as a modern substrate associated with new microbial platforms; and residues like brewery waste and soybean appear more sporadically, generally with lower productivities. This chronological analysis highlights a trajectory of maturation in the field, evolving from feasibility studies toward integrated approaches driven by efficiency, sustainability, and industrial application potential.

5. Critical analysis: what governs citric acid yield from agro-industrial wastes

5.1. Within-substrate variability exceeds between-substrate variability

A comparison across the studies compiled in Tables 2 and 3 reveals a pattern that is rarely made explicit in the literature: for most residues, the spread of reported titres within a single substrate is wider than the spread between different substrates (Fig. 1). Cassava peel illustrates this most clearly. Using the same species, *Aspergillus niger*, in submerged fermentation, Adeoye et al. (2015) reported 88.73 g L⁻¹ whereas Ajala et al. (2020) reported 1.82 g L⁻¹ — a difference of nearly fiftyfold for a nominally identical raw material. Sugarcane molasses shows the same behaviour, ranging from 14.17 g L⁻¹ (Iqbal et al. 2015) to 98.08 g L⁻¹ (Lotfy et al. 2007). Glycerol fermented by *Yarrowia lipolytica* spans 35 g L⁻¹ (Papanikolaou et al. 2002), 75.9 g L⁻¹ (Rzechonek et al. 2019) and 97.8 g L⁻¹ (Rywińska et al. 2011).

The practical implication is that the choice of residue, taken in isolation, is a weak predictor of performance. Reported titres cannot be read as intrinsic properties of a substrate, and ranking residues by the highest value found in the literature — a common practice in the field — is therefore misleading. What the data support instead is that yield is governed principally by the process-side variables discussed below.

5.2. Inducers can dominate the substrate effect

The strongest single effect identified in this survey is the addition of low-molecular-weight alcohols. In solid-state fermentation of apple pomace, Hang and Woodams (1984) obtained 883 g kg⁻¹ with 4% methanol supplementation, against 90 g kg⁻¹ for the same substrate and species without the inducer (Hang and Woodams 1986) (Fig. 2). Independent studies on apple pomace without alcohol supplementation fall in the same lower band: 46 g kg⁻¹ (Kumar et al. 2010), 124 g kg⁻¹ (Shojaosadati and Babaeipour 2002) and 128 g kg⁻¹ (Dhillon et al. 2011). The inducer effect is thus roughly an order of magnitude, far larger than the difference between any two substrates in Table 3. Methanol and ethanol are generally understood to act by increasing membrane permeability and by suppressing competing pathways, but the reviewed studies rarely quantify this trade-off against the added cost and the

environmental burden of alcohol supplementation — an omission that matters for any process claiming a circular-economy rationale.

5.3. Pretreatment and nutrient balance

Lignocellulosic residues require hydrolysis before their carbon becomes accessible, and the reviewed data indicate that the pretreatment step, rather than the biomass itself, sets the ceiling. Khosravi-Darani and Zoghi (2008) obtained 137.6 g kg^{-1} from pretreated sugarcane bagasse, against 48.7 g kg^{-1} reported by Vandenberghe et al. (2000) for untreated bagasse. Nutrient balance operates in the same direction: Majumder et al. (2010) showed that sugarcane molasses alone yielded 4.88 g L^{-1} , rising to 14.86 g L^{-1} when combined with pumpkin as a complementary substrate, indicating that nitrogen and micronutrient limitation — not carbon availability — was constraining the process.

5.4. Research gaps

Four gaps follow directly from the body of work reviewed here. First, the absence of standardised reporting: studies express results variously as g L^{-1} , g kg^{-1} of dry solids, or percentage yield, and frequently omit the substrate loading, which makes cross-study comparison unreliable and partly explains the dispersion described in Section 5.1. Second, the near-absence of techno-economic and life-cycle assessment: although cost reduction and environmental benefit are the stated motivations for using residues, almost none of the studies surveyed quantify either claim. Third, the concentration of work at flask scale, with few reports beyond the bench; the semi-pilot comparison of bioreactor configurations by Prado et al. (2005) remains an exception rather than the norm. Fourth, the scarcity of studies on downstream recovery from waste-derived broths, which are compositionally more complex than molasses and may impose purification costs that offset the savings on the carbon source.

Addressing these gaps — particularly standardised reporting and scale-up data — is a precondition for moving the field from demonstration studies towards industrial adoption.

6. Conclusions

The growing demand for citric acid, combined with the environmental cost of disposing of agro-industrial residues, has driven a substantial body of research into alternative substrates. This review compiled 65 production records from submerged and solid-state fermentation and reached three main conclusions.

First, several residues — notably cassava peel and wastewater, corn stover and crude glycerol — have reached titres comparable to those obtained with sugarcane and beet molasses, the conventional industrial substrates. Residues are therefore technically credible feedstocks, although only a minority of the studies surveyed exceeded the conventional benchmark.

Second, the performance of a given residue is not an intrinsic property of the material. The variation observed within a single substrate class exceeded the variation between classes, and the largest effects

were attributable to inducer supplementation, pretreatment and nutrient balance rather than to substrate identity. Future work should therefore report these process variables systematically, since their omission is the main obstacle to meaningful comparison across studies.

Third, the field remains concentrated at laboratory scale. The absence of techno-economic and life-cycle assessments, of standardised reporting units and of downstream recovery studies means that the cost and sustainability advantages usually attributed to residue-based processes have yet to be demonstrated quantitatively. Addressing these gaps, together with the continued development of non-sporulating producers such as *Yarrowia lipolytica* for glycerol-rich streams, represents the most direct route from the promising results reviewed here to industrial implementation.

Declarations

Competing interests

The authors have no competing interests to declare that are relevant to the content of this article.

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Data availability

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Declaration of generative AI and AI-assisted technologies in the manuscript preparation process

During the preparation of this work, the author(s) used Claude (Anthropic) in order to assist with literature organization, reference formatting, and language editing. After using this tool/service, the author(s) reviewed and edited the content as needed and take(s) full responsibility for the content of the published article.

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Author contributions

Virgínia Sanches Coelho de Oliveira Trindade: Conceptualization, Methodology, Investigation, Writing – original draft. Milena Martins Andrade: Investigation, Writing – review and editing. Daniel Tait Vareschini: Investigation, Writing – review and editing. Nathalia Gil: Investigation, Writing – review and editing. Marcelino Luiz Gimenes: Supervision, Writing – review and editing, Funding acquisition.

References

1. Aboyebi OO, Oloke JK, Arinkoola AO, Oke MA, Ishola MM (2020) Optimization of media components and fermentation conditions for citric acid production from sweet potato peel starch hydrolysate by *Aspergillus niger*. *Scientific African*
2. Adeoye AO, Lateef A, Gueguim-Kana EB (2015) Optimization of citric acid production using a mutant strain of *Aspergillus niger* on cassava peel substrate. *Biocatalysis and Agricultural Biotechnology*
3. Adeoye AO, Lateef A (2021) Biotechnological valorization of cashew apple juice for the production of citric acid by a local strain of *Aspergillus niger* LCFS 5. *Journal of Genetic Engineering and Biotechnology*
4. Afolabi FT, Adeyemo SM, Balogun HO (2018) Fermentation Conditions and Process Optimization of Citric Acid Production by Yeasts. *The International Journal of Biotechnology* 7 (2018): 51–63
5. Ajala AS, Adeoye AO, Olaniyan SA, Fasoyin OT (2020) A study on effect of fermentation conditions on citric acid production from cassava peels. *Scientific African*
6. Alam MZ, Bari MN, Muyibi SA, Jamal P, Abdullah-Al-Mamun (2011) Development of Culture Inoculum for Scale-Up Production of Citric Acid from Oil Palm Empty Fruit Bunches by *Aspergillus niger*. *Procedia Environmental Sciences*
7. Amin A, Ahmmed R, Ismail M, Islam TN, Mohasin M (2025) optimization of citric acid production from sugarcane molasses using *aspergillus niger* by submerged fermentation. *Bioresearch Communications*
8. Andonegi M, Caba KDL, Guerrero P (2020) Effect of citric acid on collagen sheets processed by compression. *Food Hydrocolloids*
9. Andrade MM, Barbosa ADM, Dekker RFH, Rezende MI, Messias JM, Dall'antonia LH (2014) Lipase production by *Botryosphaeria ribis* EC-01 on soybean meal supplemented with amino acids, and some physicochemical properties of the enzyme. *Orbital - The Electronic Journal of Chemistry*
10. Angumeenal AR, Venkappayya D (2013) An overview of citric acid production. *LWT - Food Science and Technology* 50 (2013): 367–370
11. Bari MN, Alam MZ, Muyibi SA, Jamal P, Abdullah-Al-Mamun (2009) Improvement of production of citric acid from oil palm empty fruit bunches: Optimization of media by statistical experimental designs. *Bioresource Technology*
12. Behera BC, Mishra RM, Mohapatra S (2021) Microbial citric acid: Production, properties, application, and future perspectives. *Food Frontiers*
13. Berovic M, Legisa M (2007) Citric acid production. *Biotechnology Annual Review*
14. Carsanba E, Papanikolaou S, Fickers P, Agirman B, Erten H (2019) Citric Acid Production by *Yarrowia lipolytica*. *Non-conventional Yeasts: from Basic Research to Application*. by Andriy Sibirny
15. Cavallo E, Charreau H, Cerrutti P, Foresti ML (2017) *Yarrowia lipolytica*: a model yeast for citric acid production. *FEMS Yeast Research*

16. Cavallo E, Nobile M, Cerrutti P, Foresti ML (2020) Exploring the production of citric acid with *Yarrowia lipolytica* using corn wet milling products as alternative low-cost fermentation media. *Biochemical Engineering Journal*
17. Chatzifragkou A et al (2011) Biotechnological conversions of biodiesel derived waste glycerol by yeast and fungal species. *Energy*
18. Chysirichote T (2020) Valorization of Banana Peel for Citric Acid Production under Solid State Fermentation with *Aspergillus niger*. *Chemical and Biochemical Engineering Quarterly*
19. Copetti MV (2019) Fungi as industrial producers of food ingredients. *Food Science*
20. Cremonese EB (2012) Use of oleochemical byproducts for organic acid production from *Aspergillus niger*. University of Santa Cruz do Sul – UNISC, 2012: 79
21. Currie JN (1917) The citric acid fermentation of *Aspergillus niger*. *Journal of Biological Chemistry*
22. Dai J et al (2018) Citric acid as a functional supplement in diets for juvenile turbot, *Scophthalmus maximus* L.: Effects on phosphorus discharge, growth performance, and intestinal health. *Aquaculture* 495 (2018): 643–653
23. Dhillon GS, Brar SK, Verma M, Tyagi RD (2011) Utilization of different agro-industrial wastes for sustainable bioproduction of citric acid by *Aspergillus niger*. *Biochemical Engineering Journal*
24. Di Palma L, Mecozzi R (2007) Heavy metals mobilization from harbour sediments using EDTA and citric acid as chelating agents. *Journal of Hazardous Materials*
25. Diamantopoulou P, Sarris D, Tchakouteu SS, Xenopoulos E, Papanikolaou S (2023) Growth Response of Non-Conventional Yeasts on Sugar-Rich Media: Part 1: High Production of Lipid by *Lipomyces starkeyi* and Citric Acid by *Yarrowia lipolytica*. *Microorganisms*
26. Duangmal K, Worapotpisut C, Romposa N, Katemake P (2017) Uneven enzymatic browning on fresh-cut apple and its measurement. *Acta Horticulturae*
27. Egbe NE, Ihediwa L, Abdulsalami MS, Adebayo AA (2022) Citric acid Production from Agricultural Wastes using *Aspergillus niger* Isolated from some Locations within Kaduna Metropolis, Nigeria. *Journal of Applied Sciences and Environmental Management*
28. Ekeleme KI, Makut DM, Adoga PM, Tsaku AP, Nkene HI, Oti BV (2018) Production of Citric Acid by *Trichoderma viride* Isolated from Soil in Keffi, Nigeria Using Glucose Enhanced Substrates. *South Asian Journal of Research in Microbiology*
29. Feltrin VP, Sant'Anna ES, Porto ACS, Torres RCO (2000) Production of *Lactobacillus plantarum* in sugarcane molasses. *Brazilian Archives of Biology and Technology*
30. Ferreira JL et al (2020) Calcium anacardate and citric acid as growth promoters for weaned piglets. *Livestock Science* 238 (2020)
31. Gomes MG, Gurgel LVA, Baffi MA, Pasquini D (2020) Pretreatment of sugarcane bagasse using citric acid and its use in enzymatic hydrolysis. *Renewable Energy* 157 (2020): 332–341
32. Government of Brazil (2026) Brazilian Agriculture in Numbers - February 2026. Ministry of Agriculture and Livestock

33. Goyeneche R, Agüero MV, Roura S, Scala KD (2014) Application of citric acid and mild heat shock to minimally processed sliced radish: Color evaluation. *Postharvest Biology and Technology*
34. Grand View Research (2023) Citric Acid Market (2023–2030)
35. Grewal HS, Kalra KL (1995) Fungal production of citric acid. *Biotechnology Advances* 13 (1995): 209–234
36. Hang YD, Splittstosser DF, Woodams EE, Sherman RM (1977) Citric acid fermentation of brewery waste. *Journal of Food Science*
37. Hang YD, Woodams EE (1984) Apple pomace: A potential substrate for citric acid production by *Aspergillus niger*. *Biotechnology Letters*
38. Hang YD, Woodams EE (1986) Solid state fermentation of apple pomace for citric acid production. *MIRCEN Journal of Applied Microbiology and Biotechnology*
39. Hang YD, Luh BS, Woodams EE (1987) Microbial Production of Citric Acid by Solid State Fermentation of Kiwifruit Peel. *Journal of Food Science*
40. Hu P et al (2024) Citric Acid Promotes Immune Function by Modulating the Intestinal Barrier. *International Journal of Molecular Sciences*
41. Huang H, Liu C, Yang Y, Luo S (2022) Citric acid-assisted synthesis of $\text{Na}_3\text{V}_2(\text{PO}_4)_3/\text{C}$ composite as high-performance electrode material for sodium-ion batteries. *Ionics*
42. Igliński B, Kiełkowska U, Piechota G (2022) Proecological aspects of citric acid technology. *Clean Technologies and Environmental Policy*
43. Markit IHS (2015) Chemical Economics Handbook
44. Ikram-ul H, Ali S, Qadeer MA, Iqbal J (2004) Citric acid production by selected mutants of *Aspergillus niger* from cane molasses. *Bioresource Technology* 93 (2004): 125–130
45. Imandi SB, Bandaru VVR, Somalanka SR, Bandaru SR, Garapati HR (2008) Application of statistical experimental designs for the optimization of medium constituents for the production of citric acid from pineapple waste. *Bioresource Technology* 99 (2008): 4445–4450
46. IMARC (2025) Citric Acid Market Report by Application (Food and Beverages, Household Detergents and Cleaners, Pharmaceuticals, and Others), Form (Anhydrous, Liquid), and Region 2026–2034. <https://www.imarcgroup.com/citric-acid-manufacturing-plant>
47. Iqbal J et al (2015) Isolation of *Aspergillus niger* Strains from Soil and their Screening and Optimization for Enhanced Citric Acid Production Using Cane Molasses as Carbon Source. *Journal of Applied Environmental and Biological Sciences* 4 (2015): 128–137
48. Islam MS, Begum R, Choudhury N (1986) Semi-pilot scale production of citric acid in cane molasses by gamma-ray induced mutants of *Aspergillus niger*. *Enzyme and Microbial Technology* 8 (1986): 469–471
49. Kamzolova SV, Morgunov IG (2017) Metabolic peculiarities of the citric acid overproduction from glucose in yeasts *Yarrowia lipolytica*. *Bioresource Technology*

50. Kamzolova S (2023) A Review on Citric Acid Production by *Yarrowia lipolytica* yeast. Past and present Challenges and Developments. Processes
51. Kareem SO, Akpan I, Alebiowu OO (2010) Production of citric acid by *Aspergillus niger* using pineapple waste. Malaysian Journal of Microbiology
52. Katsuki H, Kim J, Kugishim M, Kamochi N, Komarneni S, Pee J (2026) Production of clays for white porcelains from porcelain stone using eco-friendly iron impurity leaching with citric acid. Applied Clay Science
53. Khajepour F, Hosseini SA (2012) Citric acid improves growth performance and phosphorus digestibility in Beluga (*Huso huso*) fed diets where soybean meal partly replaced fish meal. Animal Feed Science and Technology
54. Khare SK, Jha K, Gandhi AP (1995) Citric acid production from Okara (soy-residue) by solid-state fermentation. Bioresource Technology
55. Khosravi-Darani K, Zoghi A (2008) Comparison of pretreatment strategies of sugarcane bagasse: experimental design for citric acid production. Bioresour Technol
56. Kim SK, Kang SW, Jin SA, Ban JY, Hong SJ, Park MS (2019) Protective Effect of Citric Acid Against Hepatic Ischemia Reperfusion Injury in Sprague-Dawley Rats. Transplantation Proceedings 51 (2019)
57. Kirimura K, Yoshioka I (2019) Citric Acid. Chapter 3 in Comprehensive Biotechnology, by Murray Moo-Young, 158–165
58. Książek E (2023) Citric Acid: Properties, Microbial Production, and Applications in Industries. Molecules
59. Kudzai CT, Ajay K, Ambika P (2016) Citric acid production by *Aspergillus niger* using different substrates. Malaysian Journal of Microbiology
60. Kumar D, Verma R, Bhalla TC (2010) Citric acid production by *Aspergillus niger* van. Tieghem MTCC 281 using waste apple pomace as a substrate. Journal of Food Science and Technology
61. Latif A et al (2023) An overview of key industrial product citric acid production by *Aspergillus niger* and its application. J Ind Microbiol Biotechnol
62. Legiša M, Matthey M (1986) Glycerol synthesis by *Aspergillus niger* under citric acid accumulating conditions. Enzyme and Microbial Technology
63. Legiša M, Matthey M (1986) Glycerol as an initiator of citric acid accumulation in *Aspergillus niger*. Enzyme and Microbial Technology
64. Li B, Qiao M, Lu F (2012) Composition, Nutrition, and Utilization of Okara (Soybean Residue). Food Reviews International
65. Liguori R et al (2015) Selection of the Strain *Lactobacillus acidophilus* ATCC 43121 and Its Application to Brewers' Spent Grain Conversion into Lactic Acid. BioMed Research International
66. Lotfy WA, Ghanem KM, El-Helow ER (2007) Citric acid production by a novel *Aspergillus niger* isolate: I. Mutagenesis and cost reduction studies. Bioresource Technology 98 (2007)

67. Lu M, Sha Y, Zhang Y, Ge M, Xu Z, Jin M (2025) Efficient production of citric acid from lignocellulose hydrolysate by metabolically engineered *Yarrowia lipolytica*. *Green Chemistry*
68. Magalhães N, Cavalcante A, Siebra AL, Wanderley CRP, Marinho G, Pessoa KDAR (2019) Citric acid production by *Aspergillus niger* AN 400 from agro-industrial residue. *Sanitary and Environmental Engineering* 24 (2019): 101–107
69. Mahmoud BSM (2014) The efficacy of grape seed extract, citric acid and lactic acid on the inactivation of *Vibrio parahaemolyticus* in shucked oysters. *Food Control*
70. Majumder L, Khalil I, Munshi MK, Alam K, Begum R, Alam N (2010) Citric Acid Production by *Aspergillus niger* Using Molasses and Pumpkin as Substrates. *European Journal of Biological Sciences* 1 (2010): 1–8
71. Makut M, Ekeleme I (2018) Citric Acid Production by *Aspergillus niger* and *Trichoderma viride* Using Hydrolysed Potato Peels Substrate. *Asian Journal of Advances in Agricultural Research*
72. Mathias TRDS, Mello PPMD, Sérvulo EFC (2014) Solid wastes in brewing process: A review. *Journal of Brewing and Distilling* 5 (2014)
73. Max B, Salgado JM, Rodríguez N, Cortés S, Converti A, Domínguez JM (2010) Biotechnological production of citric acid. *Brazilian Journal of Microbiology* 41 (2010): 862–875
74. McMurry J (2016) *Organic Chemistry*, 3rd edn. Cengage Learning, São Paulo
75. Melo FDAD (2003) Contribution to the kinetic study of extracellular polysaccharide production by *Zoogloea* sp. in sugarcane molasses. Federal University of Pernambuco, Recife
76. Meng F, Liu Q, Kim R, Wang J, Liu G, Ghahrema A (2020) Selective recovery of valuable metals from industrial waste lithium-ion batteries using citric acid under reductive conditions: Leaching optimization and kinetic analysis. *Hydrometallurgy*
77. Mordor Intelligence (2025) citric acid market size & share analysis - growth trends & forecasts (2025–2030)
78. Mostafa YS, Alamri SA (2012) Optimization of date syrup for enhancement of the production of citric acid using immobilized cells of *Aspergillus niger*. *Saudi Journal of Biological Sciences*
79. Muñoz-Villa A, Sáenz-Galindo A, López-López L, Cantú-Sifuentes L, Barajas-Bermúdez L (2014) Citric Acid: Interesting Compound. *Scientific Journal of the Autonomous University of Coahuila* 6 (2014)
80. Nangare S, Vispute Y, Tade R, Dugam S, Patil P (2021) Pharmaceutical applications of citric acid. *Future Journal of Pharmaceutical Sciences*
81. Nataraj D, Sakkara S, HN M, Reddy N (2018) Properties and applications of citric acid crosslinked banana fibre-wheat gluten films. *Industrial Crops and Products*
82. Nataraj D, Reddy R, Reddy N (2020) Crosslinking electrospun poly (vinyl) alcohol fibers with citric acid to impart aqueous stability for medical applications. *European Polymer Journal*
83. Nwosu FO, Kareem AO (2015) production of citric acid from lemon and cassava waste peels. *Nigerian Journal of Materials Science and Engineering*

84. Oliveira PZD, Vandenberghe LPDS, Rodrigues C, Pereira GVDM, Soccol CR (2022) Exploring cocoa pod husks as a potential substrate for citric acid production by solid-state fermentation using *Aspergillus niger* mutant strain. *Process Biochemistry*
85. Ozdal M, Basara EK (2019) Citric Acid Production by *Aspergillus niger* from Agro-Industrial By-Products: Molasses and Chicken Feather Peptone. *Waste and Biomass Valorization* 10 (2019): 631–640
86. Papanikolaou S, Muniglia L, Chevalot I, Aggelis G, Marc I (2002) *Yarrowia lipolytica* as a potential producer of citric acid from raw glycerol. *Journal of Applied Microbiology*
87. Papanikolaou S, Aggelis G (2010) *Yarrowia lipolytica*: A model microorganism used for the production of tailor-made lipids. *European Journal of Lipid Science and Technology*
88. Pastore NS, Hasan SM, Zempulski DA (2011) Citric Acid Production by *Aspergillus niger*: Evaluation of Different Nitrogen Sources and Sucrose Concentrations. *Engevista*
89. Patel A, Mikes F, Bühler S, Matsakas L (2018) Valorization of Brewers' Spent Grain for the Production of Lipids by Oleaginous Yeast. *Molecules*
90. Pathania S, Sharma S, Sharma K (2018) Solid state fermentation of BSG for citric acid production. *Indian Journal of Natural Products and Resources*
91. Pondja EA, Persson KM, Matsinhe NP (2017) The Potential Use of Cassava Peel for Treatment of Mine Water in Mozambique. *Journal of Environmental Protection*
92. Prado FC, Vandenberghe LPS, Woiciechowski AL, Rodríguez-León JA, Soccol CR (2005) citric acid production by solid-state fermentation on a semi-pilot scale using different percentages of treated cassava BAGASSE. *Brazilian Journal of Chemical Engineering*
93. Qiu X, Chen S, Liu G, Yang Q (2014) Quality enhancement in the Japanese sea bass (*Lateolabrax japonicas*) fillets stored at 4°C by chitosan coating incorporated with citric acid or licorice extract. *Food Chemistry*
94. Roukas T, Kotzekidou P (1986) Production of Citric Acid from Brewery Wastes by Surface Fermentation Using *Aspergillus niger*. *Journal of Food Science*
95. Roukas T, Kotzekidou P (2020) Pomegranate peel waste: a new substrate for citric acid production by *Aspergillus niger* in solid-state fermentation under non-aseptic conditions. *Environmental Science and Pollution Research*
96. Rywińska A, Juszczak P, Wojtatowicz M, Rymowicz W (2011) Chemostat study of citric acid production from glycerol by *Yarrowia lipolytica*. *Journal of Biotechnology*
97. Rzechonek DA, Dobrowolski A, Rymowicz W, Mirończuk AM (2019) Aseptic production of citric and isocitric acid from crude glycerol by genetically modified *Yarrowia lipolytica*. *Bioresource Technology*
98. Sackett D (2024) *Citric Acid: Occurrence, Biochemistry, Applications and Processing*. Nova Science Pub Inc., New York

99. Sales MDP (2009) Aspergilose: do diagnóstico ao tratamento. *Jornal Brasileiro de Pneumologia* 35 (2009)
100. Sanchís E et al (2016) Browning inhibition and microbial control in fresh-cut persimmon (*Diospyros kaki* Thunb. cv. Rojo Brillante) by apple pectin-based edible coatings. *Postharvest Biology and Technology*
101. Sawant O, Mahale S, Ramchandran V, Nagaraj G, Bankar A (2018) Fungal Citric acid production using waste materials: A mini-review. *Journal of Microbiology, Biotechnology and Food Sciences* 8 (2018)
102. Schneider M, Zimmer GF, Cremonese EB, Schneider RDCDS, Corbellini VA (2014) Byproducts from the biodiesel chain as a substrate to citric acid production by solid-state fermentation. *Waste Management & Research*
103. Shalini R, Gupta DK (2010) Utilization of pomace from apple processing industries: a review. *Journal of Food Science and Technology*
104. Shojaosadati SA, Babaeipour V (2002) Citric acid production from apple pomace in multi-layer packed bed solid-state bioreactor. *Process Biochemistry*
105. Show PL, Oladele KO, Siew QY, Zakry FAA, Lan JCW, Ling TC (2015) Overview of citric acid production from *Aspergillus niger*. *Frontiers in Life Science*
106. Skend A, Harasym J, Galanakis CM (2018) Recovery of high added-value compounds from brewing and distillate processing by-products. Chapter 7 in *Sustainable Recovery and Reutilization of Cereal Processing By-Products*, edited by Charis Michel Galanakis, 352. Chania, Greece: Woodhead Publishing
107. Soccol CR, Vandenberghe LPS, Rodrigues C, Pandey A (2006) New perspectives for citric acid production and application. *Food Technology and Biotechnology*
108. Swaggerty CL et al (2020) A microencapsulated feed additive containing organic acids, thymol, and vanillin increases in vitro functional activity of peripheral blood leukocytes from broiler chicks. *Poultry Science*
109. Torrado AM et al (2011) Citric acid production from orange peel wastes by solid-state fermentation. *Brazilian Journal of Microbiology*
110. Troiano D, Orsat V, Dumont MJ (2020) Status of filamentous fungi in integrated biorefineries. *Renewable and Sustainable Energy Reviews*
111. Uzun BB, Pütün AE, Pütün E (2006) Fast pyrolysis of soybean cake: Product yields and compositions. *Bioresource Technology*
112. Vandenberghe LPS, Soccol CR, Pandey A, Lebeault JM (2000) Solid-state fermentation for the synthesis of citric acid by *Aspergillus niger*. *Bioresource Technology*
113. Vargas DA (2012) and Josephine V. Medina. *Citric Acid: Synthesis, Properties and Applications*. Nova Science Pub Inc., New York
114. Vendruscolo F, Albuquerque PM, Streit F, Esposito E, Ninow JL (2008) Apple Pomace: A Versatile Substrate for Biotechnological Application. *Critical Reviews in Biotechnology*

115. Wang X et al (2015) Heavy metal chemical extraction from industrial and municipal mixed sludge by ultrasound-assisted citric acid. *Journal of Industrial and Engineering Chemistry*
116. Wang S, Xu C, Yu S, Wu X, Jie Z, Dai H (2019) Citric acid enhances the physical properties, cytocompatibility and osteogenesis of magnesium calcium phosphate cement. *Journal of the Mechanical Behavior of Biomedical Materials*
117. West TP (2013) Citric acid production by *Candida* species grown on a soy-based crude glycerol. *Preparative Biochemistry and Biotechnology*
118. West TP (2023) Citric Acid Production by *Aspergillus niger* Using Solid-State Fermentation of Agricultural Processing Coproducts. *Applied Biosciences*
119. Wolters N, Schabronath C, Schembecker G, Merz J (2016) Efficient conversion of pretreated brewer's spent grain and wheat bran by submerged cultivation of *Herichium erinaceus*. 222 (2016): 123–129
120. Xu J et al (2016) A novel cleaner production process of citric acid by recycling its treated wastewater. *Bioresour Technol*
121. Yahia EM (2011) *Postharvest Biology and Technology of Tropical and Subtropical Fruits*
122. Yu Z et al (2017) Enhancing phosphate adsorption capacity of SDS-based magnetite by surface modification of citric acid. *Applied Surface Science*
123. Zhang YL, Zhao CX, Liu XD, Li W, Wang JL, Hu ZG (2016) Application of poly(aspartic acid-citric acid) copolymer compound inhibitor as an effective and environmental agent against calcium phosphate in cooling water systems. *Journal of Applied Research and Technology*
124. Zhang D, Xu W, Cai J, Cheng SY, Ding WP (2020) Citric acid-incorporated cellulose nanofibrous mats as food materials-based biosorbent for removal of hexavalent chromium from aqueous solutions. *International Journal of Biological Macromolecules*
125. Zhao S et al (2019) Citric acid mitigates soybean meal induced inflammatory response and tight junction disruption by altering TLR signal transduction in the intestine of turbot. *Scophthalmus maximus* L. *Fish and Shellfish Immunology*
126. Zhou P, Meng J, Bao J (2017) Fermentative production of high titer citric acid from corn stover feedstock after dry dilute acid pretreatment and biodegradation. *Bioresource Technology*
127. Żywicka A, Junka A, Ciecholewska-Juśko D, Migdał P, Czajkowska J, Fijałkowski K (2020) Significant enhancement of citric acid production by *Yarrowia lipolytica* immobilized in bacterial cellulose-based carrier. *J Biotechnol*

Figures

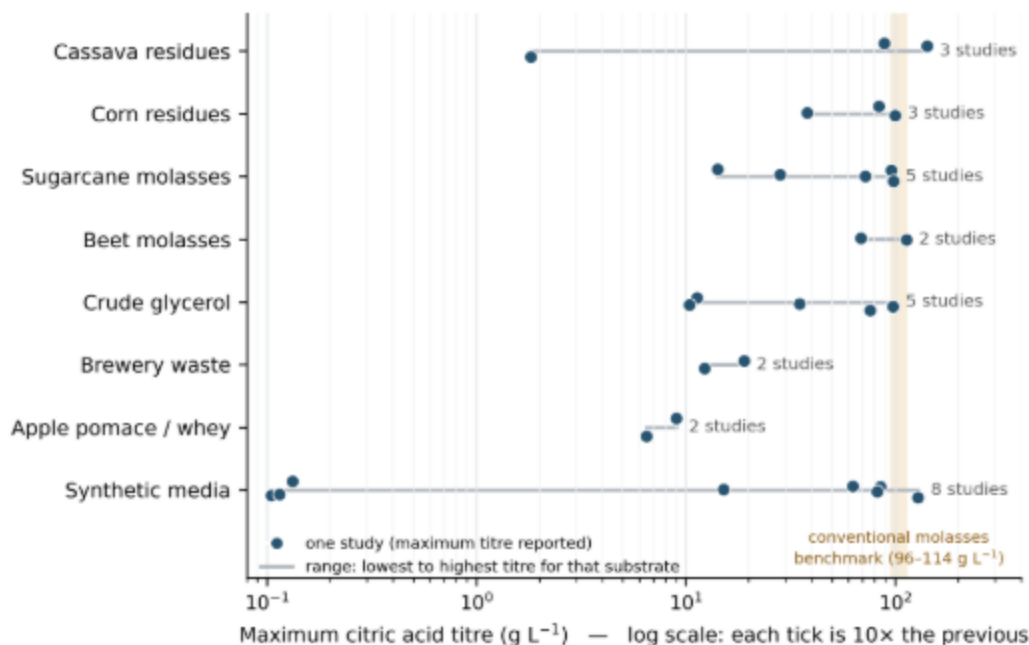


Figure 1

Maximum citric acid titres reported for submerged fermentation, grouped by substrate class (data from Table 2, log scale). Each point represents one study; the grey line connects the lowest and highest titre reported for that substrate. The shaded band marks the range obtained with conventional molasses (96–114 g L⁻¹).

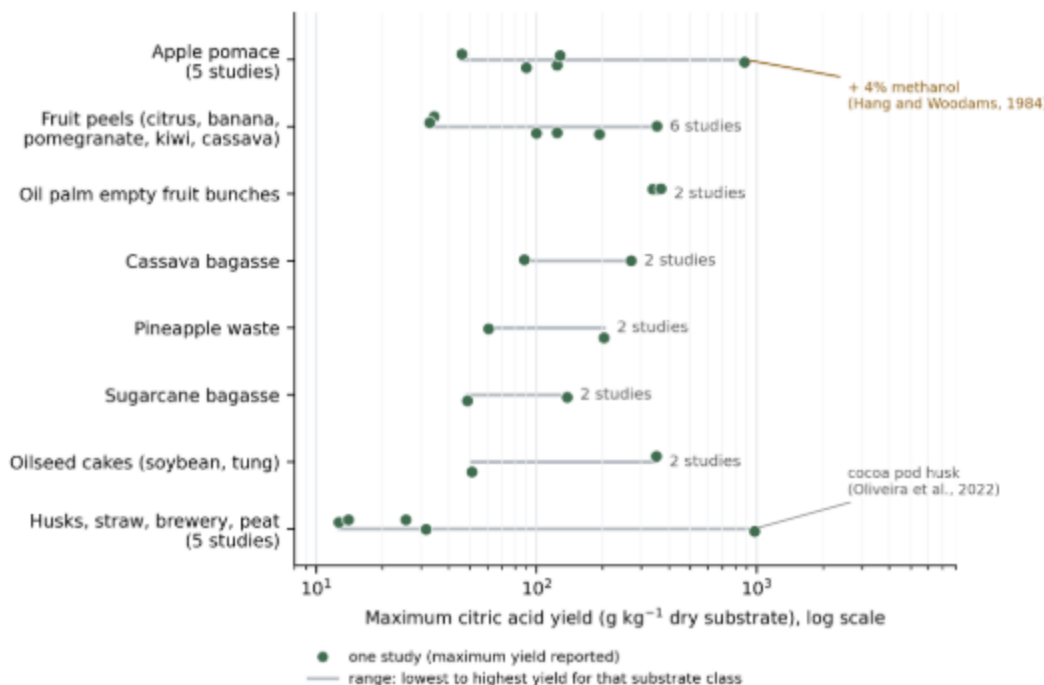


Figure 2

Maximum citric acid yields reported for solid-state fermentation, grouped by substrate class (data from Table 3, log scale). Each point represents one study; the grey line connects the lowest and highest yield reported for that class. Annotated points identify apple pomace with 4% methanol (Hang and Woodams 1984) and cocoa pod husk (Oliveira et al. 2022).