

## An Overview of Microbial Production of Lactic Acid from Molasses

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**ABSTRACT:** Lactic acid is a versatile organic acid with a wide range of industrial applications, spanning from the food and pharmaceutical sectors to biodegradable plastics and environmental remediation. This review paper aims to provide detailed information about lactic acid, encompassing its production, properties, and applications. The microbial production of lactic acid from molasses offers a promising and eco-friendly alternative to traditional chemical synthesis methods. As technology and bioprocesses continue to advance, this approach is poised to become increasingly competitive and sustainable, effectively addressing the growing demand for lactic acid across various industries. In recent years, microbial lactic acid production has garnered significant attention due to its sustainability and cost-effectiveness. Molasses, a byproduct of the sugar industry, serves as an abundant and cost-effective substrate for lactic acid production. This overview also delves into key aspects of microbial lactic acid production from molasses, including the selection of microorganisms, fermentation processes, optimization strategies, and downstream processing. It highlights both the advantages and

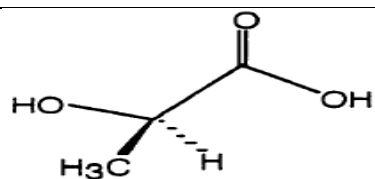
challenges associated with this biotechnological approach, emphasizing its potential as a sustainable and eco-friendly pathway for lactic acid production.

This study underscores the growing significance of microbial lactic acid production from molasses as a promising alternative to traditional chemical synthesis, with wide-reaching implications for various industries and environmental sustainability. Additionally, this paper explores the physical and microbial production of lactic acid from sucrose-derived raw materials, with molasses emerging as the most cost-effective choice

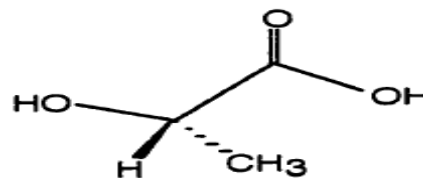
**KEYWORDS:** Lactic acid, Lactic, Molasses, microbial production, Cheap

## 1. Introduction

Lactic acid is a natural organic acid with a wide array of applications in the pharmaceutical, chemical, food, and healthcare industries and holds significant importance in pharmaceutical preparations, electroplating, the leather industry, and the pork industry, production of polypropylene oxide and biodegradable polylactic acid<sup>1-4</sup>. The fermentation process ensures the rapid and reliable generation of lactic acid, representing one of the earliest microbial products in history<sup>5</sup>. Recent years have witnessed a surge in interest in lactic acid production from biomass through fermentation process which brings garners attention due to its advantages over chemical synthesis<sup>6-8</sup>. In recent years, much work has been carried out on the optimization of lactic acid production from different biomass sources<sup>9-14</sup>. Lactic acid is an organic acid. It is 2-hydroxypropionic acid and occurs in two isomeric forms, D or L, namely, (L-(+)-LA and D-(-)-LA) as shown below



D-(-)lactic acid



L-(+) lactic acid

**2. Properties of Lactic acid:** Physical and chemical properties of lactic acid have been incorporated in Table 1.

### Properties of Lactic acid<sup>15-22</sup>

Property	Value or Description
Formula	CH <sub>3</sub> CH(OH)COOH
Molecular weight	90.08 g/mol
Odour	Odourless
Odor Threshold	The odor threshold for lactic acid is low, and it can be detected at low concentrations.
Taste	Mild acid taste
Toxicity	Oral rat LD <sub>50</sub> . 3543mg/kg
Synonyms	2-hydroxypropanoic acid, 1-hydroxyethanecarboxylic acid Ethylidenelactic acid, Alph hydroxypropionic acid
Physical state	Colourless to slightly yellow, syrupy liquid
Melting point	17°C
Boiling point	122 °C
Specific gravity	1.2
Solubility in water	Miscible
NFPA rating	Health3, Flamability 1, Reactivity 1
Flash point	112 °C
Stability	Stable under ordinary conditions
K <sub>a</sub>	1.38 x 10 <sup>-4</sup>
pK <sub>a</sub>	Lactic acid has two pK <sub>a</sub> values: 3.86 and 5.12, reflecting its two ionizable hydrogen atoms. The pK <sub>a</sub> values can change depending on concentration
Appearance	Colorless or slightly yellow liquid

State at Room Temperature	Liquid
Density	1.209 g/cm <sup>3</sup> (at 20 °C)
Solubility	Soluble in water, ethanol, and ether
Hygroscopicity	Absorbs moisture from the air
pH	2.4 (for 1 M solution)
Specific Gravity	1.21 - 1.22 (at 20 °C)
Viscosity	1.21 cP (at 25 °C)
Heat of Vaporization	42.8 kJ/mol
Freezing Point	Approximately 53 °C (127 °F) (for 80% solution)
Autoignition Temperature	Not applicable (lactic acid does not autoignite)
Electrical Conductivity	Lactic acid is a weak electrolyte and has low electrical conductivity in its pure form.
Color	Lactic acid is typically colorless, but it may appear slightly yellow in impure or concentrated forms.
Heat of Formation	Approximately -694 kJ/mol
Heat Capacity (C <sub>p</sub> )	Approximately 2.62 J/(g·K) at 25 °C
Critical Temperature	Approximately 242 °C (467.6 °F)
Partition Coefficient (log P)	Around -0.57 (n-octanol/water)
Surface Tension	Approximately 49 mN/m (at 25 °C)
Solubility in Organic Solvents	Lactic acid is soluble in various organic solvents such as ethanol, acetone, and ethyl acetate.
Heat of Solution	-55 kJ/mol (when dissolved in water)
Vapor Pressure (at 20 °C)	Very low, typically less than 1 mmHg
Octanol-Water Partition Coefficient (log P)	Approximately -0.67
Optical Activity	Lactic acid exists in two optical isomers, L-lactic acid and D-lactic acid, each with different optical activity. The optical rotation depends on the isomer and its concentration.
Specific Heat Capacity	Approximately 2.44 J/g°C (for L-lactic acid)
Combustibility	Lactic acid is not considered highly flammable, but it can support combustion under certain conditions.
Refractive Index	Approximately 1.42 (for pure lactic acid)
Solubility in Ether	Lactic acid is soluble in ether.
Solubility in Acetone	Lactic acid is soluble in acetone.
Surface Density	Approximately 7.2 mg/cm <sup>2</sup>
Solubility in Ethanol	Lactic acid is soluble in ethanol.

Optical Rotation	The optical rotation of lactic acid varies based on the specific optical isomer (L-lactic acid or D-lactic acid) and its concentration. It exhibits optical activity.
Standard Enthalpy of Formation	Approximately -694.6 kJ/mol
Partition Coefficient (log Kow)	Approximately -1.02 (for n-octanol/water)
Molecular Geometry	Lactic acid has a pyramidal geometry around the carbon atom.
Standard Gibbs Free Energy of Formation	Approximately -564.70 kJ/mol (at 298.15 K and 1 bar)
Magnetic Susceptibility	Lactic acid is diamagnetic, meaning it is not attracted to a magnetic field.
Specific Conductance	Lactic acid solutions exhibit low electrical conductivity and are weak electrolytes.
Environmental Fate	Lactic acid is biodegradable and considered environmentally friendly. It is typically broken down by microorganisms in natural systems.
Chirality	Lactic acid has chirality and exists in two optical isomers, L-lactic acid, and D-lactic acid, each with distinct optical properties.
Optical Purity	Lactic acid enantiomers may be used to express optical purity, often denoted as %ee (percent enantiomeric excess).
Acidity	Lactic acid is a weak acid with a pKa value of approximately 3.86 for the carboxylic acid group.
Partition Coefficient (log P)	Approximately -0.67 (for n-octanol/water)
Refractive Index	Approximately 1.42 (for pure lactic acid)
Heat of Solution	Approximately -55 kJ/mol (when dissolved in water)
Critical Density	0.22 g/cm <sup>3</sup>
Heat of Fusion	10.3 kJ/mol
Heat of Combustion	-1,265 kJ/mol (when burned)
Partition Coefficient (log Kow)	-1.02 (for n-octanol/water)
Molecular Geometry	Lactic acid has a pyramidal geometry around the carbon atom.
Standard Gibbs Free Energy of Formation	-564.70 kJ/mol (at 298.15 K and 1 bar)
Electrical Conductivity	Lactic acid is a weak electrolyte and has low electrical conductivity in its pure form.
Specific Heat Capacity (Cp)	2.42 J/(g·K) at 25 °C
Density (at 25°C)	1.206 g/cm <sup>3</sup>

Refractive Index (at 20°C)	1.42 (for pure lactic acid)
Solubility in Organic Solvents	Lactic acid is soluble in various organic solvents, such as acetone, ethyl acetate, and ethanol.
Vapor Density	Lactic acid vapor is heavier than air.
Partition Coefficient (log P)	Approximately -0.67 (for n-octanol/water).
Environmental Impact	Lactic acid is biodegradable and considered environmentally friendly as it can be broken down by microorganisms in natural systems.
Critical Pressure	Approximately 4.8 MPa (48 bar)
Heat of Solution	Approximately -55 kJ/mol (when dissolved in water)
Optical Rotation	The optical rotation of lactic acid varies based on the specific optical isomer (L-lactic acid or D-lactic acid) and its concentration. It exhibits optical activity.

**3. Carbon sources :** Lactic acid can be produced through fermentation using a variety of carbon sources. These carbon sources provide the necessary substrates for microorganisms to metabolize and convert into lactic acid. Lactic acid production from as a cheap carbon source have been reported by several authors<sup>23-25</sup> . Some of the common carbon sources for lactic acid production include:

**3.1.Glucose:** Glucose is one of the most commonly used carbon sources for lactic acid fermentation<sup>26-27</sup> . It can be derived from various feedstocks, such as corn starch, sugarcane, or cellulosic biomass. Efficient production of l-lactic acid using co-feeding strategy based on cane molasses/ glucose carbon sources<sup>28</sup> .

**3.2.Lactose:** Lactose, a sugar found in milk, can also be used as a carbon source for lactic acid production have also been reported<sup>29</sup> . Lactic acid bacteria, such as Lactobacillus, are often used for this purpose<sup>30</sup> .

**3.3.Molasses:** Recently much of the work have been carried out by using molasses, a byproduct of sugar refining processes, is rich in sugars and serves as an economical

carbon source for lactic acid fermentation. Cane molasses, a waste from sugar manufacturing processes, is hopeful to be utilized as a cheap carbon source for L-lactic acid fermentation<sup>31-33</sup>.

**3.4.Starch:** Starch from sources like corn, cassava, or potatoes can be enzymatically hydrolyzed into glucose or maltose, which can be used as a carbon source for the production of lactic acid<sup>34-36</sup>.

**3.5.Agro-industrial Residues:** Various agricultural and food processing residues, such as wheat bran, rice bran, and fruit pomace, can be used as carbon sources for lactic acid production after appropriate pretreatment and enzymatic hydrolysis. Relatively to substrate sources, worldwide there is a lot of interesting agro-industrial waste or sub-products with a lower value, which can be fermented by several organisms<sup>37-39</sup>.

**3.6.Cellulosic Biomass:** Cellulosic materials, like agricultural residues and dedicated energy crops, can be broken down into fermentable sugars, including glucose and xylose, and used for lactic acid production<sup>40-43</sup>.

**3.7.Glycerol:** Glycerol, a byproduct of biodiesel production, can serve as a carbon source for lactic acid fermentation, especially by certain strains of lactic acid bacteria<sup>44-45</sup>.

**3.8.Whey:** Whey, a byproduct of cheese and yogurt production, contains lactose and can be used as a carbon source for lactic acid production<sup>46-49</sup>.

**3.9.Lignocellulosic Biomass:** Advanced processes involving enzymatic hydrolysis can break down lignocellulosic biomass, such as wood or agricultural residues, into fermentable sugars like glucose and xylose for lactic acid production<sup>50-56</sup>.

The choice of carbon source depends on factors such as cost, availability, and the specific microorganism or fermentation process being used. Different strains of lactic acid

bacteria or other microorganisms may have varying preferences for carbon sources, and the selection of the most suitable source can significantly impact the efficiency and economics of lactic acid production.

**Table 2. The most common carbon for the production of lactic acid.**

Carbon Source	Description
Glucose	Commonly used from various feedstocks.
Lactose	Found in milk, utilized by lactic acid bacteria.
Molasses	Byproduct of sugar refining, economical source.
Starch	Hydrolyzed into glucose or maltose for use.
Agro-Industrial Residues	Residues like wheat bran, rice bran, etc.
Cellulosic Biomass	Agricultural and dedicated energy crops.
Glycerol	Byproduct of biodiesel production.
Whey	Byproduct of cheese and yogurt production.
Lignocellulosic Biomass	Requires enzymatic hydrolysis for sugars.

Molasses are by-products from sugar manufacturing, generally used as animal feed and for the production of bio-ethanol and yeasts<sup>57</sup>. Sucrose is the most abundant sugar in their composition, but due to its high concentration, the viscosity of the liquid is important, resulting in an increase of the operating costs<sup>58</sup>. The most common strain to ferment molasses is *Lactobacillus delbruecki*<sup>59</sup>. Lactic acid production from cane molasses by *Lactobacillus delbruecki* NCIM 2025 in submerged condition: optimization of medium component by Taguchi DOE Methodology<sup>60</sup>. Diptendu Sarkar et al<sup>61</sup>. and Arun Kumar et al<sup>62</sup> explored the optimization of lactic acid production by adjusting various process parameters, such as medium pH, temperature, inoculum size, incubation time, and shaking speed.

Some of the work on the production of lactic acid from molasses has been reported and summarized in Table 2.



**Table 2. Microbial production of lactic acid from molasses**

Organism	Lactic acid (g/L)	Yield g/g	Productivity g/(L/h)	Ref.
<i>Lb. delbrueckii</i> NCIMB 8130	90	0.97	3.8	63
<i>Lb. delbrueckii</i>	88	-	-	63
<i>Lb. delbrueckii</i> subsp. <i>delbrueckii</i> Mutant Uc-3	166	-	4.15	64
<i>Lb. delbrueckii</i>	107	0.9	1.48	65
<i>B. coagulans</i>	168.3	0.88	2.1	66
<i>E. faecalis</i> RKY1	95.7	-	4.0	67
<i>Lb. paracasei</i>	169.9	-	1.42	68
<i>E. coli</i>	75	0.85	1.18	69
Microbial consortium CEE-DL15 <i>Clostridium sensu stricto</i> (57.29%), <i>Escherichia</i> (34.22%), and <i>Enterococcus</i> (5.32%)	112.3	0.81	4.49	70
<i>Bacillus coagulans</i>	168.3	0.88	2.1	71
<i>Lb. delbrueckii</i> sp. <i>bulgaricus</i> AU	20	0.45	-	72
<i>Lb. delbrueckii</i> sp. <i>delbrueckii</i> ATCC 9649	26	0.58	-	72
<i>Lb. rhamnosus</i> ATCC 7469	18	0.40	-	72

#### 4. Conclusion:

The review of the physical-chemical properties and microbial production of lactic acid provides valuable insights into the versatile and important compound, lactic acid. This organic acid has a wide range of applications in various industries, including food, pharmaceuticals, and bioplastics. Microbial production of lactic acid, often referred to as fermentation, is a sustainable and economically viable method. Lactic acid bacteria, such as *Lactobacillus* and *Lactococcus* species, are commonly used for this purpose.

The choice of substrates for microbial production is diverse. These substrates can include carbohydrates derived from various sources, including agricultural crops, lignocellulosic biomass, and agro-industrial residues. The selection of feedstock affects the sustainability and cost-effectiveness of lactic acid production. To improve lactic acid yield, researchers

employ various strategies such as strain improvement, process optimization, and genetic engineering. These approaches aim to enhance production efficiency and reduce by-products. This review underscores the significance of lactic acid and its production processes in the context of green and sustainable chemistry.

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