Process Biochemistry 117 (2022) 53–67

Contents lists available at ScienceDirect

Process Biochemistry

journal homepage: www.elsevier.com/locate/procbio

Review

Integrated perspective on microbe-based production of itaconic acid: From metabolic and strain engineering to upstream and downstream strategies

Miguel Figueiredo Nascimento [a,b], Nuno Marques [a], Joana Correia [a], Nuno T. Faria [a,b,c,*], Nuno P. Mira [a,b,*], Frederico Castelo Ferreira [a,b,*]

[a] Institute for Bioengineering and Biosciences and Department of Bioengineering, Instituto Superior Técnico, Universidade de Lisboa, Portugal
[b] Associate Laboratory i4HB – Institute for Health and Bioeconomy, Instituto Superior Técnico, Universidade de Lisboa, Lisboa, Portugal
[c] Laboratório Nacional de Energia e Geologia, I.P., Unidade de Bioenergia, Estrada do Paço do Lumiar 22, Lisboa 1649-038, Portugal

A R T I C L E I N F O

Keywords:
Itaconic acid
Microbe-based production
Strain engineering
Fermentation
Integrated bioprocess
Aspergillus

A B S T R A C T

The discovery of itaconic acid as a product of citric acid pyrolytic distillation in 1837 opened the possibility of using it as a polymer building block. Itaconic acid, featuring two carboxylic acids and an unsaturated group, can potentially be used as a building block in several chemical syntheses, with a particular emphasis on polymer manufacture. The elucidation of biochemical pathways originating from itaconic acid, first in *Aspergillus terreus* and, recently, in several species of the *Ustilago* genus, has intensified and diversified research focused on microbe-based itaconic acid production, including at an industrial scale. These efforts include the engineering of naturally producing species/strains along with the exploration of other species that do not naturally produce itaconic acid but may offer potential benefits. The use of renewable wastes or sugar-enriched residues as substrates to produce itaconic acid, from a circular bioeconomy perspective, is another important aspect of the advancements in microbial itaconic acid production. This review provides an overview of the achievements as well as the challenges concerning the engineering of the producing strains/species, substrate selection, optimisation of bioreactor operation, and downstream itaconic acid purification methods.

1. Overview of “natural” producers of itaconic acid

Itaconic acid (IA; 2-methylidenebutanedioic acid) was recognised in 2004 by the United States Department of Energy as one of the top 12 molecules with a high potential market in a bio-based economy; consequently, research focused on this molecule has been increasing ever since [1]. This unsaturated dicarboxylic acid was first identified by Baup in 1837 as a by-product in the thermal decomposition of citric acid distillation [2–4]. In 2015, the market potential for IA was established in USD 74.5 million and 41,400 tons [5,6]. At that time, 44% of the global IA market was estimated to be in the production of styrene-butadiene rubber latexes. IA comprises two carboxylic groups and an unsaturated group that allow for simultaneous addition and condensation polymerisation reactions with a single monomer [7,8].

The other possible applications of IA, particularly in the production of methyl methacrylate and its derivatives, can expand its market potential [9]. This high potential of IA (and carboxylic acids in general) is attributed to the presence of multiple functional groups that participate in catalysis (chemical or enzymatic).

IA is industrially produced by microbial fermentation; however, the use of chemical synthesis has been previously explored through processes such as dry distillation of citric acid, followed by treatment of the anhydride with water [10], heating of calcium aconitate solution produced from the sugar cane refining process [11], the Montecatini method, involving propargyl chloride [11], oxidation of mesityl oxide, and isomerisation of citric acid [12–14]. However, compared to bio-based production, these chemical routes resulted in remarkably lower yields that constrained their exploration on an industrial scale [15].

The first example of a microorganism capable of producing IA was reported in 1932 by Kinoshita [16], who demonstrated that *Aspergillus itaconicus* could produce IA when cultivated on D-glucose. After this discovery, additional studies were performed for identifying other microbes with the capacity to biosynthesise IA from sugars. In 1939, Calam [17] reported *Aspergillus terreus* as a novel producer of IA [17] and the Northern Regional Research Laboratory (NRRL) of the United States

* Corresponding authors at: Institute for Bioengineering and Biosciences and Department of Bioengineering, Instituto Superior Técnico, Universidade de Lisboa, Portugal
E-mail addresses: nuno.faria@tecnico.ulisboa.pt (N.T. Faria), nuno.mira@tecnico.ulisboa.pt (N.P. Mira), frederico.ferreira@ist.utl.pt (F.C. Ferreira).

https://doi.org/10.1016/j.procbio.2022.03.020
Received 23 February 2021; Received in revised form 20 March 2022; Accepted 21 March 2022
Available online 24 March 2022
1359-5113/