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Innovation The Research Concept

Utilization of Feasible Agro-Waste Substrates for Succinic Acid by Microbial Cells

Abstract

Conventional, non-biological processes has been be replaced by biochemical conversion of biomass resulting in reduction of green house gas emissions and energy usage for the production of fuels and important chemicals. The technology uses the properties of living organisms, such as yeast, moulds, bacteria, and plants, to make products from renewable resources. When utilizing living systems the reactions involved generally occurs under mild conditions with high product specificity, hence reducing the formation of undesirable/harmful by-products. Succinic acid is found as one of very important chemical or chemical building blocks of top twelve manufactured from biomass. Succinic acid is currently used as a surfactant, detergent extender, foaming agent, in the food industry for pH reduction, as a flavour and antimicrobial agent, and in the manufacture of health products such as vitamins and pharmaceuticals. Succinic acid production from microorganisms using cheap renewable resources biomass has increasing interest with commercialization of bio-succinic acid. A novel industrial-scale bio refinery process is developing to convert corn-stover into succinic acid and co-products.

Keywords: Bio-Succinic Acid, Agro-Waste Substrates, Compound, Biologcal Process, Microorganisms.

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Introduction

Succinic acid is manufactured from petrochemical resources through oxidation of n-butane or benzene followed hydrolysis and finally dehydrogenation. Succinic acid is currently used as a surfactant, detergent extender, foaming agent, in the food industry for pH reduction, as a flavour and antimicrobial agent, and in the manufacture of health products such as vitamins and pharmaceuticals (Andersson et al., 2007).

Succinic acid, a di-carboxylic acid with the molecular formula $C_4H_6O_4$ was first discovered in 1546 by Georgius Agricola during dry distillation of amber. It is an intermediary product of the tri-carboxylic acid cycle, having tremendous future market potential. The tri-carboxylic acid cycle plays a key role in energy metabolism, in particular in higher cells and many microorganisms. In 2008, the turnover generated by biobased materials amounted to around 1.15 billion euros worldwide. According to estimation of Frost and Sullivan consultancy, the turnover in 2015 would be set to reach around 3.5 billion euros. Selection a right microorganism and genetic tools could be optimally suitable for the production of succinic acid (Agarwal et al., 2006). Metabolic engineering and biotechnological modification are used to drive the bacteria to maximum efficiency. Some well-established bacterial production strains are known to produce the succinic acid such as *Actinobacillus succinogenes, Corynebacterium glutamicum, Anaerobiospirillum succiniproducens, Mannheimia succiniproducens* as well as *Escherichia coli* (Lee et al., 2002; Okino et al., 2005). Bacterial strain should be very versatile with regard to use of different types of substrates such as the C6 sugars, C5 sugar (xylose) and C3 substrates (glycerine) for the production of succinic acid. It can be able to withstand the price fluctuations of the raw materials market, which is increasingly shifting towards volatile substances (Chandra et al., 2007).

The Biologically Derived Succinic Acid (BDSA) process has used a robust microorganism and new catalytic technology in the value-added step to convert corn-derived glucose to succinic acid at very high yields. Potential new markets for biobased production of succinic acid are expected to come from the synthesis of biodegradable polymers;

polybutylene succinate (PBS) and polyamides, and various green solvents (Cuklovic et al., 2008).

With a production quantity of about 30,000 tons per year, succinic acid is still a niche product for its ability to be processed into many other basic substances, thus fulfilling a decisive criterion of catalytically turning substances into platform chemical to produce 1,4-butanediol (BDO) and related products, tetrahydrofuran and γ-butyrolactone; n-methyl pyrrolidinone (NMP) and 2-pyrrolidinone; or other chemicals. (Werpy and Petersen, 2004).

Cost of Bio-Based Succinic Acid

Bio-based succinic acid production has shown some challenges like limited profitability of processes for succinic acid and high downstream process costs. At present one kilogram of succinic acid costs is come between two and three euros. If it becomes possible to produce the biobased variant of the molecule more cheaply, the market for succinic acid will grow enormously. To produce the large quantities of succinic acid by fermentation to be successful, it needs to study the impact of succinic acid on cell physiology, functionality, and viability needs to be considered (Cuklovic et al., 2008). It needs to develop the effective technologies for biological production of succinic acid from renewable waste resources or industrial waste substrates via study of biochemical conversion methods that would be cost-competitive in a long term perspective.

Production of Succinic Acid

Succinic acid can be produced by the fermentation of glucose using natural producers (*Anaerobiospirillum succiniproducens*) or engineered organisms (*Escherichia coli*), with mixture of byproducts including ethanol, lactic acid, and fumaric acid from which the succinic acid must be isolated. Process developments would be focused on reducing side-product formation through right organism selection (Chatterjee et al., 2001). Chosen of right biocatalyst would reduce the manufacturing cost of succinic acid which in turn affected by productivity and yield, raw material costs and utilization and recovery methods. High concentration of succinic acid causes cell death and need to maintain the cell viability via supply the fresh medium in fermentation to maintain high succinic acid productivity for a prolonged time. Some common problems such as sugar utilization and feedstock flexibility, and fermentation inhibition, both due to toxic compound derived from the raw material and the fermentation products themselves, are found in succinic acid fermentation. Efforts are needed to improve the efficiency of succinic acid production in terms of yields, productivities, concentrations, and product recovery. Cost reductions have also been sought through the use of cheaper feedstocks, including lignocellulosic materials of agro-waste products, sugarcane bagasse and paper and pulp (Cuklovic et al., 2008).

Liu et al has reported about process development of succinic acid production by *Escherichia coli* NZN111 using acetate as an aerobic carbon source and found that acetate showed a strong effect on both yield and productivity of succinic acid. Improved succinic acid production in the anaerobic culture of an *Escherichia coli* with double mutant (i.e. pflB and ldhA genes losses responsible for losing its ability to ferment glucose anaerobically and causes redox imbalance) was found due to enhanced anaplerotic activities in the preceding aerobic culture. Further, pflB ldhA double mutant of *Escherichia coli* is found its role in over expression of malate dehydrogenase enzyme, causes the improvements in succinic acid production concentration (31.9 g/L) and yield (1.19 mol/mol glucose) in fed-batch fermentation (Lin et al., 2005; Zhu et al., 2011).

Genome-based metabolic engineering of *Mannheimia succiniciproducens* LPK7 strain with disruption of the ldhA, pflB, pta and ackA genes is found suitable for succinic acid production without by-product formation such as acetic, formic, and lactic acids. Zhu et al 2011 had worked on metabolically engineered *Escherichia coli* strain SBS550MG (pHL413) for study of effect of culture operating conditions such as the aerobic growth phase on succinate production in a multiphase fed-batch bioreactor. They had worked on viability of Escherichia coli strain SBS550MG which had further increased the succinic acid production (Zhu et al., 2011; Vemuri et al., 2002). Bio-based succinic acid from biomass has been included in the US Department of Energy's Top Value Added Chemicals and is based on its potential as an important building block for deriving both commodity and speciality chemicals. The potential global market for succinic acid and its derivatives is estimated at 270,000 tonnes per year (Cuklovic and Stevens, 2008). However, the realisation of this global market share is found to sensitive to price reductions (up to 25%) of current cost (Thakker et al., 2006).

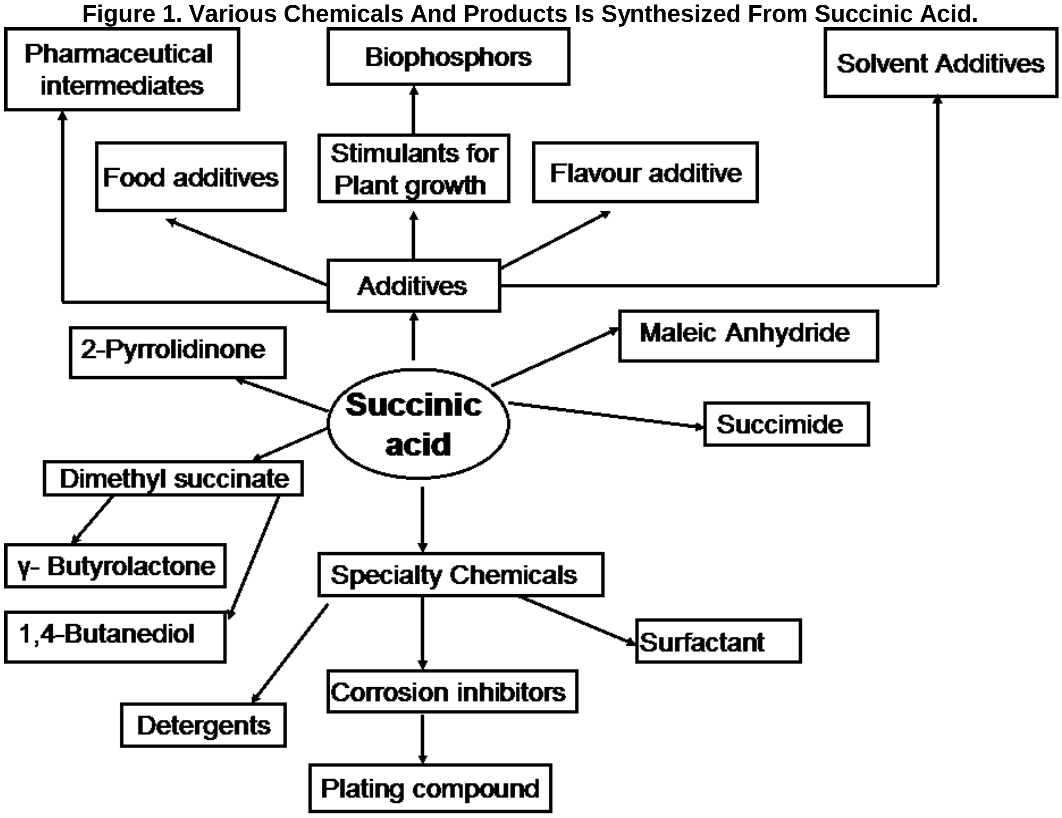
There are several companies such as Bioamber, DSM and Roquette, Myriant, BASF and Purac; and Mitsubishi Chemicals and PTT which are working on succinic acid production with use of different technology or collaboration. Bioamber, a joint Venture by DNP Green Technology and Agro Industrie Recherches et Developpments (A.R.D), have commissioned a demonstration plant in Pomacle, France with a capacity of 2,000 tonne per year (Hermann et al., 2007). DSM and Roquette are currently building a demonstration plant with a capacity of several hundred tonnes per year. Myriant, as successor to BioEnergy International, have been awarded $50 million by the US Department of Energy to help fund the construction of a succinic acid plant in Louisiana, US. BASF and Purac have announced their collaboration and intention to begin commercial production of succinic acid in the second quarter of 2010. Mitsubishi Chemicals have developed a process for the production of succinic acid, a precursor to PBS (U.S. Department of Energy, 2004; Nexant Chem Systems, 2008).

Application of Succinic Acid

Existing domestic markets for such chemicals total almost 1 billion pounds, or more than $l.3B, each year. 1, 4-butanediol is also an important basic compound for the production of polyamides and polyester. During biotechnological production, the microorganisms take up carbon dioxide. If the bacteria are fed with strongly reduced glycerine, they need to fixate one CO2 molecule from the air for each of the C3 molecules in order to create a C4 constituent of succinic acid. Thus, CO2 becomes an important raw material in the production of succinic acid (Hermann, et al., 2007).

Currently, succinic acid produced from the fermentation of carbohydrates, is mostly used within the food and beverage industry. Succinic acid is also used as precursor for 1, 4 butanediol (BDO), tertahydrofuran (THF) and γ-butyrolactone (GBL) synthesis. Succinic acid is used in the manufacture of polybutylene succinate (PBS), a biodegradable polymer sold under the names Bionolle® and GS Pla®, which are used as mulch films, rubbish bags and 'flushable' hygiene products. Succinic acid is a bulk chemical with global production at 16,000 to 30,000 tonnes with worth of US$ 2-3,000 per tonne, and annual growth of 10% (Patel, and Housing., 2006). Many application of succinic and its derivatives as a precursor to active pharmaceutical ingredients, as pharmaceutical additives, solvents, and polymers shown in **figure 1.**

Figure 1. Various Chemicals And Products Is Synthesized From Succinic Acid.



Conclusions

There are many application of succinic acid which it used as a surfactant, detergent extender, foaming agent, in the food industry for pH reduction, as a flavour and antimicrobial agent, and in the manufacture of health products such as vitamins and pharmaceuticals. From microorganisms by using cheap renewable resources biomass, succinic acid is produced. Bioprocess by using agrowaste materials has increased more interest with commercialization of bio-succinic acid. A novel industrial-scale bio refinery process is developing to convert corn-stover into succinic acid and co-products it is produced via chemical synthesis from petrochemical feedstocks that are nonrenewable, and the chemical processes suffer from problems of environment pollution. Therefore, great attention had been paid to use of renewable feedstocks through anaerobic fermentation for production of succinic acid which is among the 12 top value-added chemicals produced from biomass. As succinic acid is the main high-value chemical produced from the refinery with large quantity, market analysis of succinic acid is of crucial importance.

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