Biorenewable Production Pathway for Nylon and PET

Craig Forney

Iowa State University Research Foundation, Ames, IA, USA, ceforney@iastate.edu

ABSTRACT

Advances in biological engineering over the last several decades have increased the production of naturally (and some non-naturally) occuring molecules from biomass. However, it is evident that despite these gains, relying solely on biological methods will retard the eventual development of processes that will be suitable for industrial adoption for the production of commodity and specialty chemicals. A combination of biological and chemical reactions, beginning with biological conversion of biomass to platform chemicals and followed by chemical conversion to desired end products, will be required to realize the promise of a sustainable chemical industry. Iowa State University researchers have developed an integrated process combining fermentation using metabolically engineered yeast with electrocatalytic and chemical processing to create bio-based polyamides and terephthalic acid.

Keywords: biorenewable, fermentation, electrocatalysis, polyamides, PET

1 INTRODUCTION

Biomass conversion holds the promise to provide a sustainable feedstock for the production of chemical intermediates and products. While many groups are pursuing a direct replacement of conventional chemical intermediates through a strictly fermentative route, biomass conversion also opens up the possibility to easily add new functionality to the final product [1]. The solely fermentative approach, while capable of producing non-native molecules, is often disadvantaged by low yields, which ultimately may make the process not viable for commercial production. A hybrid approach, in which biomass-derived feedstock is biologically converted to a native molecule whose production is enhanced followed by more conventional chemical modification, appears to be able to address the yield problem and result in a scalable process.

Iowa State University researchers demonstrated the feasibility of this hybrid approach in the production of an unsaturated polyamide-6,6, beginning with fermentation of glucose to muconic acid followed by electrocatalytic hydrogenation to 3-hexenedioic acid and subsequent polycondensation with hexamethylenediamine to form the polyamide [2]. Alternately, these same initial steps can be tailored to produce a trans,trans-muconic acid which can readily be modified via a Diels Alder reaction to produce terephthalic acid for PET manufacture.

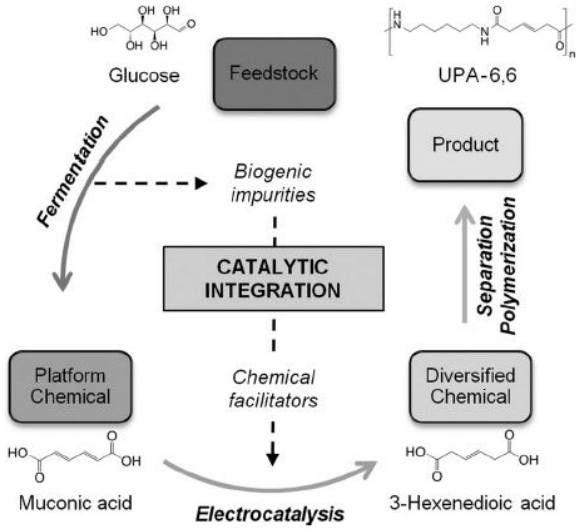

Figure 1: Biological and chemical conversion of glucose to unsaturated polyamide. Taken from [2].

2 EXPERIMENTAL

The pathway developed for this process employs a metabolically engineered yeast to convert glucose to muconic acid, followed by electrocataltic hydrogenation (ECH) of the muconic acid while still in the fermentation broth. The ECH step utilizes the salts and impurities from fermentation as electrolyte and hydrogen source, reducing the number of steps and resultant costs of production.

2.1 Strain Construction

Muconic acid production in yeast has been previously reported, though the final product titer was only 141 mg L^{-1} [4]. This low production could be attributed to low precursor availability, competing pathways, and rate-limiting enzymes. Muconic acid biosynthesis results from diverting a precursor from the shikimic acid pathway (which feeds aromatic amino acid biosynthesis). Low production issues were thus addressed by removing feedback inhibition by aromatic amino acids, increasing the production of phosphoenolpyruvate (PEP) and erythrose-4-phosphate (E4P) from glycolysis, and by increasing the utilization of E4P in the shikimic acid pathway. First, tyrosine-insensitive DAHP synthase was overexpressed to

reduce the feedback inhibition of aromatic amino acids. Erythrose-4-phosphate levels were increased by overexpression of the transketolase gene, and flux of E4P to the shikimic acid pathway was enhanced by modifying residues on the ARO1 enzyme.

Fermentation was performed in an oxygen-limiting environment to enhance conversion of protoctachuic acid (PCA) to the muconic acid precursor catechol. The final titer of muconic acid was 559.5 mg L^{-1}, a fourfold improvement over the previously-reported high [4].

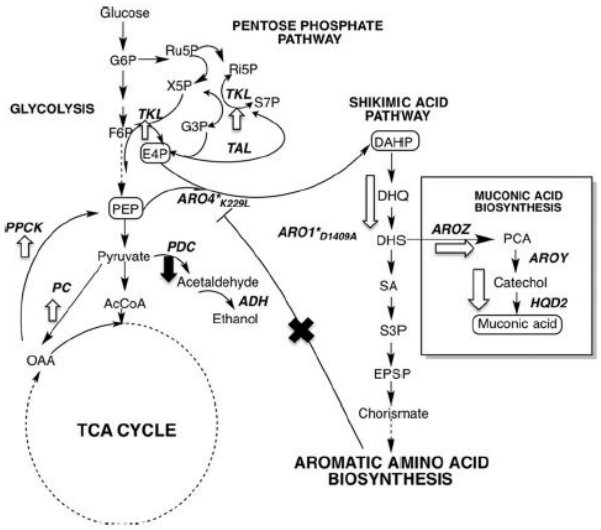

Figure 2: Metabolic pathway for overproduction of muconic acid in *Saccharomyces cerevisiae*. Reduction in feedback inhibition by aromatic amino acids, increased production of phosphoenolpyruvate (PEP) and erythrose-4-phosphate (E4P), and diversion of precursors into the shikimic acid pathway were used to increase muconic acid production. Taken from [2].

2.2 Hydrogenation of Muconic Acid

The fermentation broth was directly hydrogenated in a three-electrode electrochemical cell. This process was performed at ambient temperature and pressure, with hydrogen provided by water splitting at the electrode surface. Muconic acid was hydrogenated at the cathode surface as well according to the following steps:

$$H^+ + e^- + {}^* \rightarrow H^* \quad (1)$$

$$2H^* + C_6H_6O_4 + {}^* \rightarrow C_6H_8O_4 \quad (2)$$

The catalyst for this reaction was lead (Pb). In addition to the low cost and relative abundance of lead, the lead electrode was chemically stable and resisted poisoning. This stability allowed for the use of raw fermentation broth (containing whole yeast cells, unspent salts, and biogenic impurities) in the reaction cell, greatly simplifying the hydrogenation reaction. Voltage potential was -1.5 V vs. Ag/AgCl at the cathode; at a pH of 2.0 the maximum conversion of muconic acid to 3-hexenedioic acid (HDA) was 96% with a selectivity of 98%. Leaching of the lead cathode was minimal, determined to be approximately 6.5 ppm by elemental analysis. Deactivation of the catalyst was not observed over the course of five successive runs.

2.3 Polyamide Production

3-Hexenedioic acid was separated from the fermentation broth by a combination of vacuum evaporation, filtration over activated carbon, and crystallization. Thus purified HDA was polymerized via a polycondensation reaction with hexamethylenediamine to create an unsaturated polyamide-6,6 (UPA-6,6). A saturated nylon-6,6 was produced in similar manner from adipic acid and hexamethylenediamine for comparison purposes. Phyiscal properties of the saturated and unsaturated polyamides are compared in Table 1.

Property	Nylon-6,6	UPA-6,6
M_n (Da)	17,800	12,200
PDI	2.0	3.36
T_{melt} [°C]	250	60

Table 1: Number average molecular weight (M_n), polydispersity (PDI) and melting temperature (T_{melt}) of Nylon-6,6 and UPA-6,6.

Adipic acid and HDA may be blended to tune the properties of the resultant polyamide; the presence of HDA in these polymers provides grafting sites to further tune the properties through cross-linking as well as potentially add new functionality.

2.4 Trans,trans-Muconic Acid Isomerization

Trans,trans-muconic acid has previously been shown to undergo a Diels Alder reaction to form terephthalic acid, one of the two monomers used to produce polyethylene terephthalate (PET). Unfortunately, biologically-produced muconic acid is normally in the cis,cis- configuration. Isomerization of the cis,cis-muconic acid to trans,trans-muconic acid can be accomplished using a very small potential in the same electrochemical cell as was used for hydrogenation.

3 DISCUSSION

An integrated strategy has been developed to convert the biomass-derived feedstock glucose to polyamides through a combination of biological and chemical catalytic steps. Notably, the electrocatalytic reaction takes place in raw fermentation broth, demonstrating that costly

separation steps can, in part, be avoided using this process. Importantly, the family of unsatrated polyamides should facilitate integration of new functionality into the polyamide backbone. Finally, the flexibility of the process to produce intermediates for polyamide and polyester production provides the ability to shift production to take advantage of market needs.

REFERENCES

[1] N. Hernández, R.C. Williams, and E. Cochran, "The Battle for the "Green" Polymer. Different Approaches for Biopolymer Synthesis: Bioadvantaged vs. Bioreplacement," Organic & Biomolecular Chemistry, 12, 2834, 2014.

[2] M. Suastegui et al., "Combining Metabolic Engineering and Electrocatalysis: Application to the Production of Polyamides from Sugar," Angewandte Chemie International Edition, 55, 2368, 2016.

[3] J.E. Matthiesen, J.M. Carraher, M. Vasiliu, D.A. Dixon & J.P. Tessonnier, "Electrochemical Conversion of cis,trans-Muconic Acid to trans,trans-Muconic Acid and trans-3-Hexenedioic Acid for Bio-Based Polymer Production," 229th ECS Meeting, 1 June 2016.

[4] K.A. Curran, J.M. Leavitt, A.S. Karim & AS. Alper, "Metabolic Engineering of Muconic Acid Production in *Saccharomyces cerevisiae*," Metabolic Engineering, 15, 55, 2013.