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Optimization of methane bio-hydroxylation using waste activated sludge mixed culture of type I methanotrophs as biocatalyst

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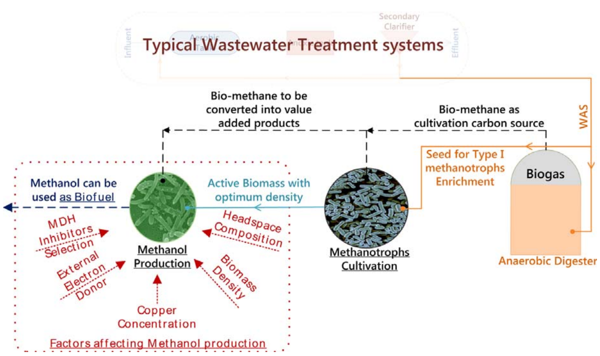
HIGHLIGHTS

- Methanotrophs enriched from waste activated sludge were used to produce methanol.
- Multiple methanol dehydrogenase inhibitors concentrations were screened.
- Headspace gas composition were optimized to promote methanol productivity.
- The attained methanol productivity obtained was comparable to pure cultures.
- Methanol concentration attained was double the reported using mixed culture.

GRAPHICAL ABSTRACT



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ABSTRACT

Captured biogas produced within wastewater treatment facilities can be the remedy to offset its increasing energy requirements. Furthermore, the conversion of methane to methanol is quite attractive as it is more transportable and has higher energy yield. Methane can be utilized by methanotrophs in which methanol is produced as a metabolic intermediate. Compared to type II, type I methanotrophs are more advantageous due to its higher growth yields and energy efficiency. This work objective is to optimize methanol bio-production using type I methanotrophs enriched from activated sludge process. This study demonstrates methanol production using mixed culture from wastewater sludge. Optimization of methanol dehydrogenase inhibitors, sodium formate, and copper concentrations, as well as, the gaseous headspace composition and biomass density resulted in a significant enhancement in methanol production. The maximum methanol concentration achieved in this study was 485 ± 21 mg/L. Whereas, the highest methanol productivity obtained was equal to 2115 ± 81 mg/L/day. Those findings show the high potential of producing methanol using mixed culture enriched from activated sludge process.

1. Introduction

Over the past few decades, our planet has witnessed the augmentation of greenhouse gases (GHG) emissions resulting in notable climate and environmental changes. Having the same GHG emissions trend, it is predicted that by 2050 the global warming would increase by 2 °C above the temperature level in 1900 [1]. Wastewater treatment facilities (WWTFs) contribute to the global warming phenomena not only by GHG emissions such as methane and carbon dioxide, but also, by consuming enormous amount of fossil fuel based energy [2]. For instance,

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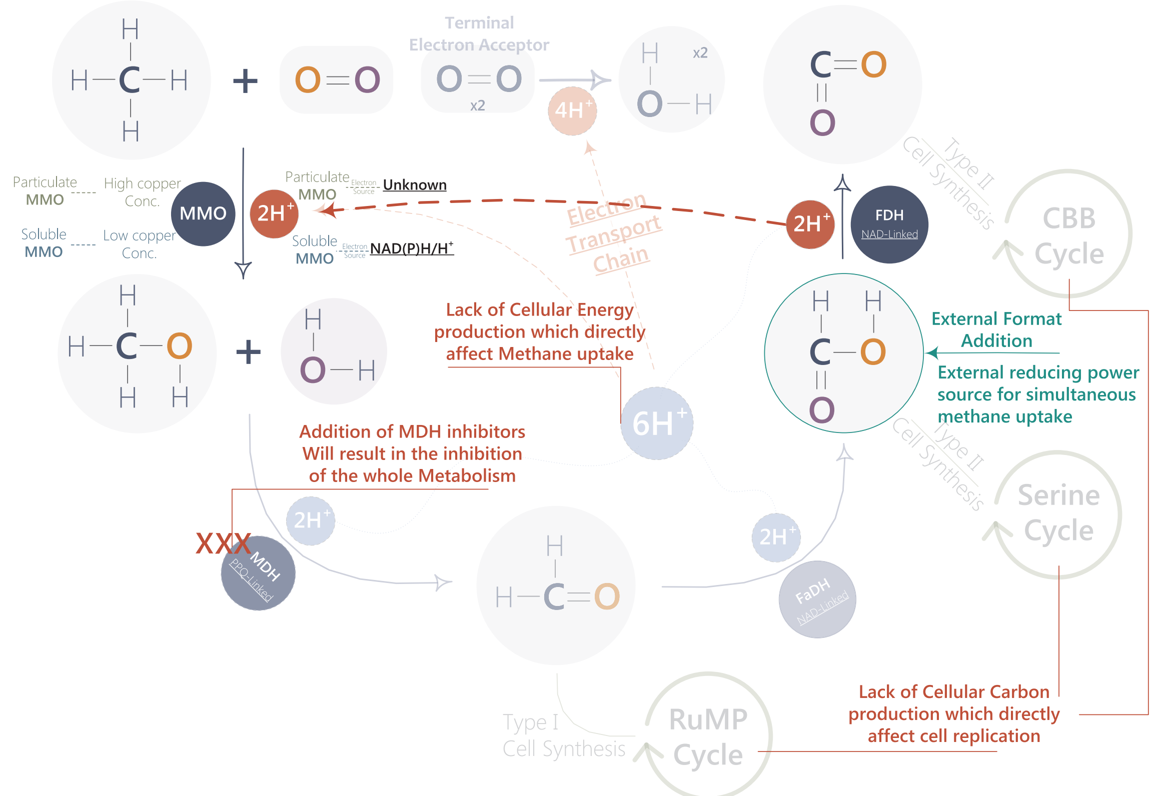

Fig. 1. Methanol Production Metabolism. MMO: Methane Monooxygenase, MDH: Methanol Dehydrogenase, FaDH: Formaldehyde Dehydrogenase, FDH: Formate Dehydrogenase, PQQ: Pyrroloquinoline Quinone, NAD(P)H: Nicotinamide Adenine Dinucleotide, RuMP: Ribulose Monophosphate pathway, CBB: Calvin-Benson-Bassaham.

WWTFs electricity consumption, which represents 4% of the United states total electrical consumption, results in about 45 million tons of GHG emissions annually [3,4]. Whereas, United states' WWTFs directly released, from the biological treatment processes, only 15 million metric tons of CO_2 equivalents of GHG in 2014 [5]. This enormous amount of energy required for wastewater treatment is not only of environmental interest but also of economic interest [2,6]. Furthermore, WWTFs are expected to receive higher wastewater volumes at the expense of higher energy inputs due to the ongoing population increase and the higher restrictions on the effluent quality [2]. Therefore, there is a great interest in resources recovery from wastewater streams to offset its operational energy inputs and reduce its net GHG emissions. Interestingly, it was reported in a study performed on WWTF located in Toronto, Canada, that wastewater has energy content (in the form of organics) up to 9 times higher than the energy consumed for its treatment [7]. Combined together, those reports led to the paradigm shift from considering the wastewater as waste needs to be treated and disposed into an energy and value-added products resource [8].

Anaerobic digestion (AD) process has been widely adopted by WWTFs for sludge minimization and biogas production as energy and resources recovery technology. Throughout the AD process, organic matters are biologically degraded anaerobically while producing biogas (consists of up to 70% of methane) [9,10]. It was estimated that WWTFs located in North America have the potential to produce about 3.90 billion m^3 of biomethane per year. This could prevent the GHG emission of similar to taking 1.18 billion passenger vehicles off the roads [11–13]. Nevertheless, multiple obstacles limit the direct energy generation of the biogas form anaerobic digestion including the existence of impurities and moisture, its low handling and collecting capabilities, and lack of convenient infrastructure for gas distribution. Moreover, the combined heat and energy technologies show low electricity efficiency ($\eta \approx$ 25–40%) [14,15]. Those obstacles, unfortunately, induce most of the WWTFs to either use the biogas for facility internal heating (in winter time only) or to flare it.

On the other hand, methanol has recently attracted the attention as an alternative fuel due to its lower cost and GHG emissions [16,17]. In comparison with methane, methanol, as a liquid fuel, is more storable, secured, compatible with the existing fueling infrastructure [1,18]. In addition, more energy can be derived from methanol (15.8 MJ/L) compared with methane (38.1×10^{-3} MJ/L) [19]. Methanol can be used as a transportation fuel either alone or blended with gasoline resulting in higher complete combustion due to methanol oxygenated content [18,20]. Using gasoline substitution ratio less than 50%, methanol-gasoline blend can be efficiently used as a transportation fuel without any engine modifications [16]. In terms of electricity generation, direct methanol fuel cells (DMFC) are intensively studied as one of the most promising power supply alternatives for portable and micro applications [21]. DMFCs are advantageous due to its higher energy density, convenient operation, and fast refueling [16,17,22]. Furthermore, methanol has been commonly used as an external carbon source used to enhance biological nitrogen removal (BNR) processes [23]. Collectively, methanol is as a multiple use commodity with a prominent role as an efficient and sustainable substitute for biomethane produced within WWTFs what makes the process of methane bio-hydroxylation more feasible.

Descending from the Methylotrophic bacterial group, methanotrophs are gram-negative bacteria that have the distinguish capability of exploiting methane as the cellular carbon and energy source [24]. Thus, methanotrophs can be employed as biological catalysts for methane hydroxylation. Owing to the possession of methane monooxygenase (MMO) enzyme, methanotrophs catalyze methane oxidation into methanol based on Eq. (1). As shown in the Equation, one oxygen molecule and two electrons should be incorporated in methane hydroxylation.

$$CH_4 + O_2 + 2e^- + 2H^+ \rightarrow CH_3OH + H_2O \quad (1)$$