# Turning C1-gases to Isobutanol Towards a Great Environmental and Economic Sustainability via Innovative Biological Routes: Two Birds With One Stone

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**Research**

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# Abstract

Background

The dramatic increase in emissions of greenhouse gases (GHGs) has led to an irreversible effect on the ecosystem, which in turn caused significant harm to human beings and other species. Exploring innovative and effective approaches to neutralizing GHGs is urgently needed. Considering the advancement of synthetic biology and the bioconversion process, C1-utilizing cell factories (CUCFs) have been modified to be able to effectively convert C1-gases includes biogas, natural gas, and carbon dioxide ($CO_2$) into chemicals or fuels via biological routes, which greatly facilitates the inedible carbon sources used in biomanufacturing, increases the potential value of GHGs and meanwhile reduces the GHG emissions.

Process design and results

Even though the current experimental results are satisfactory in lab-scale research, the evaluation of economic feasibility as well as applications of CUCFs in industrial-scale still need to be analyzed. This study designed three scenarios of CUCFs-based conversion of biogas, natural gas, and $CO_2$ into isobutanol, the detailed techno-economic analyses of these scenarios were conducted with the comparisons of capital cost, operating cost, and minimum isobutanol selling price (MISP). Results revealed that direct bio-conversion of $CO_2$ by CUCFs into isobutanol exhibited the best economic performance with a MISP of $1.38/kg isobutanol. The single sensitivity analysis showed that the gas utilization rate, flow rate, and $CO_2$ cost are the three most significant economic-driving forces on MISP of $CO_2$-derived biological isobutanol. Multiple-point sensitivity analysis presented that the MISP for the long-term case can be as low as 0.99 $/kg with using ideal targets.

Conclusions

Our findings provide a comprehensive assessment of bio-conversion of C1-gases via CUCFs to isobutanol in terms of the bioprocess design, mass/energy calculation, capital investment, operating expense, sensitivity analysis, and environmental impact. It is expected that this study may lead to the paradigm shift in isobutanol synthesis with C1-gases as substrates.


# 1. Background

As a building block chemical, isobutanol has been widely applied in various fields including food, solvents, extractants, rubber, fuel additions, and transportation fuels [1-3]. At present, isobutanol is generally produced via chemical route, i.e. the propylene carbonyl synthesis processes, of which isobutyraldehyde as the byproduct from n-butanol and 2-ethyl hexanol production is hydrogenated into isobutanol [4]. However, chemical synthesis not only requires expensive catalysts, changing temperatures, and pressures, but also contributes to greenhouse gas emissions[5]. Recently, the biological route has been widely considered as an economically feasible and environmentally sustainable approach for isobutanol synthesis due to lower raw material costs (and not competing with food crops) and less harmful gas emissions. [6, 7]. Although starchy and sugar-based substrates including corn, potato, glucose, etc. have always been the main feedstock for isobutanol production, it may not be sustainable as it inevitably compromises future food supply. While the cost of isobutanol production from food was also much higher than the current production cost of the chemical route due to the high cost of the feedstock, which was not profitable. Therefore, there is an urgent need to explore alternative inedible and low-cost carbon sources, such as greenhouse gases (GHGs), which is consistent with the best interest of global sustainability [8].

$CO_2$ and $CH_4$ are two major GHGs causing the global warming effect, which is the most important environmental problem facing the world today. $CO_2$ in the atmosphere is mainly released from the burning of fossil fuels, the ruining of plants, and industrial processes. According to the annual report of the International Energy Agency (IEA), global energy-related $CO_2$ emissions flattened in 2019 at around 33 Gigatonnes (Gt)[9]. Biogas contains $CH_4$ and $CO_2$ etc. can be produced from waste biomass by the anaerobic digestion (AD) process [10, 11], of which its global production capacity has been able to reach 370 TWh in 2017 [12]. While in China, about 80 million household biogas plants and 8000 large-scale biogas projects are expected to provide an annual production of 50 billion $m^3$ biogas by 2020 (Scarlat et al., 2018). However, biogas currently is mainly employed for generating electricity and heat through combustion, which will directly losses 50% of total carbon atom by releasing one mole $CO_2$. Although effective biohythane production from waste biomass by two-stage anaerobic fermentation for coproduction of hydrogen and methane provides higher energy recovery rate and shorter fermentation time than one-stage methane fermentation, the utilization of C1 gases is still considered as a challenging work in term of GHGs reduction and reuse of waste biomass[13]. In addition to biogas, the extraction activity of natural gas is another major source of GHG emission. It was reported that a large amount of shale gas remains due to the rapid development of fracking technologies, of which about 440 billion $m^3$ of natural gas has been wasted worldwide annually [14, 15]. It has been considered that biological valorization of GHGs into chemicals or fuels could be a promising route giving a great environmental and economic sustainability as well as reduction of GHG emission as two birds with one stone.

Currently, more efforts have been paid to convert these abundant and low-cost C1-gaseous substrates into isobutanol by biological routes via systems engineering of C1-utilizing cell factories (CUCFs) as shown in Figure 1. More than 900 mg/L of isobutanol has been demonstrated for the first time by using $CO_2$ as the sole carbon source in the autotrophic cultivation of *Synechocystis PCC* 6803 [16]. In recently years, the approaches and strategies of methanotrophic-based CUCFs are constructed and modified by using genetic-engineering tool, systematic manipulation, metabolic modeling, and carbon flux simulation[17-19]. However, the challenges and opportunities for methane bioconversion into isobutanol by methanotrophs are still remained in both scientific and industrial applications. Although Precigen Inc (formerly named as Intrexon Corporation), a biosynthesis-based company has claimed that $CH_4$-derived isobutanol has been accomplished in a lab-scale by an engineered methanotrophic bacteria [20], no scientific results have been published so far. Therefore, to verify the possibility of isobutanol biosynthesis from $CH_4$, *Methylomicrobium buryatense* (an industrial-proved methanotrophic bacteria) was genetically engineered to achieve direct conversion of $CH_4$ into isobutanol in our laboratory. By heterologous expressing α-ketoisovalerate decarboxylase (Kivd) derived from *Lactococcus lactis*, the isobutanol-producing M. buryatense was constructed (Figure S1), which accumulated about 35 mg/L in 5 days under the unoptimized condition in vials with a very limited gas transfer efficiency (Figure S2). It has been reported that a 1000-times increase in isobutanol productivity can be achieved by using C1-gaseous substrate and airlift bioreactors [21, 22]. Besides, the carbon conversion efficiency can be also significantly improved by using the genetic engineering key enzymes for isobutanol biosynthesis as previous report [16, 23]. Therefore, it is highly predictable that the industrial applications of $CO_2$/$CH_4$-based isobutanol production can be achieved with the rapid development in synthetic biology technologies and processes.

Although current experimental results are satisfactory in lab-scale research, the economic feasibility and effective applications in industrial-scale still need to be fully analyzed and evaluated. Techno-economic analysis (TEA) is usually used to decide future investigation to reach targeted goals with less time and labor cost. To the best of our knowledge, TEA on bio-conversion of $CO_2$ and $CH_4$ to isobutanol is rarely reported. Therefore, this study aims to conduct detailed techno-economic analyses on the designed three scenarios of bioconversion of the biogas, natural gas, and $CO_2$ to isobutanol, with the comparisons of capital cost, operating cost, and minimum isobutanol selling price (MISP). Sensitivity analyses were also carried out to guide the engineering practice of isobutanol production with the most economic potential. It is expected that this study may lead to the paradigm shift in isobutanol biosynthesis with C1-gases by systematically assess and compare the economic feasibility and competitiveness of various integrated bioprocesses.

# 2. Process Designs And Assumptions For Isobutanol Production From C1 Gaseous Substrates

## 2.1 Scenarios

To evaluate the technical and economic performance of isobutanol production from biogas, natural gas, and $CO_2$, the following three bio-routes were designed and entitled as scenario #1, scenario #2, and scenario #3 as shown in Figure 2. For scenario #1, both $CH_4$ and $CO_2$ in biogas are converted into isobutanol directly in a two-stage cultivation by cyanobacteria and methanotrophs, respectively. Natural gas and pure $CO_2$ are used as the sole carbon source for isobutanol biosynthesis in scenario #2 and scenario #3, respectively. The simulation was accomplished by a commercial process software (AspenTech®, Cambridge, MA, USA) for the calculation of rigorous material and energy balance.

## 2.2 Process design

A simplified overall process diagram was shown in Figure S3, including gas supply, isobutanol production, isobutanol purification, wastewater treatment (WWT), and utilities. The mass and energy flow in Aspen simulation models was calculated based on an $n^{th}$-plant of isobutanol with an annual production capacity of 50,000 tons (Table 3).

### 2.2.1 Gas supply (Area 100)

In this Area, the C1-gaseous substrate such as biogas, natural gas, or $CO_2$ will be compressed to bioreactors for isobutanol production (Fig. S1). Although the composition of biogas varies with feedstock for AD, the ratio of $CH_4$ and $CO_2$ in biogas used in scenario #1 was assumed as 50%:50%. For scenario #2, natural gas containing 90% $CH_4$ was applied and the other 10% impurities proved to be unharmful in the growth of methanotrophs was remained in the natural gas directed to bioreactors [24]. The substrate of scenario #3 was assumed as 100% $CO_2$. The air-stream routed to a pressure swing adsorption (PSA) unit was enriched into 95% $O_2$ for the cultivation of methanotrophs in scenario #1 and scenario #2 [25]. To achieve 50,000 tons of isobutanol production per year, the demand for gaseous substrates calculated in Aspen for scenario #1, scenario #2, and scenario #3 was 388,740 $m^3$/day, 278,250 $m^3$/day, and 321,338 $m^3$/day, respectively.

### 2.2.2 Isobutanol production (Area 200)

The isobutanol production process involves both biomass generation and isobutanol biosynthesis by either cyanobacterium cultured in closed tubular photobioreactors (PBR) with 50 $m^3$ working volume [26] or methanotrophs grown in 1000 $m^3$ bubble column bioreactors (BCB) with a working volume of 80% [25, 27]. The stoichiometry equations for biomass generation by cyanobacteria *(Eq.1)* and methanotrophs *(Eq.2)* applied in Aspen simulations have been proposed based upon published literature [28, 29], in which the empirical formula of “$CH_{1.934}O_{0.473}N_{0.23}$” and “$C_4H_8O_2N$” stands for the biomass from cyanobacteria and methanotrophs, respectively. The stoichiometry equation for isobutanol biosynthesis in cyanobacteria and methanotrophs has been assumed as *Eq. 3* and *Eq. 4*, respectively. For scenario #1, the biogas was sent into PBR first, where $CO_2$ can be immobilized by cyanobacterium via photosynthesis for isobutanol biosynthesis and $O_2$ production. The off-gas from PBR containing $CH_4$, $O_2$, and unused $CO_2$ will be transferred to BCB for isobutanol production by aerobic-obligated methanotrophs.

$$CO_2+0.622H_2O+0.23NH_3 \rightarrow CH_{1.934}O_{0.473}N_{0.23}+1.0745O_2 \quad (Eq.1)$$

$$CH_4+1.5O_2+0.118NH_3 \rightarrow 0.118C_4H_8O_2N+0.529CO_2+1.71H_2O \quad (Eq.2)$$

$$4CO_2+5H_2O \rightarrow C_4H_{10}O+6O_2 \quad (Eq.3)$$

$$6CH_4+6O_2 \rightarrow C_4H_{10}O+2CO_2+7H_2O \quad (Eq.4)$$

Since carbon and nitrogen sources dominate the cost of raw material[30], only gaseous substrate and ammonia were considered for economic analysis in this study. Considering the industrial application, the isobutanol productivity of 1 g/L/h and isobutanol titer of 1 g/L were assumed as the baseline of the TEA in all scenarios. The utilization rate of $CH_4$ and $CO_2$ was set as 90%, of which 20% was projected for cell growth and the rest for isobutanol biosynthesis.

### 2.2.3 Isobutanol purification (Area 300)

The effluent from A200 was first pretreated with a three-step dewatering process to separate the liquid phase and cell mass. The dewatering procedure applied in this TEA has been discussed previously [31, 32]. As described by Tao et al. [33], an isobutanol-water mixture obtained from the dewatering process will be future purified by using several distillation columns to harvest isobutanol from the liquid phase. The first column is to concentrate the overhead product to the ratio of isobutanol and water for a liquid-liquid split, and then the isobutanol-rich stream is sent to the second column for high purity of isobutanol as the final product. This separation efficiency of 90% was assumed in Aspen simulation based on published literature [33].

### 2.2.4 WWT (Area 400)

As shown in Figure S2, all effluents collected from A300 composed of wastewater, spent cell mass, and other liquid streams were directed to an on-site AD plant. The biogas generated from AD along with other gases collected from A200 will be transferred to combustion facilities for onsite energy generation [31]. A large amount of high-nitrogen sludge extracted from AD was assumed to be recovered as a byproduct for fertilization [34].

### 2.2.5 Utilities (Area 500)

Area 500 facilitates overall energy, water, and power integration, including a cooling water system, chilled water system, process water manifold, and power systems. In this area, the amount of make-up water required by the process can be determined, as well as total power requirements for the system and electricity purchased from the grid.

## 2.3 Process economics analysis

This study aims to assess and analyze the economic feasibility and competitiveness of various integrated processes of isobutanol production from C1-gaseous substrates to guide practical engineering activities. It is worth mentioning that all methods or data used in this TEA were obtained from published literature and official reports.

### 2.3.1 Capital expenses (CAPEX) and discounted cash flow method

The capital expenses were calculated by considering the total cost of purchased equipment and associated installation. The costs of units or equipment used in this study were obtained from previous studies [20], and the new equipment costs for different sizes were calculated using the exponential scaling expression (New cost = Base cost X (New size/ Base size)$^n$, where n is the economy scaling factor and varies with the equipment) based on the equipment size for the original price quote. The Chemical

Engineering Plant Cost Index (CEPCI) was used to recalculate equipment costs to $2020 [35]. The assumptions used for this TEA can be found in Table 2, which were proposed based on previous literature [36, 37].

2.3.2 Operating Expenses (OPEX)

The OPEX including gaseous substrates, other raw materials, and fixed operating costs were depicted in Table 2. In this study, all values derived from published TEA reports or literature have been converted into 2020 dollar ($2020) based on the Industrial Inorganic Chemical Index from SRI Consulting for raw materials [38] and the labor indices from the US Department of Labor Bureau of Labor Statistics for employee salaries [39]. The labor burden was estimated to be 60% of the total wage, and 2% of the inside boundary limit (ISBL) capital expenses were designated for maintenance. Besides, local property tax and property insurance were estimated at 0.5% of fixed capital investment [40]

# 3. Economic Comparison Of Isobutanol Production From C1-gaseous Substrates

### 3.1 Comparison of CAPEX and OPEX

To explore the most economic potential scenario, a detailed comparison of capital investments and additional necessary expenses (warehouse, piping, land, etc.) were obtained by using chemical engineering cost estimation techniques as shown in Figure 3. The highest CAPEX of $333 MM was observed in scenario #1 contributed to the high equipment investment of A400, which is attributed to the larger amount of wastewater produced from the two-stage culture of different microbes, while only single-stage cultures in scenario #2 and #3. The low isobutanol titer applied in this TEA may be another reason for a large amount of wastewater generation. Moreover, profiting from the low cost of PBR for cyanobacteria culture, compared with bubble column bioreactors used in methanotrophic cultivation in scenario #2 (Table S1), a total of 17% CAPEX reduction can be achieved in scenario #3. It can be seen from Table 4 that the OPEX of the three scenarios presented the major difference in the cost of carbon and nitrogen sources. Scenario #3 has the lowest total operating cost of $55.09 MM due to the relatively low cost of $CO_2$. Interestingly, the C1 gashouse substrates in all three scenarios only contribute around 30-40% of total OPEX, which was significantly lower than the proportion (60-80%) of conventional raw material (e.g., glucose), indicates that using C1 gashouse substrates as carbon sources for isobutanol production is more economically feasible and market potential.

### 3.2 Minimum isobutanol selling price (MISP) from different scenarios

Determining a minimum selling price based on investment expense refers to analyze the cost of a business decision in terms of the real-time relevant expenses. The minimum pricing is the breakeven point for that given sale. According to the global isobutanol production market, the $n^{th}$ plant with an annual capacity of 50,000 tons isobutanol was projected and the resulting MISP of three scenarios at a 10% IRR was $1.57/kg isobutanol, $1.81/kg isobutanol, and $1.38/kg isobutanol in scenario #1, #2, and

#3, respectively. Apparently, because of the lowest CAPEX and OPEX, the most inexpensive MISP of $1.38/kg isobutanol was obtained in scenario #3, which is 12% and 20% lower than the MISP from scenario #1 and scenario #2, respectively. This MISP is close to the current market price of $1.0/kg isobutanol [41] showing the economic potential of $CO_2$-based isobutanol. Given the promising economic feasibility of scenario #3, sensitivity analyses of bioconversion of $CO_2$ to isobutanol were also carried out to investigate the impact of key variables on MISP and consider options for optimizing process economics.

# 4. Sensitivity Analysis For Isobutanol Production From Co2

### 4.1 Single-point sensitivity analysis

Sensitivity analysis is an efficient approach to quantify the impact of key variables on overall economics. Therefore, a single-point sensitivity analysis was firstly performed using the Aspen model by adjusting only one single variable in reasonable ranges while all others were held constant. In this study, seven variables associated with $CO_2$-derived isobutanol production were evaluated for the influences on MISP. The baseline for all variables was the same as the assumptions used in the case of scenario #3.

As shown in the spider chart (Figure 4), the gas utilization rate was the most important cost-driving force impacting the overall cost of isobutanol production. When the gas utilization rate decreases by 50%, MISP increase from $1.38 to $2.30/kg isobutanol. This finding might be attributed to the fact that the utilization rate strongly affects the isobutanol yield, which in turn led to a significant impact on raw material expenses and bioreactor investment. It’s well known that the cost of glucose used in a bioprocess may contribute up to 60% of the total operating cost[42], while the cost of $CO_2$ in scenario #3 only accounts for 30% of the total operating cost, which dramatically reduces the MISP as shown in Figure 4. It can be expected that using wasted $CO_2$ collected from factories and power plants will further decrease the MISP. Different from sugar-based bioprocesses, the supply of gaseous substrates will be mainly determined by the gas flow rate. Unsurprisingly, the flow rate of $CO_2$ can significantly influence the MISP in a range from $1.14 to $2.63/kg isobutanol.

In addition to the aforementioned variables, other factors such as plant production capacity, flocculant usage, isobutanol titer, and productivity will also influence the MISP. The influence of plant production capacity on MISP, CAPEX, and OPEX can be seen in Figure 5. As the isobutanol production capacity increases, OPEX and CAPEX increased rapidly and then stabilized. It can be noted that the effect of these two parameters makes the MISP decline sharply with the increase of production capacity, and keep stabilization when the production capacity is higher than 100,000 ton/y. These findings suggested that the MISP will not be affected when the variables over a certain threshold. Based on these above results, several key variables with optimal values were selected as targeted goals in the multiple-point sensitivity analysis for a long-term case.

### 4.2 Multiple-point sensitivity analysis

To have a comprehensive understanding of the effects of key factors on the economic performance of $CO_2$-derived isobutanol, an exhaustive sensitivity analysis was conducted by simultaneously adjusting five key variables for MISP calculation. As shown in Figure 6, two situations including the base case (baseline in the single-point sensitivity analysis) and long-term case were illustrated and compared to project the cost potentials of this technology pathway. Given the optimization of genetic engineering, enhancement of bioconversion process, and improvement of $CO_2$ capture efficiency, the isobutanol concentration, annual production, and the $CO_2$ price were rationally predicted. The MISP for the long-term case can be as low as 0.99 $/kg isobutanol by using ideal targets. Although currently demonstrated technology and recently published results of isobutanol biosynthesis from $CO_2$ is still far away from our ideal targets, it is believed that the advanced biotechnologies can fill in the gap by constructing a robotic cell factory, reaching theoretical conversion efficiency, and owning minimum raw material cost.

# 5. Prospective And Conclusions

Currently, biogas, natural gas, and $CO_2$ are the main contributions to GHGs emissions, of which the amounts are projected to further increase in the coming decades. It is obvious that GHGs if not managed properly would cause many problems, e.g., iceberg thawing, increase in pests and diseases, and intensified desertification, thus GHGs management is becoming a pressing challenge worldwide. Although biogas and natural gas combustion can recover electric energy, the conversion efficiency is very low and the generation of GHGs gases (e.g., $CO_2$), which suggested that the combustion-based GHGs management may be challenged because of its environmental and economic sustainability. Besides, $CO_2$ from factories and power plants can be served as the carbon source for genetic engineered cyanobacteria biosynthesizing desired production, of which the isobutanol from the cyanobacteria can be further used directly as the fuel addition or platform chemical. By adopting the proposed biological routes, the GHGs includes biogas, natural gas, and $CO_2$ can be used as an alternative inedible and low-cost substrate for isobutanol production, which significantly reduces the raw material cost. Meanwhile, not any GHGs are generated from the biological process. These in turn suggested that the proposed biological routes can realize concurrent resource recovery and GHGs reduction, i.e., two birds with one stone, which is consistent with the best interest of global environmental and economic sustainability.

In this study, the TEA was applied to conduct a systematic economic assessment on calculating OPEX and CPAEX of the proposed biological routes to evaluate the economic feasibility and industrialization potential of bioconversion of C1-gaseous substrates for isobutanol production. Because of the low-cost investment for wastewater treatment and raw materials, the $CO_2$-derived isobutanol presents the lowest MISP of $1.38/kg isobutanol. By employing the single/ multiple-point sensitivity analyses, utilization efficiency, flow rate, and plant capacity are determined as key cost drivers. With the expected research targets, the promising MISP of $0.99/kg isobutanol can be achieved by reducing $CO_2$ cost and enhancing the production performance of isobutanol titer and plant capacity, which is lower than the current market price of $1.0/kg isobutanol. It is expected that this study may provide engineering practice guidance and cost optimization strategies for future biological conversion of C1 greenhouse gases to platform chemicals.

# Abbreviations

GHGs: greenhouse gases;TEA: techno-economic analysis; MISP: minimum isobutanol selling prices; R&D: research and development; CCE: carbon conversion efficiency; IRR: internal rate of return; CAPEX: capital expenses; OPEX: operating expenses; AGR: amine-based acid gas removal; ASU: air separation unit; CSTR: continuous stirred tank reactors; DAF: dissolved air flotation; AD: anaerobic digestion; WWT: wastewater treatment; ISBL: inside battery limit; FCI: fixed cost investment; SOT: state of technology; LCA: life cycle assessment; TCI: total capital investment; TDC: total direct costs; TIC: total indirect costs; TOC: total operating cost; GHG: greenhouse gas; GWP: global warming potential.

# Declarations

### Ethical Approval and Consent to participate

Not applicable

### Consent for publication

Not applicable

### Availability of supporting data

Not applicable

### Competing interests

The authors declare no competing interests.


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### Authors’ contributions

BL and RF performed the process design and wrote manuscript. YM and XH revised the manuscript. LH carried out the genetic engineering work. QF designed the process design and wrote manuscript. All authors read and approved the final manuscript.


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# Tables

**Table 1. Summary of bioproducts using $CH_4$/ $CO_2$ as the sole carbon source**

| Feedstocks | Products | Titer | Microorganisms | Refs |
|---|---|---|---|---|
| $CH_4$ | Lactic acid | 0.6 g/L | *Methylomicrobium buryatense* | [43] |
| $CH_4$ | Methanol | 59.9 mg/L | *Methylotrophus capsulatus* | [44] |
| $CH_4$ | Carbohydrate | 8.24 g/L | *Methylomicrobium buryatense* | [45] |
| $CO_2$ | Ethanol | 5.50 g/L | *Synechocystis sp.* PCC6803 | [46] |
| $CO_2$ | FAME | 0.31 g/L | *Nannochloropsis gaditana* | [47] |
| $CO_2$ | isobutanol | 911 mg/L | *Synechocystis PCC* 6803 | [16] |
| $CO_2$ | Carbohydrate | 3.81 g/L | *Chlorella vulgaris* FSP-E | [48] |

**Table 2. Costs of raw materials used in the base case study.**

| Raw materials | Cost | Unit | Ref. |
|---|---|---|---|
| **Inputs** | | | |
| Biogas | 130.3 | $/ton | [49] |
| Carbon dioxide | 74.1 | $/ton | [50] |
| Natural gas | 182.5 | $/ton | [51] |
| Ammonia | 431 | $/ton | [27] |
| Flocculant | 9670 | $/ton | [52] |
| Electricity | 0.1 | $/KW | [53] |
| Water | 0.2 | $/ton | [54] |
| Cooling tower chemicals | 3,372 | $/ton | [55] |
| Sludge disposal cost | 15.9 | $/ton | [56] |
| **Outputs** | | | |
| Electricity credit | 0.1 | $/KW | [53] |
| $CO_2$ credit | 30.2 | $/ton | [57] |
| AD sludge N credit | 271 | $/ton | [32] |

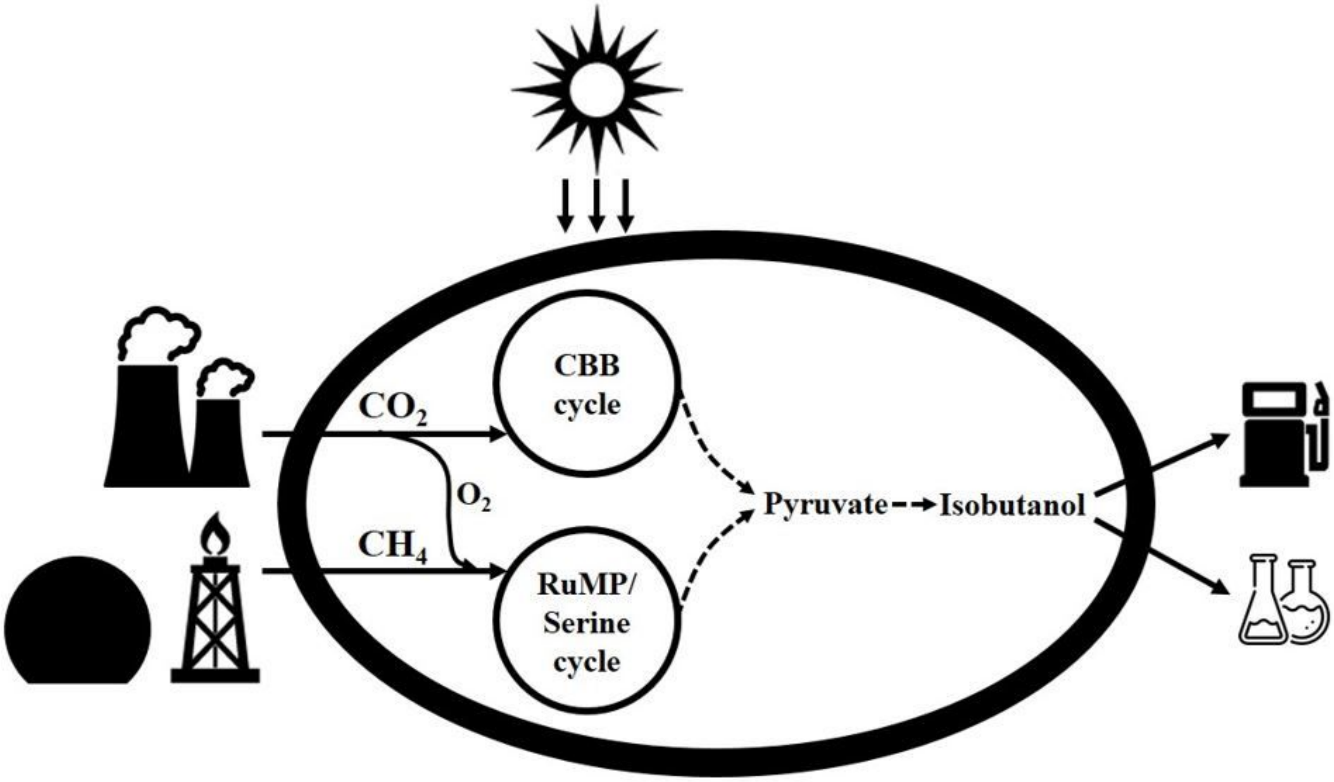
**Table 3. Assumptions for n$^{th}$-plant isobutanol produciton**

| Description of assumption | Value |
|---|---|
| Internal rate of return (IRR) | 10% |
| Plant financing by equity | 50% |
| Plant life | 30 years |
| Income tax rate | 21% |
| Interest rate for debt financing | 8% |
| Term for debt financing | 10 years |
| Working capital cost | 5% of fixed cost investment (FCI) |
| Land purchase cost | 1% of fixed cost investment (FCI) |
| Depreciation schedule | 7-year MACRS schedule |
| Plant salvage value | No value |
| Start-up time | 6 months |
| Revenue and cost during startup | Revenue = 50% of normal<br>Variable costs = 75% of normal<br>Fixed costs = 100% of normal |
| Operating hours per year | 7,920 |

**Table 4. Comparisons of operating costs in three scenarios with an annual production of 50000 tons**

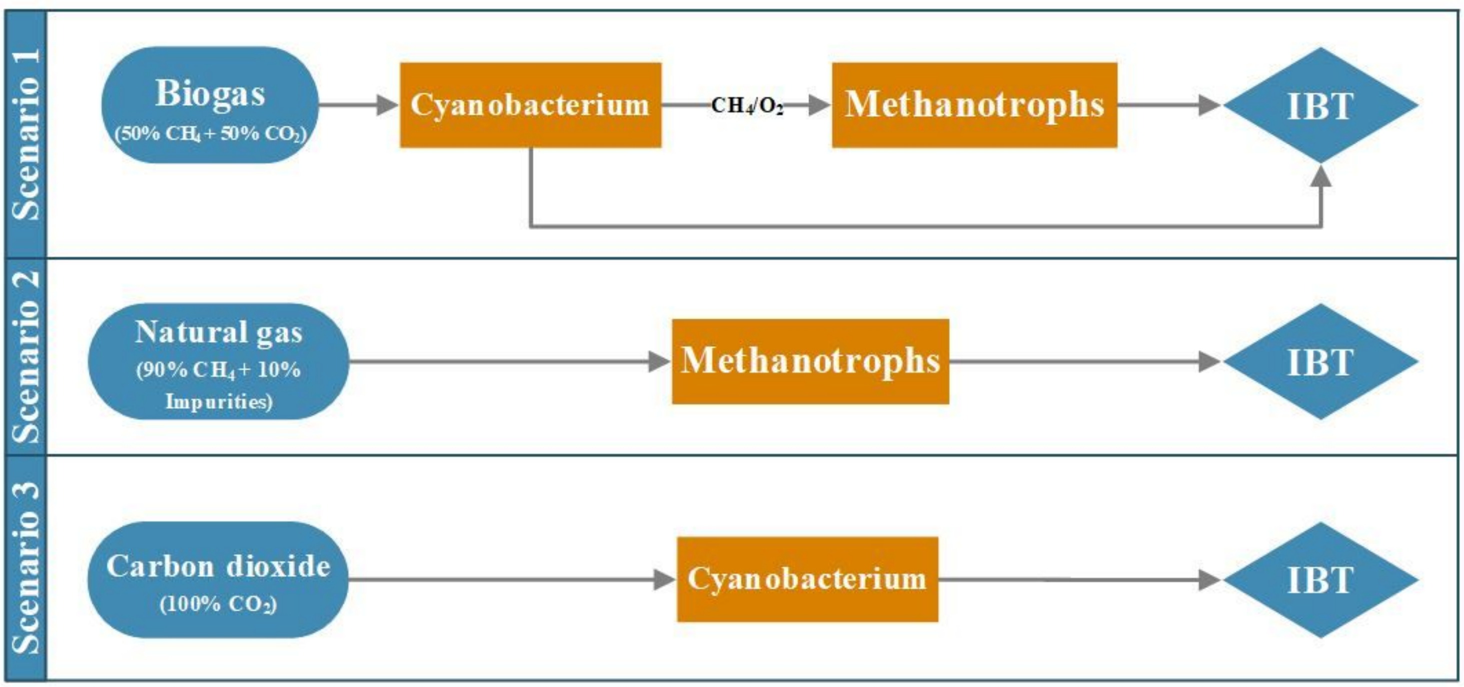
| Manufacturing Costs | | | |
|---|---|---|---|
| Item | Annual cost (MM$/yr) | | |
| Variable operating costs-raw materials | Scenario #1 | Scenario #2 | Scenario #3 |
| Gaseous feedstock | 24.71 | 30.32 | 17.61 |
| Ammonia | 2.41 | 2.28 | 3.24 |
| Flocculant | 12.75 | 12.84 | 13.05 |
| Sludge disposal cost | 13.13 | 14.30 | 14.44 |
| Makeup water | 0.18 | 0.15 | 0.20 |
| Cooling tower chems | 0.06 | 0.06 | 0.06 |
| Electricity | 0 | 2.34 | 0 |
| Sum of variable operating cost of raw materials | 53.23 | 62.29 | 48.60 |
| Variable operating costs-byproduct credits | | | |
| AD sludge N credit | 1.13 | 0.96 | 1.27 |
| $CO_2$ credit | 1.35 | 4.77 | 0 |
| Grid electricity | 0.82 | 0 | 1.44 |
| Sum of byproduct credits | 3.30 | 5.73 | 2.71 |
| Total variable operating costs (VOC) | 49.93 | 56.56 | 45.89 |
| Fixed operating costs | | | |
| Salaries | 2.33 | 2.33 | 2.33 |
| Labor burden | 1.40 | 1.40 | 1.40 |
| Facility maintenance | 4.00 | 4.50 | 3.58 |
| Property insurance & tax | 2.29 | 2.30 | 1.89 |
| Sum of fixed operating costs (FOC) | 10.02 | 10.53 | 9.20 |
| Total operating cost (VOC+FOC) | 59.95 | 67.09 | 55.09 |

# Figures



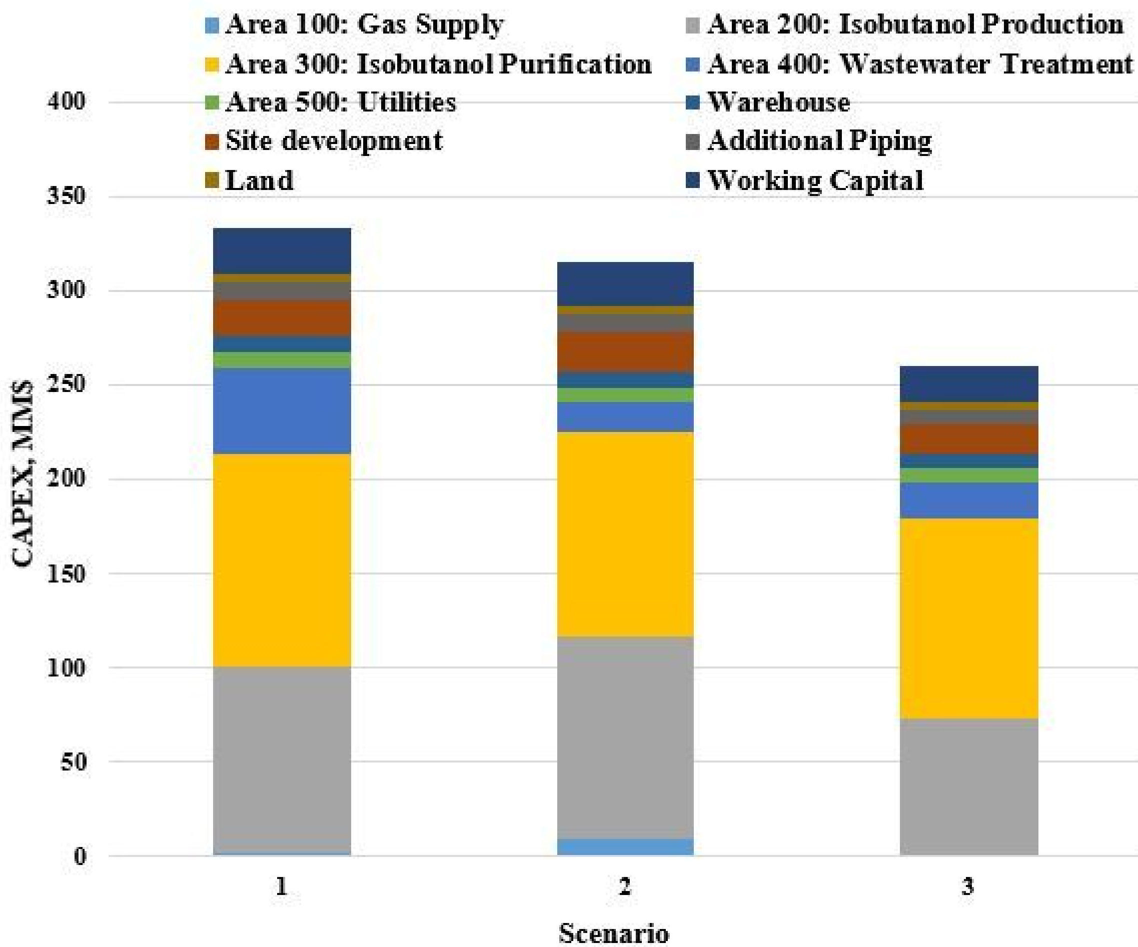

## Figure 1

Simplified pathway of isobutanol production from CH4 and CO2.



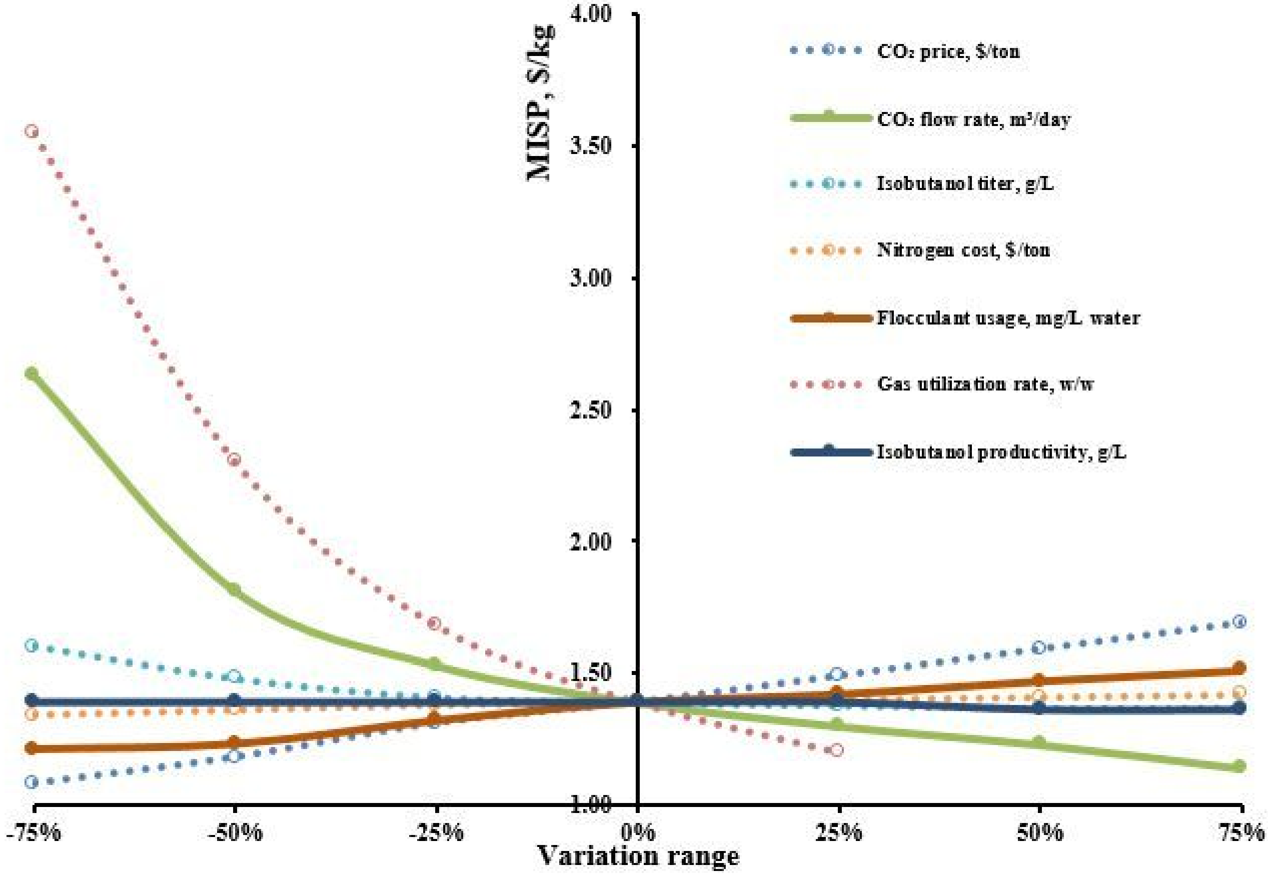

## Figure 2

Three process scenarios of isobutanol production.



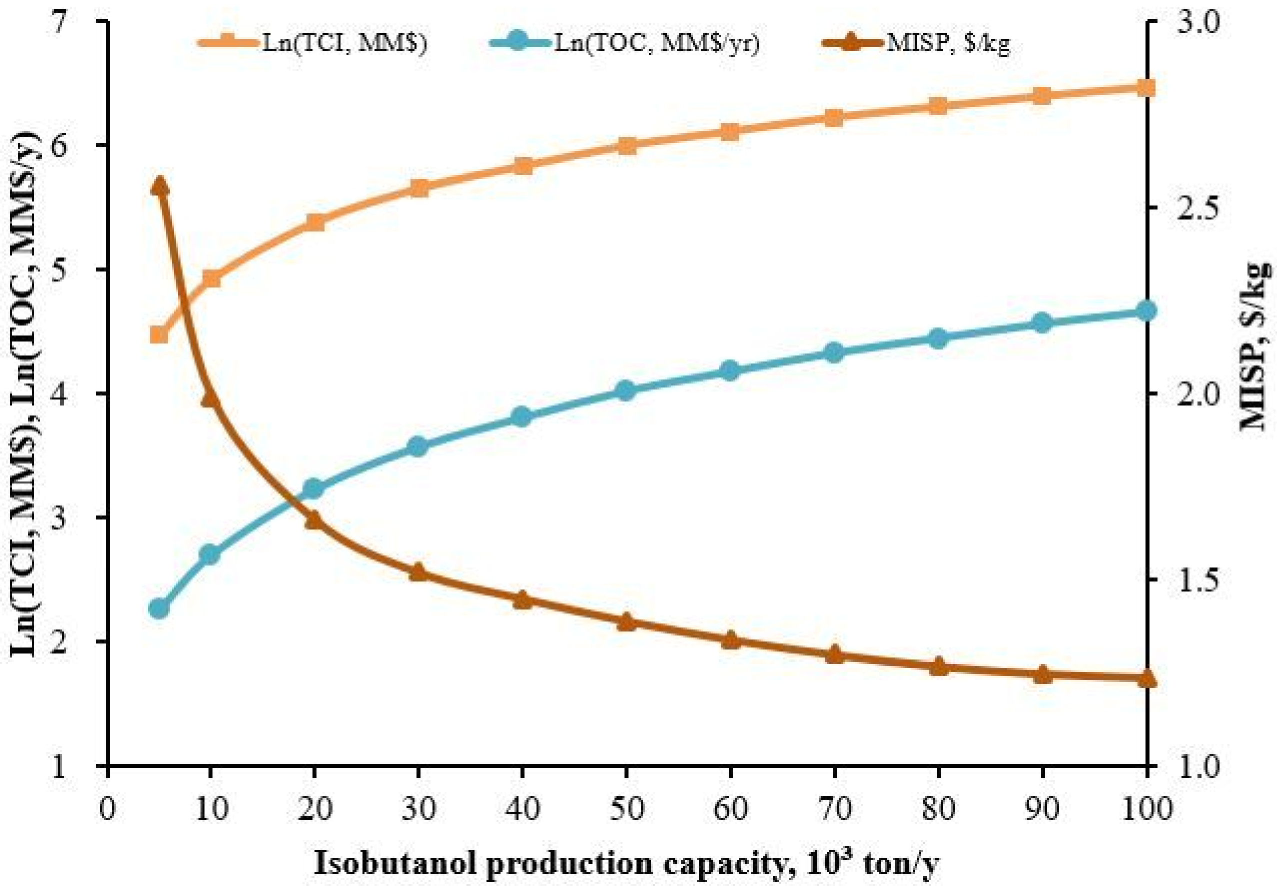

## Figure 3

Capital cost comparison of three scenarios with an annual production of 50,000 tons



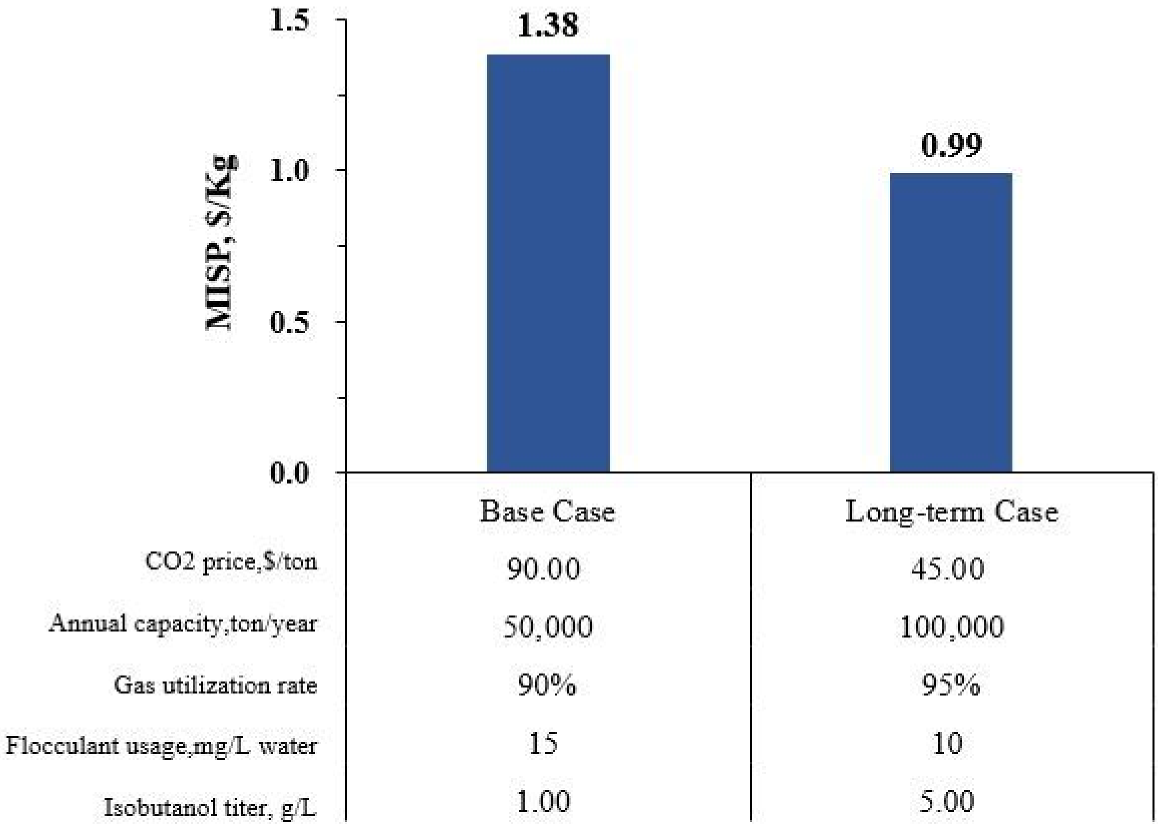

## Figure 4

Single-point sensitivity analysis on scenario 3.




## Figure 5

Effects of isobutanol production capacity on MISP, CAPEX, OPEX production.




## Figure 6

MISP of various prospective targets in scenario 3.