Life cycle design of bioprocess system applying simulation-based approach

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Abstract

In this study, we are tackling the development of a simple dynamic model of bioprocesses that enables life cycle design through computer-aided simulation. The material and energy balances of bioprocesses are significantly affected by scale. The dynamic model considers changes in microbial growth and metabolic production over time and the associated energy balance, such as heating and cooling, as the fermenter is scaled up. The dynamic model developed in this study will theoretically enable efficient bioprocess design by predicting future productivity and environmental impacts and their hotspots and bottlenecks on a commercial scale from early-stage research data and feeding them back to basic laboratory-scale research. As a case study, we simulated the ethanol fermentation process using a prototype of the dynamic model developed in this study. As a result, we confirmed no significant differences in the rates of yeast growth or ethanol production depending on the scale of the fermenter. Still, there were substantial differences in the heat balance. Furthermore, a gate-to-gate LCA limited to the ethanol fermentation process revealed that scaling up from a 5 L jar fermenter to a 50 kL commercial-scale fermenter reduced Greenhouse gas (GHG) emissions per product by 88 % and visualized the impact of fermenter scale on GHG emissions hot spots.

**Keywords**: microorganism, scale up, prospective life cycle assessment

* 1. Introduction

The global decarbonization trend accelerates research and development on plant-derived products using bioprocesses (Yaashikaa *et al.,* 2023). Bioprocesses utilize enzymatic reactions and other biochemical reactions by microorganisms. They are widely used in various industries, such as pharmaceuticals (Jones and Gerogiorgis, 2022), foods (Shimada *et al.,* 2021), bioplastics (Kikuchi *et al.*, 2022), and biofuels (Ohara *et al.*, 2012). The life cycle design of bioprocesses often differs from that of the chemical industry because it deals with plant-derived raw materials and complex reactions by microorganisms. In particular, the process's scale significantly affects the material and energy balance of a process such as fermentation, in which microbial growth and metabolite production occur simultaneously (Imamoglu and Sukan, 2013). For example, the processes of media preparation, sterilization, incubation, separation, and purification at the laboratory scale (flasks, jars) significantly differ from the plant scale regarding energy, utility use, and productivity. Therefore, it is common in bioprocess technology development to conduct primary research at the lab scale followed by scale-up to the plant scale for validation. However, there is a concern with this conventional method of developing bioprocesses. If environmental or economic problems are found during the demonstration stage in a large-scale plant, the process will revert to the primary research level. In chemical processes, models have been proposed that can quantitatively forecast the material balance, energy balance, and environmental impact of future scale-up in the lab-scale phase of research (Piccinno *et al.*, 2016). Quantitative models of microbial growth and metabolite productivity have been reported for the bioprocesses (Luong *et al.*, 1988). Still, no models have been reported, including energy consumption at each scale and its hot spots.

In this study, we develop a simple dynamic model that enables computer-aided simulation of bioprocesses' material and energy balance. This dynamic model would allow efficient bioprocess design by forecasting future productivity, environmental impacts, and costs on a commercial scale, as well as their hotspots and bottlenecks, from early-stage research data and feeding them back into basic laboratory-scale research. First, we will develop a model for each culture tank from jar fermenter scale (5 L) to pilot plant scale (500 L) and commercial scale (50 kL), taking as an example the ethanol production process, which is a typical bioprocess. Furthermore, as a case study, a gate-to-gate LCA (Lifecycle Assessment) is performed to clarify the influence of the scale of the culture tank on the environmental load per product and its hotspot and the usefulness of dynamic models in bioprocess life cycle design is discussed.

* 1. Materials and methodologies
     1. Modeling of fermenter in bioprocess

Depending on the strain type, microbial reactions differ in conditions (e.g., growth rate, fermentation rate, resistance to fermentation inhibitors, optimum temperature, etc.). This study selected ethanol fermentation by general yeast (ex. *Saccharomyces cerevisiae*), which has been reported at various scales (Ouchida *et al.*, 2017), as the first model. Simulations were performed in an Excel-based in-house code based on the microbial kinetics model, in which the medium was heated after the start of incubation, yeast growth started at 35 °C, and the total fermentation time was set to 100 h. The initial glucose concentration of the medium was set at 100 g/L, and it was assumed that all glucose was consumed by yeast growth and ethanol fermentation. The final yeast yield was set at 90 g-dry/mol-glucose. The Monod equation shown by Eq. (1) determined the yeast growth rate and ethanol productivity from the concentration of yeast and glucose in the medium (Nosrati-Ghods *et al.*, 2020).

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| --- | --- |
| , | (1) |

where *μ* is the specific yeast growth rate [h-1], *μmax* is the maximum yeast growth rate [h-1], *S* is the substrate concentration [g/L], *ν* is the specific ethanol productivity [h-1], *νmax* is the maximum ethanol productivity [h-1], *Ks* and *Kp* are substrate utilization constant [g/L] equal to substrate concentration when *μ* and *ν* are half of *μmax* and *νmax*, respectively.

The heat balance considered direct heating and cooling of the fermenter, fermentation heat, heat dissipation from the walls, and heat loss due to ventilation. Heating and cooling conditions were different for each scale. The fermentation heat was calculated by Eq. (2).

|  |  |
| --- | --- |
|  | (2) |

where ∆*Hc* is fermentation heat [J], ∆*Hr* is glucose consumption heat [J], ∆*Hα* is the heat of yeast combustion [J/g], ∆*X* is yeast growth [g], ∆*Hp* is the heat of products combustion [J/g], and ∆*P* is products amount [g].

Heat dissipation from the fermenter wall was calculated assuming natural convection from the wall. Heat loss due to aeration was calculated assuming cooling by evaporative latent heat based on the saturated vapor pressure at the medium temperature and the assumption that 90 % of the saturated vapor content of the aerated air evaporates.

* + 1. Scale-up fermenter configuration

The ethanol fermenter was step-wise scaled up from a 5 L jar fermenter to a 500 L pilot-scale and 50 kL commercial-scale fermenter, as shown in Table 1 and Fig. 1. The 5 L jar fermenter was modeled after the Middle Scale Bioreactor BMS-P manufactured by ABLE Corporation. An electric heater (capacity: 0.15 kW) was wrapped around the glass vessel, and the heater surface temperature was calculated by considering the heat transfer between the heater and the outside air, and the net heat added to the liquid medium in the vessel was calculated. The liquid and vessel were assumed to be heated by heat transfer between the heater and the glass, and the heater was assumed to be controlled to maintain the target temperature by on/off control. For cooling, a chiller (0.05 kW) was constantly operated to maintain the target temperature by on/off control. 500 L and 50 kL fermenters were modeled as stainless-steel tanks with jackets attached to the outer walls. The fermenters were maintained at the target temperature by on/off control of heating with 60 °C hot water passing through the jacket and cooling with 20 °C cold water passing through the jacket. The flow rate of hot and cold water was set at 60 kg/h. The minimum flow rate of hot water was literately determined to raise the liquid temperature to the target temperature based on the heat balance. Also, assuming that yeast growth is completed in 40 h after the liquid reaches the target temperature, in the 500 L and 50 kL scale, the aeration was assumed to be stopped when yeast growth was saturated.

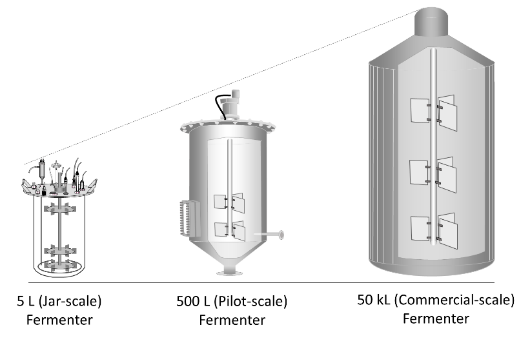
Table 1. Fermenter configurations modeled at each scale

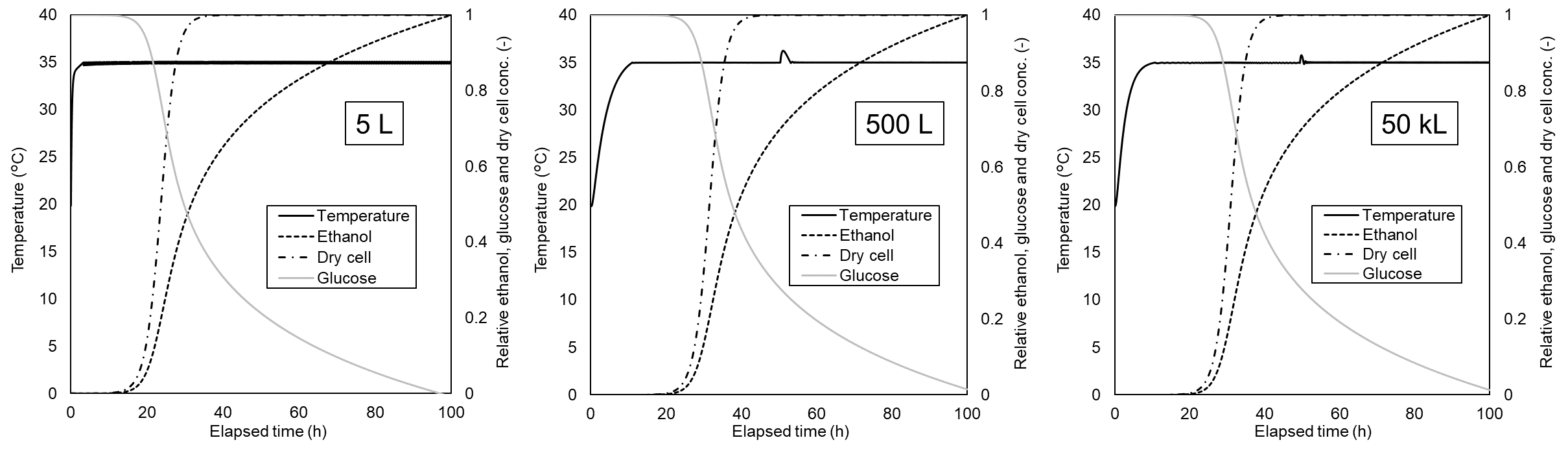
Fig.1 Overview of fermenter scale-up

* + 1. Setting for gate-to-gate LCA

Gate-to-gate LCA was performed to determine the environmental load per product at each fermenter scale and the impact of its hot spots. The system boundary is from the start to the end of the fermentation process, and only the energy-derived environmental impact input to the fermentation process is considered, excluding the environmental impact related to the production of raw and auxiliary materials, disposal of by-products, etc. The environmental load item was the Global Warming Potential (GWP), calculated as CO2-equivalent. The functional unit was 1 kg of ethanol contained in the fermentation liquid. Foreground data for ethanol fermentation were taken from Ouchida *et al.* (2017), and background data were taken from IDEA v2.2, a Japanese LCA database.

* 1. Results and discussion
     1. Simulation of fermenter at each scale

Simulated fermentation profiles showing relative values for glucose consumption, yeast growth, ethanol production, and medium temperature at 5 L, 500 L, and 50 kL scales are shown in Fig. 2. Starting from a medium temperature of 20 °C, there was a difference in scale in the rate at which the medium reached the target temperature of 35 °C. This was attributed to differences in heating method and heating capacity. Since the heating capacity depends on the model settings, for example, at plant scale, the time to reach the target temperature can be easily reduced by increasing the amount of hot water passed through the jacket or by raising the hot water temperature. In terms of kinetics, ethanol production started after 3.3 h in the 5 L jar and after 11.0 h in the 500 L and 50 kL fermenters. This is due to the time required to reach the target temperature. The 500 L and 50 kL scales designed under the same conditions of heating and cooling showed the same performance as expected. The slight temperature increase after about 50 h in the 500 L and 50 kL was due to disappearing the latent heat of vaporization loss by stopping aeration after yeast growth saturation, and it took time for cooling.



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自動的に生成された説明Fig.2 Simulated fermentation profiles at each scale of fermenter

Fig.3 Simulated heat balance at each scale of fermenter

Figure 3 shows the results of the heat balance at each scale. The graph's vertical axis is expressed as relative values, with the maximum heating value as 100 and negative values for cooling. In the 5 L scale, heating, cooling, and heat dissipation from the glass vessel were relatively large. In contrast, heat loss from fermentation and latent heat of evaporation due to aeration were slight. In the 500 L and 50 kL scales, heating, heat loss due to hot water discharge, and latent heat loss due to aeration were relatively significant in the early fermentation stage. In the late stages of fermentation, heat loss decreased because aeration was set to stop when yeast growth was saturated.

* + 1. Greenhouse gas emissions from fermentation process at each scale

The results of the gate-to-gate LCA limited to the ethanol fermentation process are shown in Fig. 4. The results are shown as relative emissions values at each scale, with the total emissions at the 5 L scale as 100 %. The Greenhouse gas (GHG) emissions per weight of ethanol were reduced by 87.0 % and 88.5 % for the 500 L and 50 kL scales, respectively, compared to the 5 L scale. In other words, GHG emissions were reduced by scale-up.

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自動的に生成された説明In the 5 L jar scale, GHG emissions from electricity consumption in the chiller accounted for 42.5 % of the total GHG emissions. In contrast, emissions from electricity consumption in the compressor for aeration, heater, and agitation were 19.5 %, 19.0 %, and 19.1 %, respectively. Since the chiller of the jar fermenter is in constant operation, its electricity consumption is significant. Furthermore, the heater was controlled on/off while the cooling chiller was constantly running, resulting in higher emissions from the heater power. The 500 L and 50 kL stainless steel fermenters have a larger diameter than the glass 5 L jar fermenter, resulting in lower heat dissipation. Therefore, once the target temperature is reached, energy consumption for heating and cooling is lower, resulting in lower GHG emissions derived from heating and cooling. In the scale-up from 500 L to 50 kL, the environmental impact from heating is slightly lower because the heat dissipation is reduced due to the larger body diameter. In the 500 L and 50 kL scales, GHG emissions were mainly attributable to compressor power consumption for aeration. This amount was equivalent to that of the 5 L jar scale. This is because the model was set up assuming that the ratio of dissolved oxygen required for yeast growth and ethanol production would be the same regardless of scale. GHG emissions derived from stirring power decreased with scale-up. This is because the mechanical loss of the stirring device becomes relatively smaller with a larger capacity, resulting in smaller stirring power per liquid volume [kW/m3] (Murakami *et al.*, 2000).

Fig.4 Relative greenhouse gas emissions and their breakdown at each scale of fermenter

In this case study, the simulation-based scale-up enabled visualization of the total GHG emissions and hot spots as shown in Fig. 4. Scale-up prediction in the early stages of the study can provide feedback on improvement points from environmental and economic aspects at the commercial scale, which will enable appropriate process design.

* 1. Conclusions

This study developed a prototype of a fermenter model that contributes to the bioprocesses' simulation-based prospective life cycle design. Further improvement of the accuracy of the dynamic model will enable simulation of the material and energy balance of the fermenter from laboratory-scale to commercial scale. It will help design bioprocesses for higher productivity and lower environmental impact requirements.

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