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Evaluation of Productivity of Zymotis Solid-State Bioreactor Based on Total Reactor Volume

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Summary

In this work a method of analyzing the performance of solid-state fermentation bioreactors is described. The method is used to investigate the optimal value for the spacing between the cooling plates of the Zymotis bioreactor, using simulated fermentation data supplied by a mathematical model. The Zymotis bioreactor has good potential for those solid-state fermentation processes in which the substrate bed must remain static. The current work addresses two design parameters introduced by the presence of the internal heat transfer plates: the width of the heat transfer plate, which is governed by the amount of heat to be removed and the pressure drop of the cooling water, and the spacing between these heat transfer plates. In order to analyze the performance of the bioreactor a productivity term is introduced that takes into account the volume occupied within the bioreactor by the heat transfer plates. As part of this analysis, it is shown that, for logistic growth kinetics, the time at which the biomass reaches 90 % of its maximum possible value is a good estimate of the optimum harvesting time for maximizing productivity. Application of the productivity analysis to the simulated fermentation results suggests that, with typical fast growing fungi (μ = 0.324 h^{-1}), the optimal spacing between heat transfer plates is of the order of 6 cm. The general applicability of this approach to evaluate the productivity of solid-state bioreactors is demonstrated.

Key words: Zymotis bioreactor, packed-bed bioreactors, volumetric productivity, solid-state fermentation, modeling, large scale cultivation, heat transfer

Introduction

Solid-state fermentation (SSF) involves the growth of microorganisms on water insoluble substrates in the absence of visible water between the substrate particles. This cultivation technique has potential to be used at commercial scale for the production of some microbial products, especially in those situations where higher yields or better product qualities are obtained in SSF than in submerged liquid fermentation (SLF) processes.

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One example is the production of fungal spores, which have potential applications as inocula for fermented foods or as biopesticides: the yield of fungal spores is usually higher in SSF than in SLF, and spores produced by SSF are typically more robust than those produced in SLF *(1-3)*. However, in the production of fungal spores by SSF, agitation can retard growth and damage the reproductive hyphae, greatly reducing spore yields *(4,5)*. As a consequence, it is necessary to optimize the design of SSF bioreactors in which the substrate bed remains static throughout the fermentation. Although tray bioreactors can be used, packed-beds are more appropriate because the forced aeration allows some control over fermentation parameters through manipulation of the flowrate and the temperature of the air used in the fermentation *(6)*.

Traditional packed-bed bioreactors do not have internal heat transfer surfaces and at large-scale are sufficiently wide that conduction to the bioreactor walls contributes negligibly to heat removal *(7,8)*. In this situation, the combination of metabolic heat production with the dynamics of convective cooling causes a rise in temperature between the air inlet and the air outlet *(9)*. Experimentally, temperature rises within packed beds of over 20 °C have been recorded *(10)*. Such high temperatures lead to regions of poor growth and product formation within the bioreactor *(6,10-13)* and are therefore undesirable. Unfortunately, strategies such as increasing the aeration rate or decreasing the temperature of the inlet air are only moderately effective: they reduce but do not eliminate the temperature gradients in the bed *(6)*. Furthermore, there are practical and economic limits on the air flowrates and inlet air temperatures that can be used. As a result, there are limitations on the bed heights that can be used for large scale traditional packed beds *(8)*.

Roussos *et al. (14)* proposed a modified packed-bed bioreactor, called »Zymotis«, which has internal heat transfer plates (Fig. 1). Heat removal by these plates minimizes axial temperature gradients, allowing near optimal conditions for growth. However, this design has received little experimental attention and therefore optimal design parameters are not known. Recent mathematical modeling work showed that the bioreactor has the potential to lead to high productivities per unit of bed volume and indicated that it could be the most appropriate bioreactor design for those SSF processes in which the substrate bed must remain static, since it potentially allows quite large bioreactor heights *(15)*.

Three new design parameters are introduced by the incorporation of internal heat transfer plates: the spacing between the internal heat transfer plates, the temperature of the cooling water in the plates, and the thickness of the plates themselves. The model suggests that optimal productivities will be achieved with the Zymotis bioreactor by using a strategy in which the temperature of the cooling water is decreased in response to the increase of temperature at the air outlet end of the bed *(15)*. However, since the model does not take the thickness of the internal heat transfer plates into account, it predicts that optimum productivity will be achieved with zero spacing between the plates, because under these conditions all the substrate can be maintained at the optimum temperature. However, this is clearly not a useful optimum.

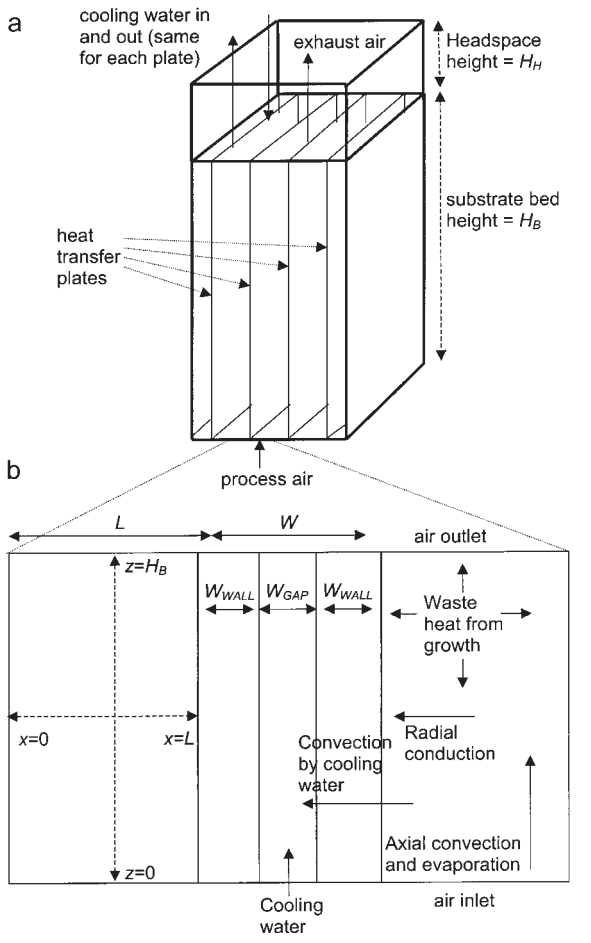

Fig. 1. (a) The Zymotis bioreactor of Roussos *et al. (14)* and (b) details of a repeating unit within the bioreactor, showing two »half-slabs« of substrate on either side of the heat transfer plate

Therefore the current work proposes a method, based on the total bioreactor volume, for characterizing overall productivity of the Zymotis bioreactor, and demonstrates the application of this method in a case study using simulated fermentation data generated by a dynamic model of the bioreactor *(15)*. An optimum plate spacing greater than zero is demonstrated. It is also shown that this approach provides a useful basis for comparing the performance of different SSF bioreactors. Such a comparative measure of performance has not previously been proposed.

Methods

In the current work the model of Mitchell and von Meien *(15)* was used to provide simulated data. The model is reproduced only briefly here, since it has already been described in detail *(15)*. It describes the growth of *Aspergillus niger* on a starchy substrate.

System and assumptions

The Zymotis bioreactor *(14)* is a rectangular packed bed bioreactor, aerated from the bottom with moist air

Table 1. The mathematical model of Mitchell and von Meien *(15)*

Equation		Eq. No.
Biomass growth	$\frac{dX}{dt} = \mu X\left(1 - \frac{X}{X_m}\right)$	/1/
Effect of temperature on specific growth rate	$\mu = \frac{A.\exp\left(\frac{-E_{a1}}{R(T + 273.16)}\right)}{1 + B.\exp\left(\frac{-E_{a2}}{R(T + 273.16)}\right)}$	/2/
Energy balance	$\rho_b C_{pb} \frac{\partial T}{\partial t} + \rho_a (C_{pa} + f\lambda) \frac{\partial T}{\partial z} = k_b \frac{\partial^2 T}{\partial x^2} + \rho_s (1 - \varepsilon) \frac{dX}{dt}$	/3/
Weighted bed properties	$\rho_b = \varepsilon.\rho_a + (1\text{-}\varepsilon)\rho_s$ $k_b = \varepsilon.k_a + (1\text{-}\varepsilon)k_s$ $C_{pb} = (\varepsilon\rho_a(C_{pa} + f.\lambda) + (1\text{-}\varepsilon)\rho_s C_{ps})/\rho_b$	/4a/ /4b/ /4c/
Boundary conditions	$z = 0 \quad T = T_a$ $x = 0 \quad \frac{dT}{dx} = 0$ $x = L \quad k_b \frac{dT}{dx} = -h(T - T_w)$	/5a/ /5b/ /5c/
Initial conditions	at $t = 0 \quad T = T_0 \quad 0 \le z \le H_B$ and $0 \le x \le L$ at $t = 0 \quad X = X_0 \quad 0 \le z \le H_B$ and $0 \le x \le L$	/6a/ /6b/
Control strategy	$T_w = T_{opt} - F.(T_{LHB} - T_{opt})$	/7/

(Fig. 1a). During the process the substrate bed remains static. The outer casing is assumed to be insulated, such that there are no temperature gradients from front to back in the bioreactor. The system modeled is a repeating unit within this bioreactor (Fig. 1b). This repeating unit extends from the central plane halfway between two plates, across the heat transfer plate, to the central plane halfway between the two plates that make the adjacent compartment. The plate in the center of this repeating unit is reponsible for removing heat from the two »half-slabs« of fermenting substrate on either side of it.

The model describes heat transfer but not mass transfer. Sangsurasak and Mitchell *(13)* discussed most of the assumptions in the model, in the context of heat transfer in a traditional packed-bed. Since their model described well the experimental data of Ghildyal *et al*. *(10)* and Saucedo-Castaneda *et al*. *(16)*, the assumptions are accepted as reasonable.

Dynamic heat transfer model

The model, consisting of Equations /1/ to /7/, is shown in Table 1. Growth is assumed to follow logistic growth kinetics, with the specific growth rate constant expressed empirically as a function of temperature *(16)*. The only heat transfer processes taken into account are convection and evaporation in the vertical direction and conduction in the horizontal direction. The factor $f\lambda$ characterizes evaporative heat removal, assuming that the air and the moist solid at any particular location within the bed are in thermal and moisture equilibrium *(13)*.

Values for density, thermal conductivity and heat capacity of the bed are calculated as weighted averages of the properties of the air and substrate within the bed. Density and thermal conductivity are volume-weighted, while heat capacity is mass-weighted. Implicit in these equations is the assumption that the thermal properties of the microbe are equal to those of the substrate and that the void fraction does not change with time.

The boundary conditions correspond to the bottom of the bed being maintained at the inlet air temperature, the absence of heat transfer through the central plane between two cooling plates, and convective heat transfer from the edge of the bed to the cooling water. The initial conditions correspond to an even inoculum concentration (X_o) and initial temperature (T_o) at all points in the column. Since the bioreactor performs best when the temperature of the cooling water is varied in response to the temperature measured at the top of the bed midway between two heat transfer plates (T_{LHB}) *(15)*, this control strategy was used in the simulations in the current work.

The parameters used in the model are in Table 2 *(15–22)*. They were estimated for the growth of *Aspergillus niger* on a starchy substrate in a packed-bed bioreactor *(8)*, the system used by Saucedo-Castaneda *et al*. *(16)*. A value of 2 for the control factor F in Equation /7/ gives good performance, while allowing cooling water temperatures that avoid the need for refrigeration *(15)*. The equation system was converted into dimensionless form and solved, using the method of characteristics, by the method of orthogonal collocation, applied in both the vertical and horizontal dimensions *(15)*. The resulting differential-algebraic system was solved using the DASSL routine *(23)*.

Productivity analysis

The performance of the bioreactor is analyzed on the basis of the productivity of biomass, that is, the quantity of biomass produced per unit time and volume. Given the spatial heterogeneity within the bioreactor during the periods of static operation, it is necessary to calculate a volume-weighted biomass concentration,

which is done on the basis of weights calculated according to Gauss' quadrature *(15).*

The analysis is done for an organism showing logistic growth kinetics:

$$X = \frac{X_m}{1+\left(\frac{X_m}{X_0}-1\right)e^{-\mu_{opt}t}} \qquad /8/$$

where X is the volume-weighted biomass concentration in terms of kg of biomass per kg of initial substrate.

The productivity of biomass production at any time during the fermentation can be plotted as:

$$P_S = \frac{X - X_0}{t_T + t_L + t} = \frac{X - X_0}{t_N + t} \qquad /9/$$

where t is the time which has elapsed since the microorganism entered the active growth phase, t_L is the length of the lag phase, and t_T is the turnaround time, that is, the time required to empty the bioreactor, clean it and charge it with the next load of inoculated substrate. Since both the lag phase and the turnaround time are nonproductive, they are added together and the period is denoted t_N (*i.e.* nonproductive). Note that P_S has the units of kg of biomass produced per kg of substrate initially present per hour, and the subscript 'S' denotes that it is on the basis of the amount of substrate, to distinguish it from the volumetric productivity.

A model of the growth kinetics allows calculation of the values of P_S that would occur if growth were biologically-limited and not limited by transport phenomena. This gives a standard, denoted P_{SB}, against which the performance of fermentation runs can be compared. For an organism exhibiting logistic growth kinetics, this can be obtained by substituting Equation /8/ into Equation /9/:

$$P_{SB} = \frac{\dfrac{X_m}{1+\left(\frac{X_m}{X_0}-1\right)e^{-\mu_{(T=opt)}t}} - X_0}{t + t_N} \qquad /10/$$

If Equation /10/ is plotted against time, a maximum value will be obtained. This maximum possible productivity, which depends on the values of X_o, X_m, $\mu_{(T=opt)}$ and t_N, is denoted P_{SBmax}.

Equations /9/ and /10/ express the productivity on the basis of the mass of substrate present. The volumetric productivity (P_V) of the bed, that is, taking into account only the volume occupied by the substrate itself, is:

$$P_V = \frac{(X - X_0)\rho_b}{t + t_N} = \frac{(X - X_0)(1-\varepsilon)\rho_S}{t + t_N} \qquad /11/$$

where P_V has the units of kg of biomass per cubic meter of bed per hour. The bed density is easily measured experimentally. It can also be calculated as the volume fraction occupied by the substrate ($1-\varepsilon$) times the density of the substrate itself (ρ_s) *(15).*

The volumetric productivity can also be expressed on the basis of the total volume occupied by the bioreactor. The fraction of the total volume of the Zymotis bioreactor occupied by the substrate bed (V_F) is given by:

$$V_F = \frac{V_B}{V_F + V_P + V_H} = \frac{2LH_BD}{2LH_BD + WH_BD + (2L+W)H_HD} =$$

$$= \frac{1}{\left(1+\frac{W}{2L}\right)\left(1+\frac{H_H}{H_B}\right)} \qquad /12/$$

where V_B,V_P and V_H are the volumes occupied by the bed, plates and headspace, respectively, H_B and H_H are the heights of the bed and headspace, respectively, D is the depth from front to back, L is half of the distance between heat transfer plates and W is the width of each plate. The factor of 2 in the expression for V_B arises because, with the plate layout shown in Fig. 1a, a repeating unit of bioreactor consists of 2 half-slabs of substrate of width L, one on either side of one heat transfer plate. Roussos *et al. (14)* did not specify the headspace volume of their Zymotis bioreactor. Since large headspaces are not necessary in packed-beds, a headspace equal to $0.1H_B$ is assumed.

The productivity of the bioreactor based on the overall volume of the bioreactor is therefore:

$$P_B = V_F \cdot P_V = \frac{1}{\left(1+\frac{W}{2L}\right)\left(1+\frac{H_H}{H_B}\right)} \cdot \frac{(X - X_0)(1-\varepsilon)\rho_S}{t + t_N} \qquad /13/$$

Note that the units of P_B are kg of biomass per hour per cubic meter of bioreactor volume.

Results and Discussion

In the current work optimal performance is considered in terms of the production of biomass, using simulated growth data supplied by the bioreactor model. Although the analysis of performance is done with simulated data, it can be applied equally as well to actual fermentation data. Furthermore, the same concepts can be applied to situations where the desire is to characterise production of a product other than biomass, as long as either kinetic equations for product formation or experimental product concentration profiles are available. The focus on biomass production is simply used as a means to illustrate the concepts.

Productivity per kilogram of substrate

The aim of this section is to identify the time of harvesting of the batch fermentation that will lead to the maximum productivity of the process. The dynamic heat transfer model was used to generate simulated results for the volume-weighted biomass concentration, using the parameter values in Table 2. These results were substituted into Equation /9/ to calculate the substrate-based productivity (P_S) as a function of time (Fig. 2). This is compared against the value of P_S for logistic growth with $\mu = \mu_{(T=opt)}$. In both cases the value of t_N was taken as 10 h.

The maximum productivity obtained during the process can be determined by inspection of Fig 2. In fact, determination of the maximum productivity does not require the mathematical model since it is simple to

determine the maximum productivity of the fermentation graphically. In real fermentations the challenge is to remove sufficient samples from a heterogeneous bed to give a good estimate of the volume-weighted biomass concentration.

Maximum possible substrate-based productivity

Substituting the values for X_m, X_o and $\mu_{(T=opt)}$ given in Table 2 into Equation /10/ gives that P_{SBmax}, the maximum possible value of P_{SB} will be achieved at the time at which $X = 0.113$, which corresponds to $0.904X_m$. The

Table 2. Description of symbols and listing of parameter values used in the model and other calculations

Symbol	Description	Value	*Ref.*
A	Frequency factor in equation /2/	7.483×10^7 s^{-1}	*(16)*
B	Constant in equation /2/	1.300×10^{47}	*(16)*
C_{pa}	Heat capacity of air	1180 J kg^{-1} °C^{-1}	*(17)*
C_{pb}	Heat capacity of the bed	J kg^{-1} °C^{-1} (Equation /4c/)	
C_{ps}	Heat capacity of moist substrate	2500 J kg^{-1} °C^{-1}	*(18)*
C_{PW}	Heat capacity of water	4184 J kg^{-1} °C^{-1}	*(17)*
D	Depth of bioreactor from front to back	1 m	
E_{a1}	Activation energy in numerator of equation /2/	70225 J mol^{-1}	*(16)*
E_{a2}	Activation energy in denominator of equation /2/	283356 J mol^{-1}	*(16)*
f	Change in water carrying capacity of air with temperature $\frac{m(\text{water})}{m(\text{air})T}$	0.00246 kg kg^{-1} °C^{-1}	*(17)*
F	Factor used in the scheme for controlling cooling water temperature	2	*(15)*
G	Mass flow rate of cooling water	kg s^{-1} (Equation /18/)	
h	Overall coefficient for convective cooling at the heat transfer plate	95 W m^{-2} °C^{-1}	*(16)*
H_B	Overall height of the bed	2.5 m	*(15)*
H_H	Height of headspace	0.25 m	
H_P	Height of the heat transfer plate	2.5 m	
k_a	Thermal conductivity of the air phase	0.0206 W m^{-1} °C^{-1}	*(19)*
k_b	Thermal conductivity of the bed	W m^{-1} °C^{-1} (Equation /4b/)	
k_s	Thermal conductivity of the substrate phase	0.3 W m^{-1} °C^{-1}	*(20)*
L	Half of the distance between two heat-transfer plates (»plate half-spacing«)	0.03 m	*(15)*
L_c	Characteristic length	m (Equation /20/)	
P_B	Productivity of the bioreactor based on total bioreactor volume $\frac{m(\text{biomass})}{V(\text{bioreactor})t}$	kg m^{-3} h^{-1} (Equation /13/)	
P_S	Substrate-based productivity $\frac{m(\text{biomass})}{m(\text{initial substrate})t}$	kg kg^{-1} h^{-1} (Equation /9/)	
P_{SB}	Substrate based productivity when growth is biologically limited throughout the growth phase $\frac{m(\text{biomass})}{m(\text{substrate})t}$	kg kg^{-1} h^{-1} (Equation /10/)	
P_{SBmax}	Maximum value of P_{SB} reached during the growth phase $\frac{m(\text{biomass})}{m(\text{substrate})t}$	kg kg^{-1} h^{-1}	
P_V	Productivity based on the volume of the substrate bed $\frac{m(\text{biomass})}{V(\text{bed})t}$	kg m^{-3} h^{-1} (Equation /11/)	
Q_{duty}	Maximum required rate of heat removal per heat transfer plate	W (Equation /16/)	
q_{peak}	Peak volumetric rate of waste metabolic heat production	J m^{-3} h^{-1} (Equation /15/)	
R	Universal gas constant	8.314 J mol^{-1} °C^{-1}	*(17)*
Re	Reynolds number	Equation /21/	
t	Time since growth commenced	independent variable s	
t_{90}	Time for the volume-averaged biomass concentration to reach 90 % of X_m	determined by inspection of results h	
t_{cycle}	Time of a total fermentation cycle	h	
t_L	Length of the lag phase	10 h	
t_N	Total nonproductive time	h	
t_T	Turnaround time (time for emptying, cleaning and preparing the bioreactor)	h	
T	Bed temperature	dependent variable in °C	
T_a	Inlet air temperature	38 °C	*(15)*
T_{LHB}	Temperature at $x = L$ and $z = H_B$	determined from model output in °C	
T_o	Initial bed temperature	38 °C	*(15)*
T_w	Cooling water temperature	°C (Equation /7/)	
T_{opt}	Optimum temperature for growth	38 °C	*(16)*
v	Superficial velocity of water flowing through the heat transfer plate	m s^{-1} (Equation /19/)	
V_z	Superficial air velocity	0.01 m s^{-1}	*(16)*
V_B	Volume occupied by the bed	m^3	
V_F	Fraction of the total bioreactor volume occupied by the bed	m^3 (Equation /12a/)	

Symbol	Description	Value	*Ref.*
V_H	Volume occupied by the bioreactor headspace	Calculated m^3	
V_N	Total nonproductive volume within the bioreactor	m^3	
V_P	Volume occupied by the heat transfer plates	Calculated m^3	
W	Total width of the heat transfer plate	0.01 m	
W_{GAP}	Width of the internal gap of the heat transfer plate	0.004 m	
W_{WALL}	Thickness of the walls of the heat transfer plate	0.003 m	
x	Horizontal position within bed	independent variable m	
X	Biomass concentration $\frac{m(\text{biomass})}{m(\text{initial substrate})}$	dependent variable kg kg^{-1}	
$X_{harvest}$	Biomass concentration at harvest $\frac{m(\text{biomass})}{m(\text{initial substrate})}$	kg kg^{-1}	
X_o	Initial biomass concentration	0.001 kg kg^{-1}	*(16)*
X_m	Maximum possible biomass concentration $\frac{m(\text{biomass})}{m(\text{initial substrate})}$	0.125 kg kg^{-1}	*(21)*
Y	Growth heat yield coefficient $\frac{E}{m(\text{biomass})}$	8.366 x 10^6 J kg^{-1}	*(16)*
z	Vertical position within bed	independent variable m	
ε	Void fraction of the bed	0.35	*(22)*
μ	Specific growth rate	s^{-1} (Equation /2/)	
$\mu_{(T=opt)}$	Specific growth rate at the optimal temperature for growth	0.3242 h^{-1}	*(16)*
μ_W	Viscosity of water	0.001 Pa s^{-1}	*(19)*
λ	Enthalpy of vaporization of water $\frac{E}{m(\text{water})}$	2414300 J kg^{-1}	*(17)*
ρ_a	Density of air	1.14 kg m^{-3}	*(19)*
ρ_b	Bed density	kg m^{-3} (Equation /4a/)	
ρ_s	Density of the substrate particles	700 kg m^{-3}	*(16)*
ρ_W	Density of water	1000 kg m^{-3}	*(19)*
ΔP	Pressure drop of cooling water across the heat transfer plate	Pa (Equation /22/)	
$\Delta T_{driving}$	Driving force for heat transfer	10 °C	
ΔT_{water}	Allowable temperature rise as the cooling water flows through the heat transfer plates	1 °C	

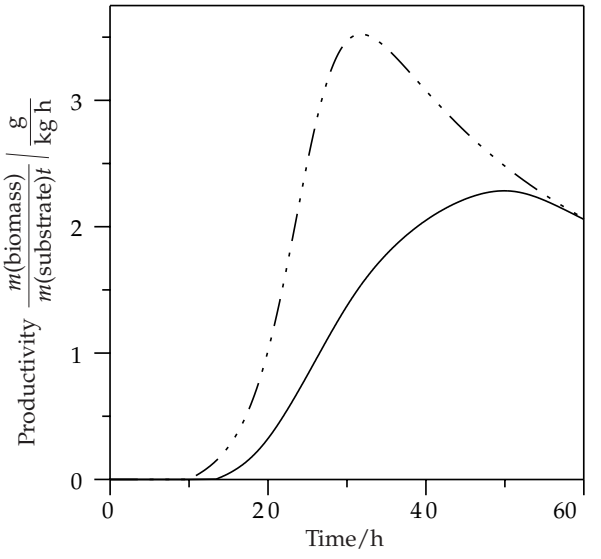

Fig. 2. Productivity (P_S) profiles for a fermentation simulated using the parameter values in Table 2 (——) and for logistic growth with μ = 0.324 h^{-1} (- ··· -). A 10 h nonproductive time has been added at the beginning of the fermentation.

time at which the volume-averaged biomass concentration reaches 90 % of X_m is therefore selected as the harvest time. The symbol t_{90} is then defined as the time between the start of the active growth phase and the time at which $0.9X_m$ is reached. For logistic growth with μ = $\mu_{(T=opt)}$ = 0.324 h^{-1}, combined with a nonproductive time of 10 h, the maximum possible value of P_{SM} (biomass per substrate per hour) is 3.5 $g \cdot kg^{-1} \cdot h^{-1}$ at 31.8 h, and the value of t_{90} is 21.6 h.

The fraction of X_m that corresponds to the maximum overall productivity depends on the various parameters within Equation /10/. In Table 3 the value of $\mu_{(T=opt)}$ was varied from 0.081 h^{-1} to 0.648 h^{-1}, X_m was varied from 0.031 to 0.187 kg-biomass · kg-substrate^{-1} and t_N was varied from 0 to 30 h, which span the range of values that can be expected in SSF processes. The absolute value of the maximum productivity (P_{SM}) and the time at which it is reached vary widely as the parameters $\mu_{(T=opt)}$, X_m and t_N are varied. In contrast, the variation in the fraction of X_m that corresponds to maximum overall productivity is less pronounced, ranging from $0.849X_m$ (obtained with X_m = 0.125, $\mu_{(T=opt)}$ = 0.324 and t_N = 0) to $0.943X_m$ (obtained with X_m = 0.125, $\mu_{(T=opt)}$ = 0.324 and t_N = 30). The time at which $0.9X_m$ is reached is therefore a reasonable approximation of the best harvest time, over a wide range of parameter values.

Volumetric productivity of the bioreactor

Equation /11/ can be used to calculate the volumetric productivity of the bioreactor, substituting $0.9X_m$ for X and t_{90} for t. However, for a Zymotis type bioreactor, in which the half-plate spacing is an important parameter, use of this volumetric productivity as a criterion to find the optimal plate half-spacing leads to an optimal

Table 3. Effect of different values of the specific growth rate constant at the optimum temperature ($\mu_{(T=opt)}$), the maximum biomass concentration (X_m), and the non-productive time (t_N) on the maximum value of P_{SB} (P_{SBmax}), the time at which this maximum occurs and the fractional biomass concentration (X/X_m) at this time

Varied parameters			Calculated results		
$\mu_{(T=opt)}$ / h^{-1}	X_m / kg-biomass kg-initial-substrate^{-1}	t_N / (h)	P_{SBmax} / kg-biomass kg-initial-substrate^{-1} h^{-1}	Time at which P_{SBmax} attained* / h	X/X_m when P_{SBmax} attained
Base case					
0.3242	0.125	10	0.00352	21.8	0.904
Effect of $\mu_{(T=opt)}$					
0.0811	0.125	10	0.00116	82.7	0.868
0.1621	0.125	10	0.00210	42.2	0.883
0.6484	0.125	10	0.00538	11.4	0.929
Effect of X_m					
0.3242	0.031	10	0.00098	16.9	0.889
0.3242	0.062	10	0.00186	19.3	0.895
0.3242	0.187	10	0.00508	23.2	0.908
Effect of t_N					
0.3242	0.125	0	0.00520	20.2	0.849
0.3242	0.125	20	0.00269	23.0	0.933
0.3242	0.125	30	0.00218	23.5	0.943

*measured from the commencement of growth and not the time of inoculation, and therefore does not include the nonproductive time

spacing of zero. This occurs because, with a plate half-spacing of zero, and with cooling water at T_{opt} within the plates, then the whole bed would be maintained at T_{opt}, giving optimal growth. However, the reactor would then be full of heat transfer plates and have no actual substrate, meaning that although the productivity per volume of bed would be maximal, the productivity of the reactor itself would be zero. Using the values in Table 2 for the bioreactor, microorganism and substrate parameters gives:

$$P_B = \frac{50.7325}{\left(1+\frac{W}{2L}\right)(1+0.1)(t_{90}+t_N)} \qquad /14/$$

The optimum value of L cannot be obtained by simple differentiation of Equation /14/ because t_{90} for a fermentation run depends on L in a complex way that cannot be expressed as a simple equation, due to the effects of L on heat transfer and therefore the temporal temperature profile experienced by the microorganism. Note that t_{90} will also depend on other variables such as the superficial air velocity used *(15)*. In the current work the value of t_{90} is obtained by solving the heat transfer model of Mitchell and von Meien *(15)*, which has the plate half-spacing (L) as one of the parameters, although of course in the absence of a model it could be obtained graphically from the fermentation data. However, even though t_{90} values can be obtained from the model, it is still not possible to solve for the overall bioreactor productivity, because a value is needed for W. Selection of an appropriate value for W is addressed in the next section.

Appropriate values for the cooling plate thickness

The appropriate value for the cooling plate thickness (W) depends on considerations of pressure drop. The inner gap in the plate (W_{GAP}) must be sufficiently large to give an acceptably small pressure drop when water is flowing through the plate at a rate sufficient to remove the waste metabolic heat at the time of peak heat generation. It also depends on the thickness of the plate wall (W_{WALL}), which is a strength consideration. Table 4 shows the calculation of the pressure drop on the basis of the water flow rate required to limit the temperature rise of the cooling water between the entrance and exit of the cooling plate to 1 °C, at the time of maximum metabolic heat production. The value of 2 in Equation /16/ arises because each heat transfer plate must remove the heat from two half-slabs. The required heat transfer coefficient calculated using Equation /17/ is within the range of typical heat transfer coefficients for liquids on both sides of the plate *(19)* and therefore the plate can probably remove the heat at the required rate. Note that Perry *et al. (19)* do not list values directly applicable to the situation with a solid on one side and a liquid on the other.

The pressure drop across the plate depends on the internal gap between the plate walls, W_{GAP}. An overall plate width of 10 mm was chosen, being two 2 walls each of 3 mm width and a gap of 4 mm between the plates (*i.e* $W = 0.01$ m $= 2W_{WALL} + W_{GAP}$). Such a plate should have sufficient mechanical strength to be used in large scale bioreactors, since 3 mm walls are typical of other large-scale applications *(19)*. The calculated Reynolds number of 1275 belongs to the transient flow regime and consequently the pressure drop was calculated with the correlation for turbulent flow regime (Equation /21/). The pressure drop of 213 Pa is quite acceptable, meaning that a value of W_{GAP} of 4 mm is suitable.

Determination of the optimal plate half-spacing

With a value for W, it is possible to calculate the optimal plate half-spacing. The model was used to simu-

late fermentations at a range of values of L, with a control factor of 2 in the water temperature control algorithm. The value of t_{90} was obtained by inspection of the results and substituted into Equation /14/, using the value for W of 0.01 m that was calculated in the previous section. There is a broad peak for the bioreactor productivity (P_B), spanning from L = 0.015 to 0.04 cm, with the actual optimum occurring from 0.02 to 0.03 m (Fig. 3b). From handling considerations, and to minimize the expense of heat transfer plates, the gap should be made as large as possible and therefore a plate half-spacing of 0.03 m might be chosen (*i.e.* a 6 cm gap between plates).

The usefulness of P_B, the productivity based on the overall volume, as the criterion for evaluating bioreactor performance is clear. In contrast to P_V, P_B gives an optimum L which is not equal to zero. Fig. 3 shows the various influences that combine to give this optimum. Firstly, the better temperature control as the plate half-spacing is decreased leads to better values of t_{90} (Fig. 3a), and therefore increasingly greater values of P_V (Fig. 3b). However, the fraction of the bioreactor occupied by the bed (V_F) falls (Fig. 3a), with the fall becoming more rapid as the plate half-spacing decreases below 0.02 m. These opposing curves for P_V and V_F combine to give the optimum. Fig. 3b also shows the maximum possible value for P_B, that is, the value which would be obtained if the growth occurred at the $\mu_{(T=opt)}$ of 0.324 h^{-1} throughout the growth phase. This increases monotonically as the plate half-spacing increases because it does not take into account the difficulties in temperature control for wider slabs. Note that the overall bioreactor productivity predicted by the model is close to this maximum possible value for low values of the plate half-spacing, but, due to the difficulties in maintaining the bed temperature near the optimum at higher plate half-spacings, the predicted performance deviates greatly from the maximum possible value.

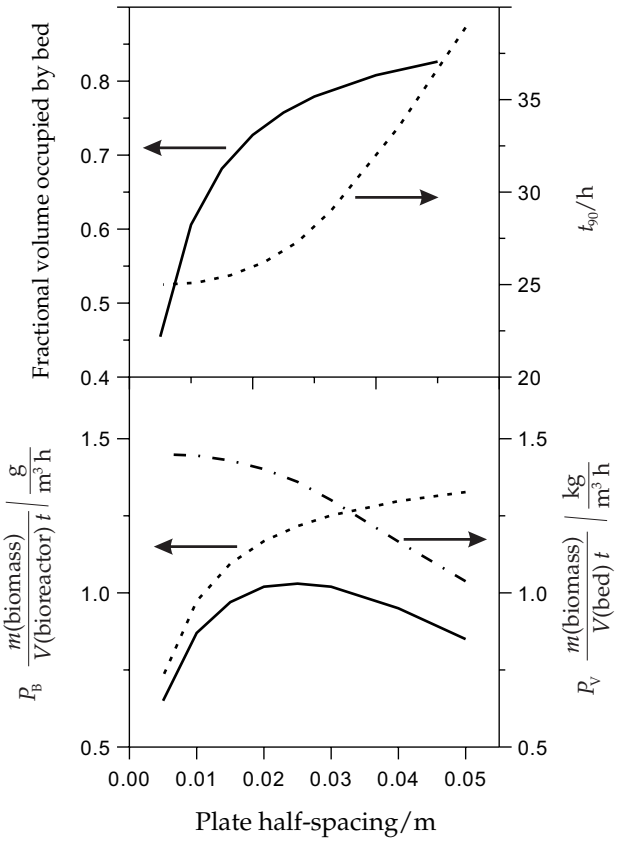

Fig. 3. Predicted performance of the Zymotis bioreactor under the conditions given in Table 2 with different plate half-spacings. (a) Fractional volume occupied by the bed (——) and the time taken to reach 90 % of the maximum biomass concentration, t_{90} (– – –). (b) Productivities based on the overall bioreactor volume (P_B) (——) and the volume of the bed itself (P_V) (– · – · –). The maxium possible value of P_B is also shown (– – –). It represents the value of P_B that would be obtained if it were possible to maintain the bed at the optimum temperature for growth throughout the entire fermentation.

Table 4. Steps in the calculation of the pressure drop of the cooling water across the heat transfer plate

Step in the calculation	Equation used	Eq.No.	Value calculated*
Maximum heat production rate for logistic growth kinetics *(8,24)*	$q_{peak} = 0.25\rho_S(1\text{-}\varepsilon)Y\mu_{(T=opt)}X_m$	/15/	10712 W m^{-3}
Maximum required rate of heat removal per heat transfer plate	$Q_{duty} = 2L.H.D.\ q_{peak}$	/16/	2680 W
Overall coefficient required for convective cooling at the heat transfer plate	$h = \dfrac{Q_{duty}}{H_p D \Delta T_{driving}}$	/17/	107.2 W m^{-2} °C^{-1}
Mass flow rate of cooling water required to give the desired cooling rate	$G = \dfrac{Q_{duty}}{C_{PW}\Delta T_{water}}$	/18/	0.6405 kg s^{-1}
Superficial velocity required for water flowing through the heat transfer plate	$v = \dfrac{G}{DW_{GAP}\rho_W}$	/19/	0.160 m s^{-1}
Characteristic length of the flow area for calculation of the Reynold's number	$L_c = \dfrac{W_{GAP}D}{2(W_{GAP} + D)}$	/20/	0.00199 m
Reynold's number	$\mathrm{Re} = \dfrac{4L_c\rho_W v}{\mu_W}$	/21/	1275
Pressure drop	$\Delta P = \dfrac{1}{2}\rho_W v^2 \dfrac{H_p}{L_c}\left(\dfrac{0.0791}{\mathrm{Re}^{0.25}}\right)$	/22/	213 Pa

*calculated using the values given in Table 2

General applicability of the productivity analysis to SSF

Although the analysis of productivity in the current work has been done for one specific bioreactor, and on the basis of assumed logistic growth kinetics, the general approach is obviously easily extendable to other bioreactors and other growth kinetics. As demonstrated above, the maximum value of P_V (productivity in kg of biomass per m^3 of bed) is easily determined. The appropriate growth kinetic equation can be substituted into Equation /9/, which allows P_V to be plotted directly or the graphical method can be applied directly to the experimental biomass profile, taking care to use a volume-weighted biomass concentration if the bioreactor is not well-mixed. To convert the P_V to an overall bioreactor productivity, it is simply necessary to know the ratio between productive and non-productive volume in the bioreactor:

$$P_B = V_F \cdot P_V = \frac{1}{\left(1+\frac{V_N}{V_B}\right)} \cdot \frac{X_{harvest}\rho_b}{t_{cycle}} \qquad /23/$$

where V_N is the non-productive volume within the bioreactor. This productivity then gives a useful criterion for comparing the performance of different bioreactors, or, as shown in the present paper, the same bioreactor under different design and operating conditions.

Conclusions

In the current work we have developed a criterion for comparing bioreactor performance, namely the overall bioreactor productivity, which takes into account the whole bioreactor volume and not just the bed volume. The applicability of this productivity calculation is not limited to the Zymotis packed-bed. It can be used to compare the performance of the range of different SSF bioreactors. This productivity calculation is a useful tool for use as an objective function to be maximized in a program attempting to optimize bioreactor operation.

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Procjena produktivnosti bioreaktora Zymotis sa čvrstim slojem zasnovana na ukupnom volumenu reaktora

Sažetak

Opisana je metoda analize rada fermentacijskog bioreaktora sa čvrstim slojem. Postupak je primijenjen da bi se ispitala optimalna vrijednost za razmak između ploča za hlađenje u bioreaktoru Zymotis, koristeći simulirane fermentacijske podatke dobivene prema matematičkom modelu. Bioreaktor Zymotis je vrlo prikladan za one fermentacijske procese u čvrstom sloju u kojima supstratni sloj mora ostati statičan. Opisani postupak koristi dva određena parametra uvjetovana prisutnošću internih ploča za prijenos topline. Širina ploča za prijenos topline uvjetovana je količinom topline koju treba ukloniti i padom tlaka rashladne vode te razmakom između tih ploča. Da bi se analizirao učinak bioreaktora, uveden je pojam produktivnosti koji uzima u obzir volumen unutar bioreaktora što ga zauzimaju ploče za prijenos topline. Kao dio ove analize pokazalo se da je, prema logistici kinetike rasta, optimalno vrijeme za povećavanje produktivnosti upravo ono kada biomasa postiže 90 % svoje maksimalne moguće vrijednosti.