T H E

ACETONE BUTANOL ETHANOL

FERMENTATION:

PRELIMINARY STUDIES ON SOME PRACTICAL ASPECTS

by

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ABSTRACT

The dilute nature of solvents at the end of fermentation and slow overall rate of fermentation are major economic burdens on a commercial plant producing acetone, butanol, and ethanol. A preliminary feasibility costing of such a plant showed the cost of fermenters represents almost 50% of the total purchased equipment cost, and emphasised the need for improvements in the fermentation.

Experiments were performed in 10-litre and 30-litre pressure vessels, a 1.5-litre vessel at atmospheric pressure, and trial runs in 100 ml bottles.

Good correlations were found for the different fermentation headspace pressures (100 to 250 kPa abs.) and minimum observed pH's (pH 4.2 to pH 4.65) with final butanol yields (0.92 to 11.6 g/l); increases in both parameters correlating with increased butanol concentration. Ethanol was found to be correlated with pressure only, and acetone with neither parameter directly. Other chemical species present in the broth were also correlated with each other. It was found that a tree diagram drawn using the strongest correlations resembled closely the known metabolism of the organism in terms of the metabolic pathways, specification of active forms of the metabolites, and effect of external influences. Use of multiple linear regression in this manner was named The Factor Correlation Method, and is potentially useful for research on metabolism and similar investigation on a much broader basis. Application of this technique showed that the pressure effect was possibly due to more than a single metabolic cause, and further experiments also emphasised the complex nature of the pressure effect.

The experimental work also highlighted the potential hazard of culture degeneration leading to substandard fermentation yields and eventual nonviability. Discussion on the experimental results and of the literature suggests the phenomenon is due to infection by lysogenic phage rather than spontaneous mutation, and an approximate model based on simultaneous partial differential equations parallels some observed characteristics of the phenomenon.

Other topics include theoretical exercises with laboratory work on the water tolerance of methanol-petrol mixtures, the error associated with cell enumeration using a haemocytometer, and evaluation of growth and solvent production characteristics and some relevant parameters.

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CONTENTS

LIST OF VARIABLES AND ABBREVIATIONS

Variable	Name	Units
a	a constant	\$
a_a	specific area of packing	$m^2\ m^{-3}$
acet	acetone	
A	area	m^2
A_a	active area on the plate	m^2
$[AA]_f$	final total acetate concentration	(g/l)
$[AA^-]_f$	final, calculated, dissociated acetic acid concentration	(g/l)
A.B.E.	acetone butanol ethanol fermentation	-
$[A]_f$	final acetone concentration	(g/l)
A_s	area free for gas flow	m^2
b	a constant	$\$m^3$
b.p.	boiling point	°C
but	butanol	
B	bottoms flow	$mol\ s^{-1}$
$[BA]_f$	final total butyrate concentration	(g/l)
$[BA^-]_f$	final, calculated, dissociated butyric acid concentration	(g/l)
$[B]_f$	final n-butanol concentration	(g/l)
BOD	Biological Oxygen Demand	ppm
c	specific heat	$Jg^{-1}\ K^{-1}$
c′	total cost of fermenters	\$
CM	Cooked Meat Medium	
CoA	Coenzyme A	-
COD	Chemical Oxygen Demand	ppm
CW	cooling water	-
d.f.	degrees of freedom	-
d_g	gas density	$kg\ m^{-3}$
d_l	liquid density	$kg\ m^{-3}$

D	diameter	m
D′	distillate flow	$mol\ s^{-1}$
DC	direct cost	$
DE	time taken between viral infection and cell burst	hr
Dist	distillation section	-
DPC	direct product cost	$pa
DR	rate of death of active infected cells	t_d^{-1}
e	error	
e_a	human absolute error	cells
equ.	equation	
eth	ethanol	
E	number of cells per square expected	cells
$[E]_f$	final ethanol concentration	(g/l)
f	fractional error	-
f_a^2	human fractional error	-
frac.	fraction	
F_g	fraction of cross sectional area open to gas flow	%
FC	fixed charges	$pa
FCI	fixed capital investment	$
Fd	ferridoxin	
Ferm	fermentation section	-
FFAP	Free Fatty Acid Phase	
FP	feed plate position (numbered from the top of the column).	-
GE	general expenses	$pa
GLC	gas-liquid chromatography	
Glu	d-glucose	
h	liquid hold up as a fraction of the total bed volume	-
h_c	heat of combustion	Jg^{-1}
h_v	heat of vapourisation	Jg^{-1}

$[H^+]_f$	final hydrogen ion concentration	(M)
$[H^+]_m$	minimum hydrogen ion concentration observed	(M)
$[HAA]_f$	final, calculated, undissociated acetic acid concentration	(g/l)
$[HBA]_f$	final, calculated, undissociated butyric acid concentration	(g/l)
HTU	height of a transfer unit	m
H_2O	water	
i	inhibition power constant	-
I	inhibitor concentration	g/l
IC	indirect cost	$
IPC	individual purchase cost of item updated to September 1980	$
k	growth rate estimated from the logistic equation	hr^{-1}
K´	a constant	
K_m	a constant	
K_v	a constant	-
L	length	m
LMTD	log mean temperature difference	K
LN	log mean cell concentration	10^6/ml
m	mass flow rate	kg hr^{-1}
m_f	slope of feed line for a McCabe-Thiele Construction	
mol	mole	
m_r	slope of rectification section operating line	-
MC	manufacturing costs	$pa
MEK	methyl ethyl ketone	
M_i	general term for a property	-
MLR	Multiple Linear Regression	

M_m	general term for a property of a mixture	-
MON	motor octane number	-
Mr	molecular weight	g/mol
MU	growth rate	hr^{-1}
M15	mixture of 15% (v/v) methanol with 85% (v/v) petrol	-
M85	mixture of 85% (v/v) methanol with 15% (v/v) petrol	-
n	number of squares counted	squares
no.	number of operating fermenters (excluding spare).	-
N	cell concentration	10^6/ml
NT	number of trays	-
N.C.P.	National Chemical Products Ltd	-
N_{log}	cell population estimated by the logistic equation	10^6/ml
N_o	cell concentration immediately after inoculation	10^6/ml
NPN	non-proteinaceous nitrogen	
N_t	average stationary cell population	(X 10^6 cells/ml)
N_s	stationary phase cell population	10^6/ml
N_{sm}	cell population smoothed using three point moving averages	10^6/hr
NTU	number of transfer units	-
NV	burst size	$cell^{-1}$
O	observed cell population	10^6/ml
OD	oven dried	
p	pressure	kPa
pa	per annum	
pH_f	final broth pH	

pH_m	minimum observed pH	
PEC	purchased equipment cost	$
P_l	wetted column pressure drop	$Pa\ m^{-1}$
PLB	rate of conversion of lysogenic to virial cell	t_d^{-1}
P_o	dry column pressure drop	$Pa\ m^{-1}$
Prod	product section	-
PS	success constant	
P_{sm}	product concentration smoothed using three point moving averages	g/l
q	specific rate of production of product	μg/l hr cell
q′	ratio of heat required to vapour in the feed to a distillation column to its heat of vapourisation	-
Q	average WP flow rate ($15\ m^3\ hr^{-1}$)	$m^3\ hr^{-1}$
Q_l	liquid flow rate	$mol\ s^{-1}$
Q_g	gas flow rate	$mol\ s^{-1}$
r	ratio of fermenter filling rate to Q.	-
rpm	revs per minute	$rev.\ min^{-1}$
R	ratio of fermenter emptying rate to Q.	-
R_m	minimum reflux ratio	-
RON	research octane number	-
R_r	external reflux ratio	-
R′	maximum rate of butanol production	$g\ l^{-1}hr^{-1}$
RR	$RR = 0.8\ (r^{-1} + R'^{-1})$	-
s	absolute standard error	cells
s & t	shell and tube heat exchanger	-
s.g.	specific gravity	-
std.	standard	-

s^2	variance of cell counts	$cells^2$
S	substrate concentration	g/l
t	time	s
t_d	doubling time for a healthy cell	hr
t_{sm}	time smoothed using three point moving averages	hr
t_t	turnaround time	hr
T	temperature	K
TCI	total capital investment	$
TLV	threshold limit value	ppm
TN	total nitrogen	
U	overall heat transfer coefficient	$kW\ m^{-1}\ K^{-1}$
v	volume of single fermenter	m^3
v′	velocity	$m\ s^{-1}$
V	total fermentation volume	m^3
V_f	flooding velocity	$m\ s^{-1}$
V_o	optimum velocity	$m\ s^{-1}$
V_{tt}	Total tank volume	m^3
V'_t	superficial velocity without downcomers	$m\ s^{-1}$
w	heat flux	W
WC	working capital	$
WP	Sulphuric acid casein whey permeate	-
WSH	Wood Sugar Hydrolysate	
WSR	Wood Sugar Hydrolysate Residue	
$\bar{x}$	arithmetic mean	-
x_i	composition	mol. frac.
y	composition (operating line)	mol. frac.
y*	equilibrium composition	mol. frac.
y_d	distillate composition	mol. frac.
Y	conc. of healthy cells	10^6/ml
YALL	YALL = Y + YY + YL	10^6/ml
YE	yeast extract powder	-
YL	conc. of lysogenic cells	10^6/ml

y_o	bottoms composition	mol. frac.
YY	conc. of infected cells	10^6/ml
YYV	conc. of infected cells a period D before the time of interest	10^6/ml
$Y_{.68}$	68% confidence level envelope	cells
ΔG_o	standard change in free energy	J/mol
$\Delta G'$	change in free energy	J/mol
α	a constant	-
β	a constant	-
μ	growth rate	hr^{-1}
$\int$	integral sign	
¢	voidage	-
Ø	a constant	-

LIST OF FIGURES

LIST OF TABLES