Appendix A

The kinetic model used in this work was described by the next equations, r (mmol/h):

$$r_1=\frac{V_{max1}[\text{Glucosa}][\text{Biomasa}]}{K_{m1}\left(1+\frac{[\text{Glucosa}]}{K_{m1A}}\right)+[\text{Glucosa}](1+[\text{Butanol}])}.F \quad \text{(A.1)}$$

$$r_2=\frac{V_{max2}[\text{FGP}][\text{Biomasa}]}{K_{m2}+[\text{FGP}]}.F \quad \text{(A.2)}$$

$$r_3=\frac{V_{max3}[\text{G3P}][\text{Biomasa}]}{K_{m3}+[\text{G3P}]}.F \quad \text{(A.3)}$$

$$r_4=\frac{V_{max4}[\text{Lactato}][\text{Biomasa}]}{K_{m4}+[\text{Lactato}]}.F \quad \text{(A.4)}$$

$$r_5=\frac{V_{max5}[\text{Piruvato}][\text{Biomasa}]}{K_{m5}+[\text{Piruvato}]}.F \quad \text{(A.5)}$$

$$r_6=\frac{V_{max6}[\text{Piruvato}][\text{Biomasa}]}{K_{m6}+[\text{Piruvato}]}.F \quad \text{(A.6)}$$

$$r_7=\frac{V_{max7}[\text{Actetato}][\text{Biomasa}]}{K_{m6}+[\text{Acetato}]}.F \quad \text{(A.7)}$$

$$r_8=V_{max8}\left(\frac{1}{1+[K_{m8A}/\text{Acetato}]}\right)\left(\frac{1}{1+[K_{m8B}/\text{Acetato}]}\right)[\text{Biomasa}] \quad \text{(A.8)}$$

$$r_9=\frac{V_{max11}[\text{ACoA}][\text{Biomasa}]}{K_{m11}+[\text{ACoA}]}.F \quad \text{(A.9)}$$

$$r_{10}=\frac{V_{max11}[\text{ACoA}][\text{Biomasa}]}{K_{m11}+[\text{ACoA}]}.F \quad \text{(A.10)}$$

$$r_{11}=\frac{V_{max11}[\text{ACoA}][\text{Biomasa}]}{K_{m11}+[\text{ACoA}]}.F \quad \text{(A.11)}$$

$$r_{12}=\left(\frac{V_{max12}[\text{Glucose}][\text{Biomasa}]}{\text{Glucosa}+K_{m12}+K_{m12A}/[ACoA]}\right).INH \quad \text{(A.12)}$$

$$r_{13}=k_{13}[Biomasa] \quad \text{(A.13)}$$

$$r_{14}=\frac{V_{max18}[AACoA][Biomasa]}{K_{m18}+[AACoA]}.F \quad \text{(A.14)}$$

$$r_{15}=V_{max15}\left(\frac{1}{1+(K_{m15A}/[Butirato])}\right)\left(\frac{1}{1+(K_{m15B}/[AACoA])}\right)[Biomasa] \quad \text{(A.15)}$$

$$r_{16}=r_8+r_{15} \quad \text{(A.16)}$$

$$r_{17}=\frac{V_{\max 17}[Glucosa][Biomasa]}{K_{m17}+[Butirato]+K_{m17A}/[Butirato]+K_{m17B}/[Butanol]}\cdot F \quad \text{(A.17)}$$

$$r_{18}=\frac{V_{\max 18}[BCoA][Biomasa]}{K_{m18}+[BCoA]} \quad \text{(A.18)}$$

$$r_{19}=\frac{V_{\max 19}[BCoA][Biomasa]}{K_{m19}\left(1+K_{m19A}/[Butirato]\right)+[BCoA]\left(1+[Butanol]/K_{m19B}\right)}\cdot F \quad \text{(A.19)}$$

$$r_{20}=\frac{V_{\max 20}[Xilosa][Biomasa]}{K_{m20}\left(1+[Xilosa]/K_{m20A}\right)+[Xilosa]\left(1+[Butanol]\right)}\cdot F \quad \text{(A.20)}$$

$$r_{21}=\frac{V_{\max 21}[X5P][Biomasa]}{K_{m21}+[X5P]}\cdot F \quad \text{(A.21)}$$

$$r_{22}=\frac{V_{\max 22}[X5P][Biomasa]}{K_{m22}+[X5P]}\cdot F \quad \text{(A.22)}$$

$$r_{23}=\left(\frac{V_{\max 23}[Xilosa][Biomasa]}{[Xilosa]+K_{m23}+K_{m23A}/[ACoA]}\right)\cdot INH \quad \text{(A.23)}$$

$$INH=\left(1-\frac{Butanol}{Butanol_{\max}}\right)\cdot\left(1-\frac{Acetato}{Acetato_{\max}}\right)\cdot\left(1-\frac{Butirato}{Butirato_{\max}}\right)\cdot\left(1-\frac{Biototal}{Biototal_{\max}}\right) \quad \text{(A.24)}$$

Where F is equal to 1 if substrate concentration is bigger than 1 (mM) on the contrary case is equal to cero, this is the reason why this model is called 'on-off'. Please note that the Global reactions are without hydrolysis.

To solve the entire model we also considered the following differential equations to describe the rate of all involved components during fermentation.

$$\frac{d[Glucosa]}{dt}=-r_1 \quad \text{(A.25)}$$

$$\frac{d[F6P]}{dt}=r_1-r_2+r_{21} \quad \text{(A.26)}$$

$$\frac{d[Piruvato]}{dt}=r_3+r_4+r_5-r_6 \quad \text{(A.27)}$$

$$\frac{d[Lactato]}{dt}=r_5-r_4 \quad \text{(A.28)}$$

$$\frac{d[ACoA]}{dt}=r_6+r_7+r_8-r_9-r_{10}-r_{11}-r_{12}-r_{23} \quad \text{(A.29)}$$

$$\frac{d[Biomasa]}{dt}=r_{12}+r_{23}-r_{13} \quad \text{(A.30)}$$

$$\frac{d[\text{Acetato}]}{dt}=r_9-r_7-r_8 \quad (A.31)$$

$$\frac{d[\text{Etanol}]}{dt}=r_{11} \quad (A.32)$$

$$\frac{d[\text{BCoA}]}{dt}=r_{14}+r_{15}+r_{17}-r_{18}-r_{19} \quad (A.33)$$

$$\frac{d[\text{Butirato}]}{dt}=r_{18}-r_{15}-r_{17} \quad (A.34)$$

$$\frac{d[\text{Acetona}]}{dt}=r_{16} \quad (A.35)$$

$$\frac{d[\text{Butanol}]}{dt}=r_{19} \quad (A.36)$$

$$\frac{d[CO_2]}{dt}=r_{16}+r_6 \quad (A.37)$$

$$\frac{d[\text{Xilosa}]}{dt}=-r_{20} \quad (A.38)$$

$$\frac{d[\text{X5P}]}{dt}=-r_{22}-r_{21}+r_{20} \quad (A.39)$$

Table A.1. List of fermentation model constants

Reaction	V_{max}	K_m	K_{ma}	K_{mb}
r_1	1.A0	5.45	A3.11	3A.4A
r_2	21.8A	1.54		

r_3	42.15	4.0A		
r_4	2.15	173.16		
r_5	7.47	131.35		
r_6	67.64	3.12		
r_7	1A.50	22.21		
r_8	8.08	4.5A	45.04	
r_A	34.84	0.71		
r_{10}	33.A8	10.AA		
r_{11}	4.60	7.40		
r_{12}	0.53	18.25	6.37	
r_{13}	0.01A			
r_{14}	A2.47	16.46		
r_{15}	0.23	2.20	0.00105	
r_{16}	11.A6	0.48	24.74	175.2A
r_{17}	31.10	1.33		
r_{18}	A2.81	1.30	86.04	148.15
r_{1A}	1.55	3.33	51.80	A6.20
r_{20}	AA.47	4.66		
r_{21}	131.35	25.78		
r_{22}	0.15	0.06	0.11	
r_{23}	1.A0	5.45	A3.11	3A.4A

For hydrolysis, Kadam et al. (2004) utilized a Langmuir-type isotherm to describe the enzymatic hydrolysis of cellulose; in this model, the inhibitory effect on celluloses and other sugars present in the biomass hydrolyzated was considered as well in the effect of temperature

$$E_{iB} = \frac{E_{i\max} K_{iad} E_{iF} S}{1 + K_{iad} E_{iF}} \tag{A.40}$$

$$r_1 = \frac{k_{1r} E_{1B} R_S S}{1 + \frac{G_2}{K_{1IG2}} + \frac{G}{K_{1IG}} + \frac{X}{K_{1IX}}} \tag{A.41}$$

$$r_2 = \frac{k_{2r} (E_{1B} + E_{2B}) R_S S}{1 + \frac{G_2}{K_{2IG2}} + \frac{G}{K_{2IG}} + \frac{X}{K_{2IX}}} \tag{A.42}$$

$$r_3 = \frac{k_{3r} E_{2F} G_2}{K_{3M} \left(1 + \frac{G}{K_{3IG}} + \frac{X}{K_{3IX}} \right) + G_2} \tag{A.43}$$

$$\frac{dS}{dt} = -r_1 - r_2 \tag{A.44}$$

$$\frac{dG_2}{dt} = 1.056 r_2 - r_3 \tag{A.45}$$

$$\frac{dG}{dt} = 1.111 r_2 + 1.053 r_3 \quad \text{(A.46)}$$

$$E_{Ti} = E_{Fi} + E_{Bi} \quad \text{(A.47)}$$

$$k_{ir(T2)} = k_{ir(T1)} e^{-E_{ai}/R\{1/T1 - 1/T2\}} \quad \text{(A.48)}$$

$$R_s = \frac{S}{S_0} \quad \text{(A.49)}$$

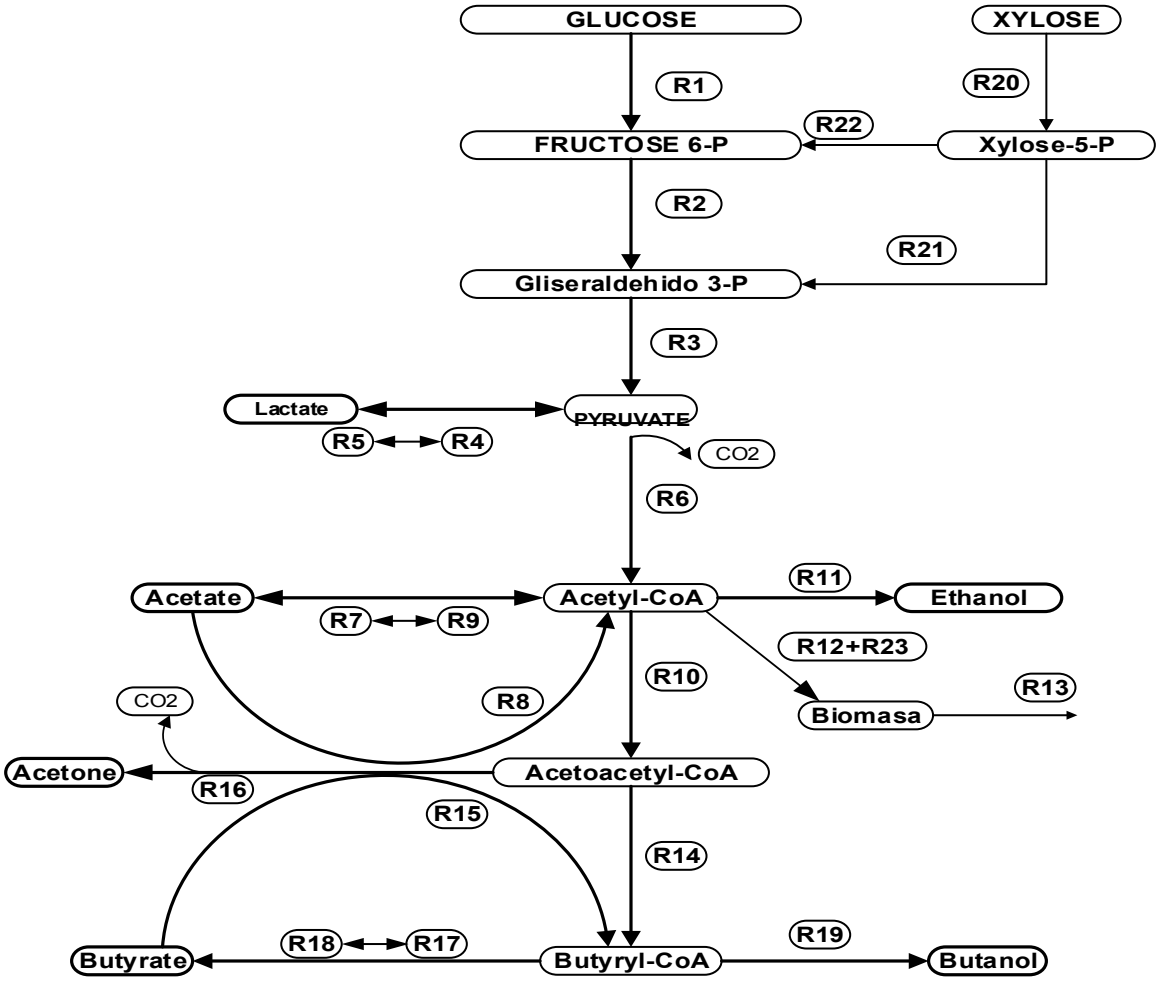

Figure A-1. Metabolic pathway for xylose and glucose consumption

Shinto, H.; Tashiro, Y.; Kobayashi, G.; Sekiguchi, T.; Hanai, T.; Kuriya, Y.; Okamoto, M.; Sonomoto, K. Kinetic Study of Substrate Dependency for Higher Butanol Production in Acetone-Butanol-Ethanol Fermentation. *Process Biochem.*

Kadam, K. L.; Rydholm, E. C.; McMillan, J. D. Development and Validation of a Kinetic Model for Enzymatic Saccharification of Lignocellulosic Biomass. *Biotechnol. Prog.* **2004**, *20* (3), 698.