PRODUCTION OF MALTOSE BY USING ENZYMATIC STARCH HYDROLYSIS

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INTRODUCTION

Maltose :

- **Malt sugar**
- **Disaccharide**
- **Consist of 2 polymeric carbohydrate**
  i. **Amylose**
  ii. **Amylopectin**

Maltose syrups applications :

i. Food
ii. Pharmaceutical
iii. Fine chemical industries
- bulking agents
- crystallization inhibitors
- moisture conditioners
- stabilizers

(Saha BC, Zeikus JG ;Biotechnol Bioeng 1989;34:299 –303)

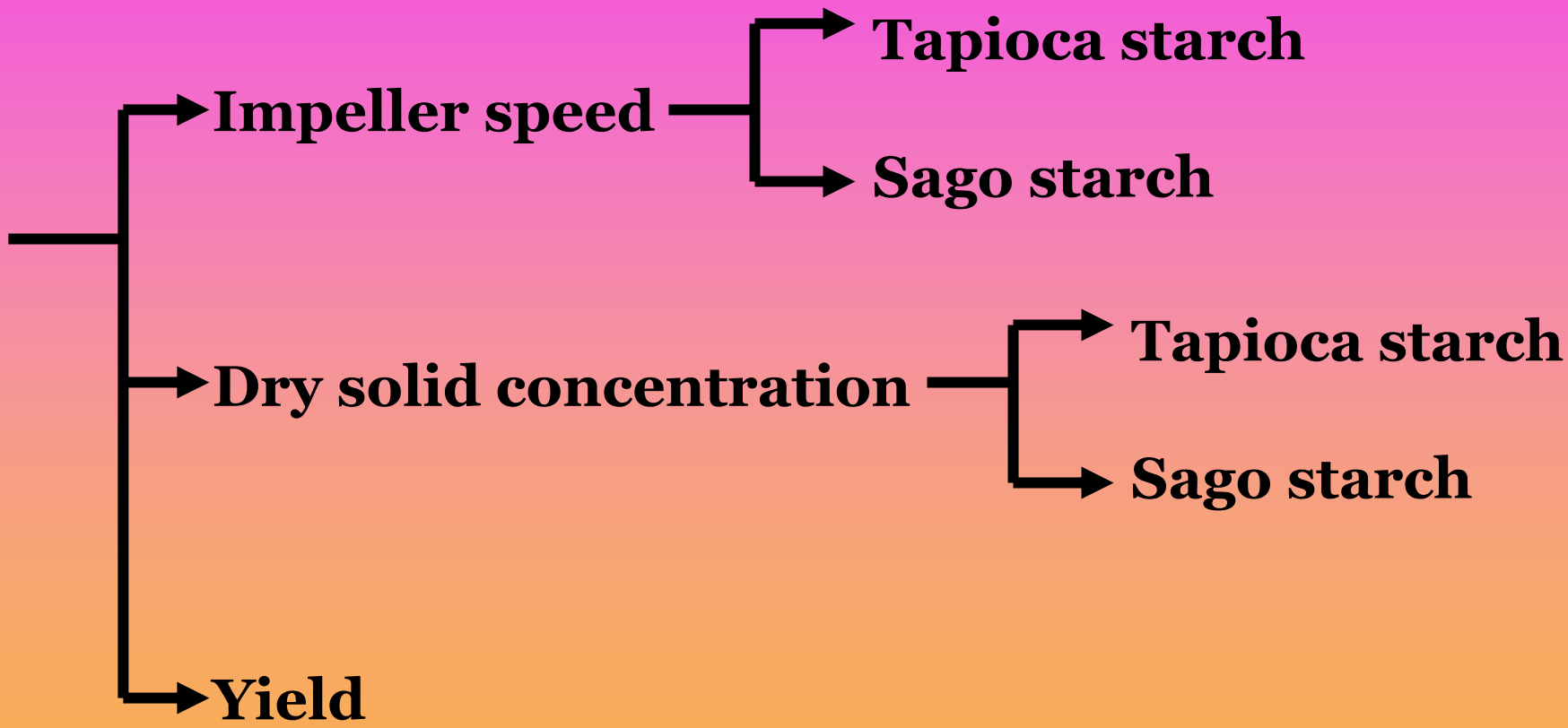
PROBLEM STATEMENT

1. **Most researcher used 2 types of enzyme in low concentration of dry solid (< 5.0 % w/v)**

   (C.W. Wong, S.K.S. Muhammad, M.H. Dzulkifly, N. Saari, H.M. Ghazali ; Food chem, 2005)

2. **Only several researcher investigated on the effect of impeller speed on the production of maltose.**

OBJECTIVES


Impeller speed
Tapioca starch
Sago starch
Dry solid concentration
Tapioca starch
Sago starch
Yield

- ✓ **2L fermenter**
- ✓ **Fungal alpha amylase (from *Aspergillus Oryzae*)**
- ✓ **Different impeller speed at 100, 200 and 300 rpm**
- ✓ **Different dry solid concentration at 8, 10 and 12% w/v**

METHODOLOGY

Mixing :

1L of 8, **10** and 12% Tapioca starch / Sago starch
(G.S Ayernor, T.K Hammond, A.Graffham ; AJST, Vol 1, June 2002)

Sodium acetate buffer (pH 5)

↓ Sample was heated at 50°C in 5 min

Add 100 mL of 1.4 g/L α-amylase

↓ Operating condition :

- Duration = 2.5 hour
- Temperature = 50°C
- Agitation with different impeller speed for each process (100, **200** and 300 rpm)

Product was analyzed using high performance liquid chromatography (HPLC) → Maltose concentration versus Impeller speed

No of Experiment	Tapioca starch		Sago starch	
	Dry solid (% w/v)	Impeller speed (rpm)	Dry solid (% w/v)	Impeller speed (rpm)
1	8.0	100	8.0	100
2	8.0	200	8.0	200
3	8.0	300	8.0	300
4	10.0	100	10.0	100
5	10.0	200	10.0	200
6	10.0	300	10.0	300
7	12.0	100	12.0	100
8	12.0	200	12.0	200
9	12.0	300	12.0	300

Table 1 : Experimental design

RESULT

Table 2 : Gross yield for sago starch

dry solid (%w/v)	Impeller speed (rpm)		
	100	200	300
8	**32.63**	32.49	32.09
10	26.09	25.79	25.63
12	21.72	21.07	21.29

Table 3 : Gross yield for tapioca starch

dry solid (%w/v)	Impeller speed (rpm)		
	100	200	300
8	**31.96**	31.79	31.71
10	25.49	25.62	25.53
12	21.21	21.01	21.32

1. Both sago and tapioca starch :

i. The maximum value of dry solid concentration
= 8% w/v
= 80 g in 1 L

ii. The optimum speed of impeller = 100 rpm

2. Yield

1. $Y_{\text{sago starch}}$ **= 32.63 %**
2. $Y_{\text{tapioca starch}}$ **= 31.96 %**

Yield, Y = 50% to 60% maltose

(Norman BE ; London : Academic Press, 1979. p. 339 - 376)

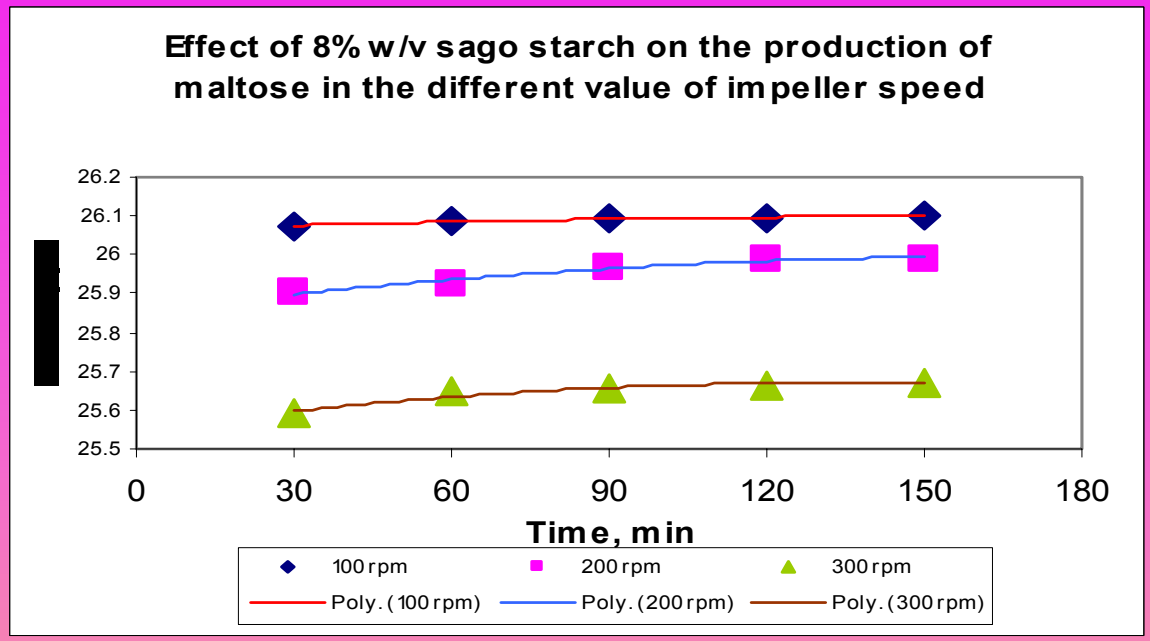
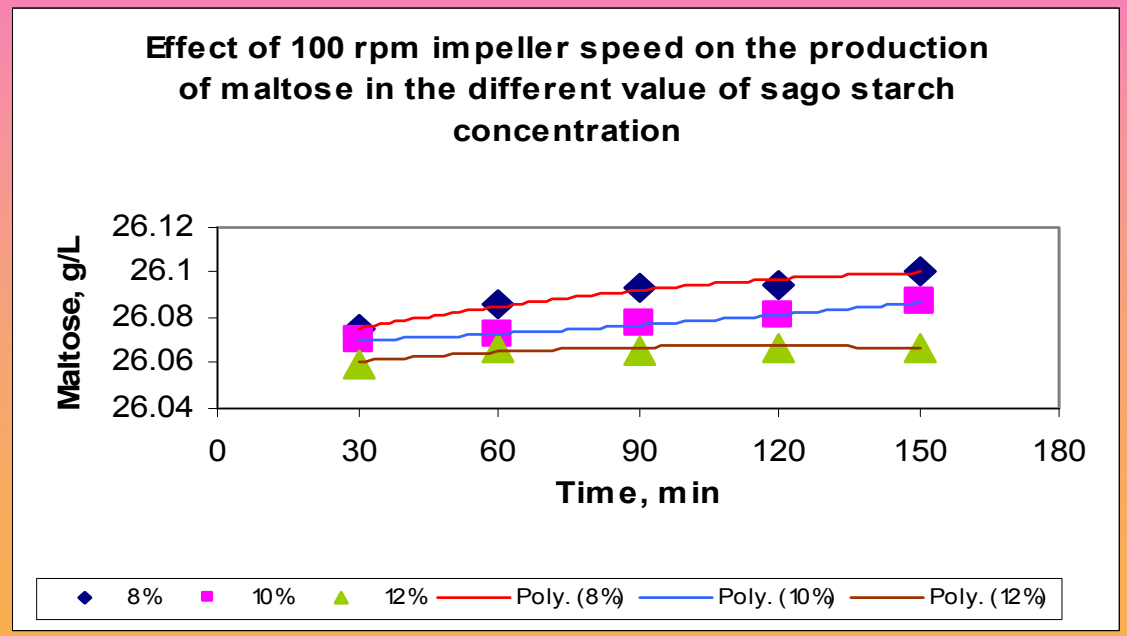
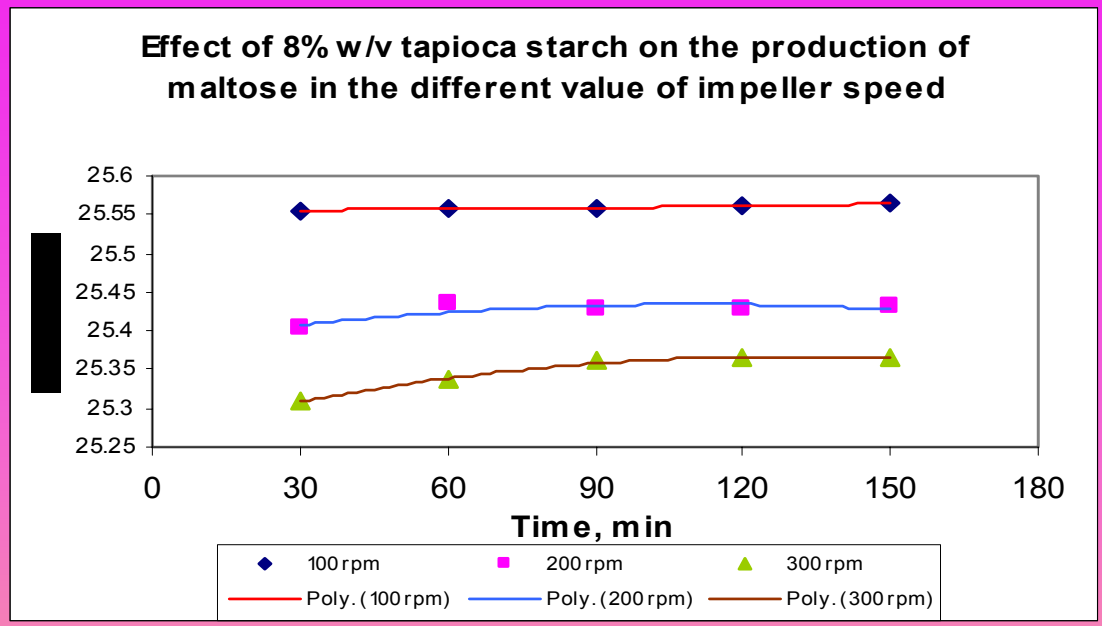


Figure 1

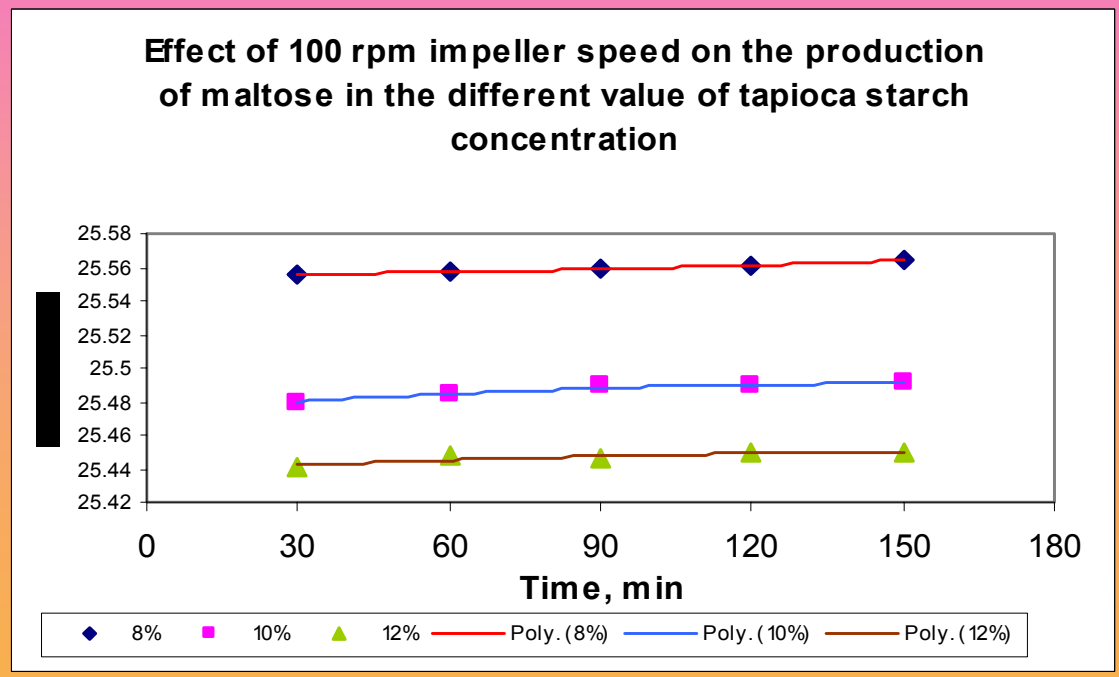
Figure 2



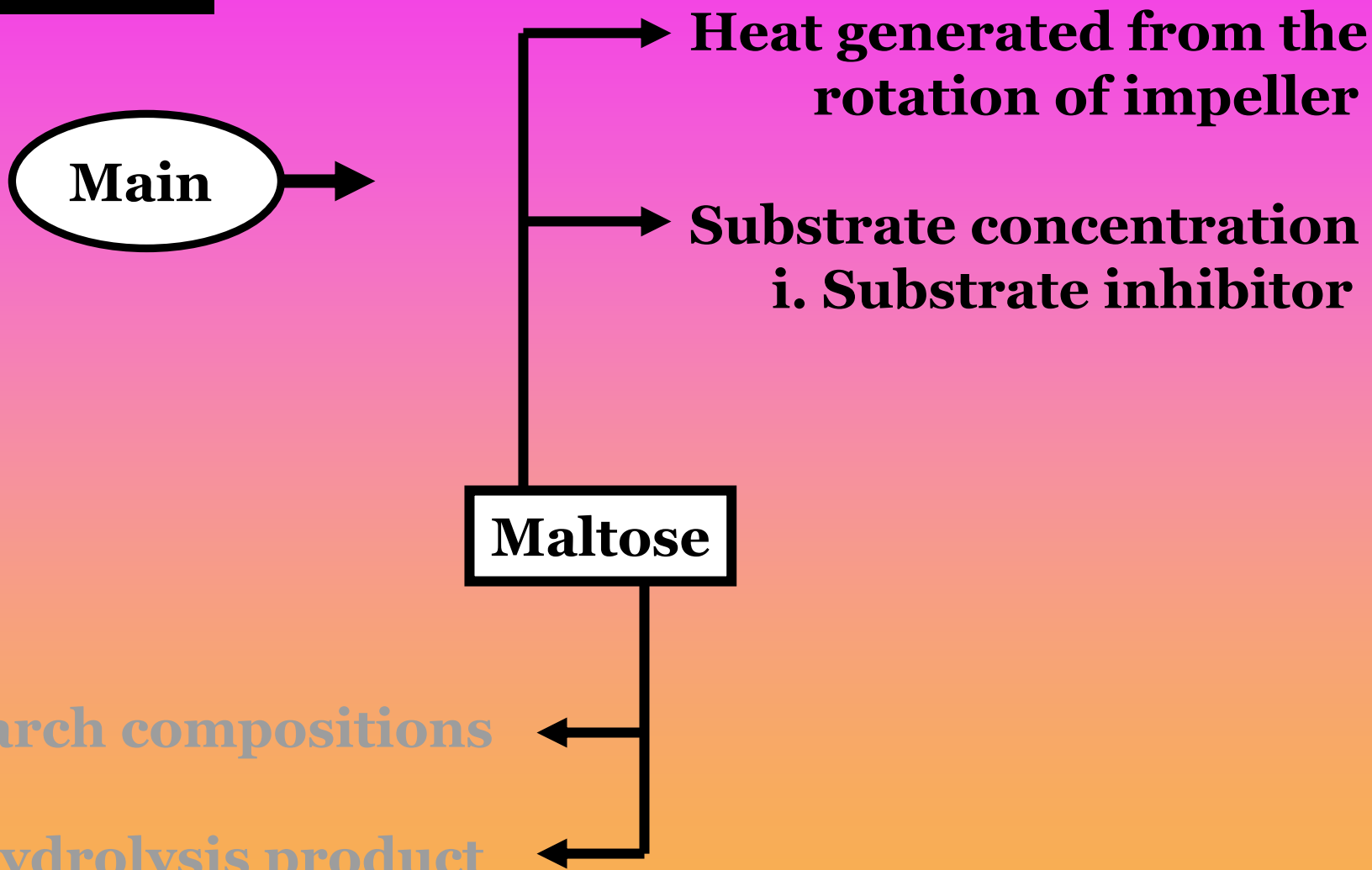


← Figure 3

Figure 4 →



HOW


Main
Heat generated from the rotation of impeller
Substrate concentration
i. Substrate inhibitor
Maltose
Starch compositions
Hydrolysis product

1. Heat Generated

Heat, Q = Power consumed in liquid x time

$$= P.t$$

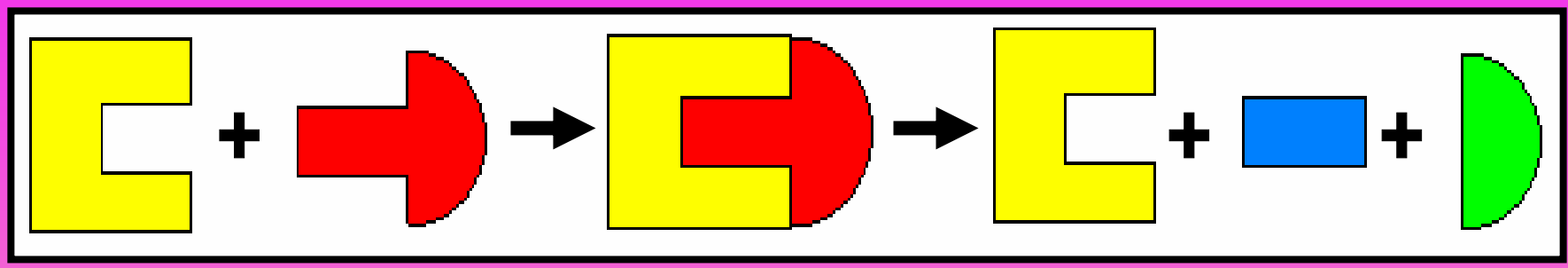
$$= D^5N^3 \rho \, . \, (P_o) \, . \, t$$

$$= D^5N^3 \rho . K \, (N_{Re})^n \, (F_r)^m \, . \, t$$

as N_{Re} : Reynold number
F_r : Froude number
P_o : Power number
N : Rotational frequency of impeller

$N \uparrow Q \uparrow$ → **Enzyme denatured**

2. Substrate Concentration



$$v = \frac{V_{max}[S]}{K_M + [S]}$$

v = Rate of product formation
$= k_2 [ES]$

V_{max} = Maximum velocity
$= k_2 [E_o]$

Enzyme used : α-amylase
: E_o = 1.4 g/L

i. Substrate inhibition

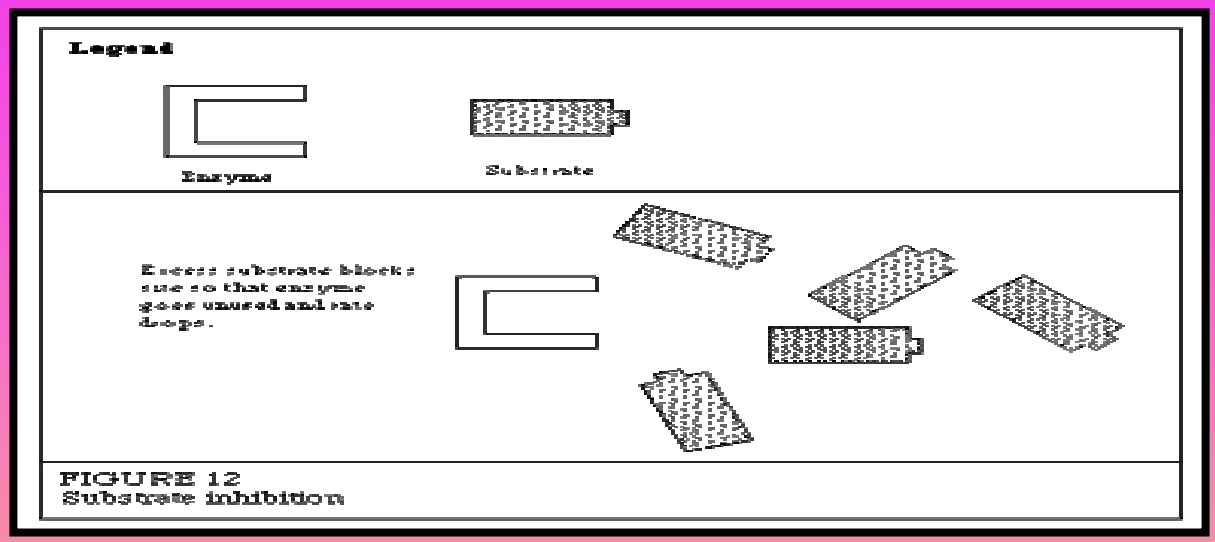
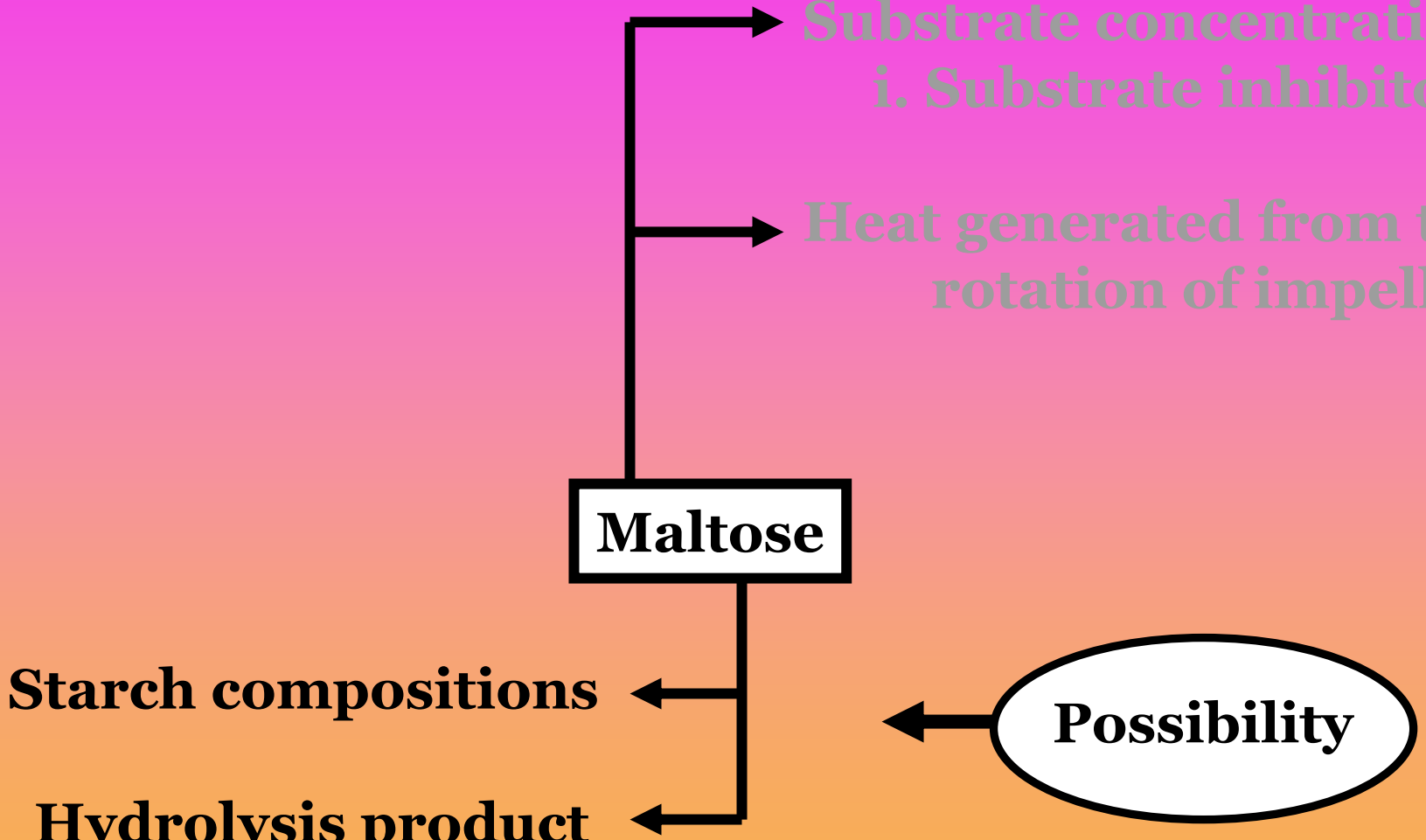


FIGURE 12
Substrate inhibition

$$V = \frac{Vm}{1 + \frac{[S]}{K_{si}}}$$

[S] ↑ v ↑ → Maltose ↓

HOW


Substrate concentration
i. Substrate inhibitor
Heat generated from the rotation of impeller
Maltose
Starch compositions
Hydrolysis product
Possibility

3. Starch Compositions

1. Sago starch
   Amylose = 26%
   Amylopectin = 74%

2.Tapioca starch
   Amylose = 17%
   Amylopectin = 83%

CONCLUSION

1. Yield
   i. $Y_{\text{sago starch}}$ = 32.63 %
   ii. $Y_{\text{tapioca starch}}$ = 31.96 %

2. $\text{Maltose}_{\text{sago}} > \text{Maltose}_{\text{tapioca}}$

3. Maximum dry solid concentration = 8% w/v

4. Optimum impeller speed = 100 rpm

5. Substrate and enzyme concentration effected the production rate

RECOMMENDATION

1. **Use higher value of enzyme concentration**
   = 8 % w/v amylase for 10% dry solid

2. **Use Rice-malt extract**

→ **82% maltose**

(G.S Ayernor, T.K Hammond and A.Graffham ;
AJST, Vol 3, No 1 : June 2002)

THANK YOU

Hydolysis Product

High concentration of hydrolysis products

i. **Mass transfer decreased**
ii. **Osmotic effect (reducing the water activity in the vicinity of ES complex**

(Hill et all)

Penicillium expensum

Both enzymes catalyze transglycosylation reactions during the concentration-dependent degradation of maltooligosaccharides produced on hydrolysis of starch.

The significantly higher yields of maltose achieved with the P.expansum a-amylase, compared to the A. oryzae enzyme, appear to be due to the mechanism of action of P. expansum on maltotriose.

The A. oryzae enzyme only switches from unimolecular to multimolecular events at high concentrations of the trisaccharide.

(Evelyn M. Doyle, Agnes M. Noone, Catherine T. Kelly, William M. Fogarty
Enzyme and Microbiology Technology 25(1999) 330-335)

RME was prepared by malting paddy rice for 10 days at 28°C and diastatic power determined.

The fresh ground malt was weighed into 100ml of citric acid-sodium phosphate buffer (pH was adjusted using 0.1M HCl and 0.1M NaOH) and allowed to stand for an hour with intermittent stirring, after which the mixture was centrifuged for 15 minutes at 3000 rpm. The supernatant was used as rice malt extract (RME) for the hydrolysis.

At high starch concentration :

i. **10% w/v starch**
ii. **8% w/v RME**