The Behavior of Invert Sugar in Sugar Processing

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INTRODUCTION

The mixture of equal amounts of glucose and fructose is called "invert" sugar in the sugar industry. They occur naturally in sugar cane juice at 3 to 5% of total soluble solids (Clarke, 1988) and in beet juice at 0.4 to 0.8% (McGinnis, 1971). The content of glucose and fructose in sugar cane juice varies greatly depending on the varieties and the maturity of cane, weather and geographic conditions, and the condition of fertilization (Meade and Chen, 1977; Legendre, 1988). These monosaccharides are also produced during sugar processing through sucrose hydrolysis under even slight acidic conditions (Edye and Clarke, 1992).

The presence of invert in raw material has following undesirable effects during sugar processing. (1) It has been shown the autocatalysis of sucrose in the presence of invert because the degradation of invert forms carboxylic acids. These acids lower the pH of sugar solution and causes further conversion of sucrose into glucose and fructose. In both raw sugar mill and refinery even a low level of hydrolysis of sucrose will result in significant sucrose loss with economic consequence (de. Bruijn, 1986a, 1987). (2) The formation of coloring materials due to degradation, Maillard reaction of invert with amino acids and the caramelization of invert increases the cost of decolorization, and increases the color of final products (Ramaiah and Kumar, 1968; Fleming, et al., 1968; Clarke, et al., 1987). (3) The existence of invert in either raw sugar or refined sugar increases the possibility of caking during storage, particularly when fructose exists because fructose is moisturized easily and caking of sugar is caused by the migration of moisture; in addition, trace amount of invert, particularly on the surface of crystal affects the flowability of sugar crystals hence, the handling characteristics of sugar. (4) The presence of invert increases the viscosity of sugar liquor in the later stage of sugar boiling (Swindells, et al. 1958; Hinton, 1929).

In the sugar refinery, invert comes from raw sugar and produced during refining process. The invert contents in raw sugar vary from 0.2% to 0.6% of dry mass depending on sources and quality of raw sugar. After affination (sugar washing), the percentage of invert in washed sugar is reduced 0.06 - 0.1%. This indicates that molasses film of raw sugar crystals is high in invert. The concentration of invert in affination syrup varies from 3 to 5%. Although the affination syrup consists of only about 7% of melt sugar, the cost for recovering sucrose from affination syrup accounts for up to 25% of the refining costs. In addition, the carbonate cake generated by carbonation process is costly to dispose. For example, in a sugar refinery 0.8% CaO on the weight of raw sugar is usually used to treat melt liquor (washed sugar liquor), and 1.5 to 1.6% $CaCO_3$ is generated by the carbonation, about 25 tons of wet carbonate cake is produced per day for a 1000 ton raw sugar/day refinery. The total cost of disposing the wet cake is $20 to $30 per ton depending on the distance of shipping. Therefore, it is imperative to clarify the behavior of invert in sugar refining process and to find the optimum condition to destroy the invert in washed sugar and affination syrup when necessary with the minimal use of lime and the least loss of sucrose to improve the yield of the refining operation.

Regarding the alkaline degradation of invert, many mechanisms and kinetic models has been proposed. The kinetic models derived by and by de Wilt and Lindhout (1973), de Wit (1979), and de Bruijn (1987) reflect the mechanism and the role of enediol ions as an important intermediate during the degradation of monosaccharides the most. However, the models involve sugar anions which are difficult to be determined. The simplified reactions of monosaccharides degradation in alkaline solution (de Bruijn, 1987) is shown in Figure 1.
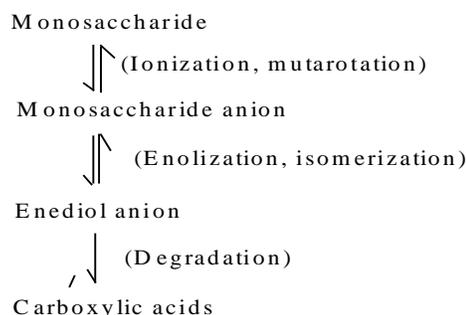
Monosaccharide
$\Updownarrow$ (Ionization, mutarotation)
Monosaccharide anion
$\Updownarrow$ (Enolization, isomerization)
Enediol anion
$\downarrow$ (Degradation)
Carboxylic acids

Figure 1 The simplified reactions of monosaccharides degradation in alkaline solution (de Bruijn, 1987*)*

Many kinetic models have been proposed to describe the interconversion and degradation of monosaccharide under alkaline conditions. An empirical kinetic equation established by Vukov (1965a) and still in use is

$$\log k = 16.9 - 5620/T - pOH$$

where T is reaction temperature in K and k is rate constant in min^{-1}

This equation reflects the effects of pH and temperature on rate constants of invert alkaline degradation. It is valid from pH 10 to 12.5 and temperature 60 to 130°C. According to this equation, rate constants increase 10 times as pH increases one unit at constant temperature. This is much larger than the results reported by de Bruijn et al. (1987).

In this work, the kinetic model of invert alkaline degradation was further simplified based on the experiment results, the correlation between the degradation rate of invert and the major factors such as pH and temperature was established, the nature of base and the concentration of sucrose on invert degradation and the formation of lactic acid and color were evaluated, and the effect of invert destruction on the efficiency of purification in a simulated continuous carbonation process was evaluated through the measurement of color removal, calcium removal and filterability of carbonated sugar liquid.

MATERIALS AND METHOD

Materials: Sucrose, D-glucose, D-fructose, mannose, psicose and lactic acid were the products of Sigma Chemical Company (St. Louis, MO). Raw sugar and washed sugar were from Domino Sugar Co. (Chalmette, LA). Alkaline chemicals: Calcium oxide (CaO), sodium hydroxide (NaOH) and potassium hydroxide (KOH) were purchased from Fisher Scientific.

Experiments: The parameters in present experiments were chosen based on the sugar refining conditions. pH ranged from 9 to 11, temperature 70 to 90°C, and the concentration of invert sugar 0.02 to 0.04 M. CaO was dissolved in water as 15% suspension, NaOH and KOH were prepared as 5 M solutions. Invert sugar solutions were made by dissolving equal amounts of glucose and fructose in a 1,000 ml beaker with 800 ml of deionized water, then adjusted to desired pH with $Ca(OH)_2$, or NaOH or KOH solution. The invert solution was then transferred into a volumetric flask and DI water was added to the mark of the flask. The reactions were

conducted in a jacketed beaker reactor (1 liter) connected with a temperature controlled circulating water bath. The jacketed beaker was preheated to the desired temperature, then the invert solution was poured into the reactor and heated rapidly to the desired temperature (usually in 15 minutes). The pH of reaction solution was monitored using a pH temperature compensated pH metore and the pH was manually controlled by adding alkaline solutions. Samples were taken into 20 ml glass bottles after the desired temperature was reached, then cooled to room temperature immediately to stop the reaction. The samples were then filtered through 0.45 μm membrane and stored in the freezer for further analysis. The sampling interval varied according to the reaction conditions. Ten samples were taken for each experimental condition.

Analysis of Invert: An HPLC method using a post column derivatization reaction procedure (Wnukoski, 1983) was used to determine the glucose and fructose in samples taken at different reaction time. The column was 4.6 mm x 250 mm steel cartridge column containing 4 μm Nova-pack® spherical silica bonded with trifunctional amino propylsilane. 15 μl of Filtered sample was injected by WISP 710 B Automatic Injector (driven by 40 psi compressed air). The mobile phase was 80% acetonitrile in deionized water; the flow rate was 0.8 ml/min. After separation, the fructose flew out first then the glucose. They reacted with 0.2% tetrazolium blue chloride in 0.1 N NaOH at 90ºC in the heating coil respectively and formed dark blue pigments with maximum absorbance at 430 nm. 486 Tunable Absorbance Detector was used to determine the residual glucose and fructose after the post column reaction. The signal detected was integrated by Waters 730 Data Model.

Analysis of lactic acid: Ion Chromatography system (DIONEX) was used to separate the organic acids and to determine lactic acid quantitatively. The system consists of BIO-RAD Module AS-100 HPLC®automatic sampler, gradient pump, Dionex Conductivity Detector, Dionex Eluent degas Module and Dionex 4400 integrator. The column used is IONPAC ® AS11 (4 x 250 mm). Gradient elution procedure was used for the better resolution. Mobile phases include HPLC grade water (Eluent 1), 5 mM NaOH solution (Eluent 2) and 100 mM NaOH solution (Eluent 3). The signal to noise ratio was improved by Cation Self-Regenerating Suppressor. The use of chemical suppressor eliminates the conductivity of mobile phase while the conductivity of analyte is enhanced. The injection volume was 20 μl, flow rate was 1.5 ml/min. The linear gradient is shown below:

Elution time (min)	% Eluent 1 (water)	% Eluent 2 (5 mM NaOH)	% Eluent3 (100mM NaOH)
0	90	10	0
2.0	90	10	0
20	0	100	0
26	0	65	35
35	0	0	100

Color measurement: The color of all samples was measured by ICUMSA (International Commission for Uniform Methods of Sugar Analysis) method GS-1-7 1994. Samples were filtered through 0.45 μm membrane to remove turbidity. The pH of the sample then adjusted to 7.0 ± 0.1 The absorbance (Abs) of the neutralized solution was measured at a wavelength of 420 nm by Spectrophotometer (Gilford Response II, Ciba Corning Diagnostics Corporation, Ohio).

RESULTS AND DISCUSSION

1. KINETICS OF INVERT ALKALINE DEGRADATION

1.1. Simplification of Kinetic Models of Invert Alkaline Degradation

As pointed out earlier, the kinetic models derived by de Wilt and Lindhout (1973), de Wit (1979), and be Bruijn (1987) involve sugar anions which are difficult to be determined. In this study, the mixture of glucose, fructose, mannose and psicose, and the invert alkaline reaction mixture obtained at pH 11.0, 90ºC were analyzed by HPLC. Mannose and psicose were not found in the reaction mixture. The result indicates that the alkaline degradation may not form mannose and psicose or their concentrations are too low to be detected, thus can be ignored when calculating rate constant of invert degradation. According to this result, following assumptions were made to emphasize the role of enediol ions and to simplify the kinetic model of invert alkaline degradation. 1) The conversions between invert sugars and their anions are very fast, and these inter-conversion reactions are in a fast equilibrium state. The concentrations of glucose and fructose measured by HPLC include the concentrations of their anions. 2) The presence of psicose in the mixture can be ignored, and mannose and glucose together can be treated as glucose. 3) The enediol anions intermediates have pseudo steady-state concentrations during the

reaction, i.e., $dE^-/dt = 0$ since the concentration of enediol anions are very low (less than 1 mol% of total sugars) in the equilibrium mixture (de Wit, 1979). 4) The formation of enediol anions from sugar anions is the rate-limiting step of invert degradation.

$$G \underset{k_2}{\overset{k_1}{\rightleftarrows}} E^- \overset{k_5}{\longrightarrow} A$$
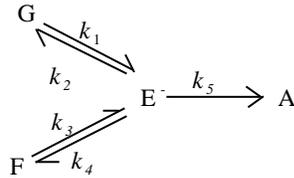$$F \underset{k_4}{\overset{k_3}{\rightleftarrows}}$$

Figure 2. Simplified kinetic model of invert alkaline degradation (G=glucose, F=fructose, E=enediol ions and A=acids)

Based on the above assumptions, the kinetic model of invert alkaline degradation shown in Figure 1 could be simplified as shown in Figure 2. According to this model, following equation can be derived:

$$\frac{k_g}{k_f} = \frac{k_1}{k_3} \tag{1}$$

This equation demonstrates that the degradation rate of glucose and fructose are determined by the formation of enediol ions from corresponding sugars.

1.2 Factors Influencing the Kinetic Behavior of Invert Sugar

Figure 3 shows the pattern of invert alkaline degradation with time at different temperature and pH. It has been proven that the degradation of invert under alkaline condition is first order reaction. According to the data in figure 3, the rate constants of glucose, fructose and total invert were calculated and listed in table 1 and table 2.

Table 1 and Table 2 show that rate constants of glucose, fructose and total invert degradation increased 4.5 to 7.5 times with one unit increase in pH at 80°C, increased 3 to 4 times with a 10°C increase in temperature at pH 10.0, when the initial concentration of invert was 5,000 mg/L (equal amount of glucose and fructose), and $Ca(OH)_2$ suspension was used as a base. Table 1 and 2 also show that the rate constant of fructose degradation was almost always larger than that of glucose degradation under same conditions. This indicates that the conversion

of fructose to enediol ions was faster than the conversion of glucose to enediol ions under the experimental conditions. A further analysis revealed that the rate constants of total invert were the average of the rate constants of glucose and fructose under the experiment conditions. This was confirmed by the results of other experiments in which NaOH, KOH and Ca(OH)$_2$ were used as bases (Table 3). This relationship should be always correct because the total invert concentration is the sum of the concentrations of glucose and fructose, and rate constant is concentration independent at constant pH and temperature.
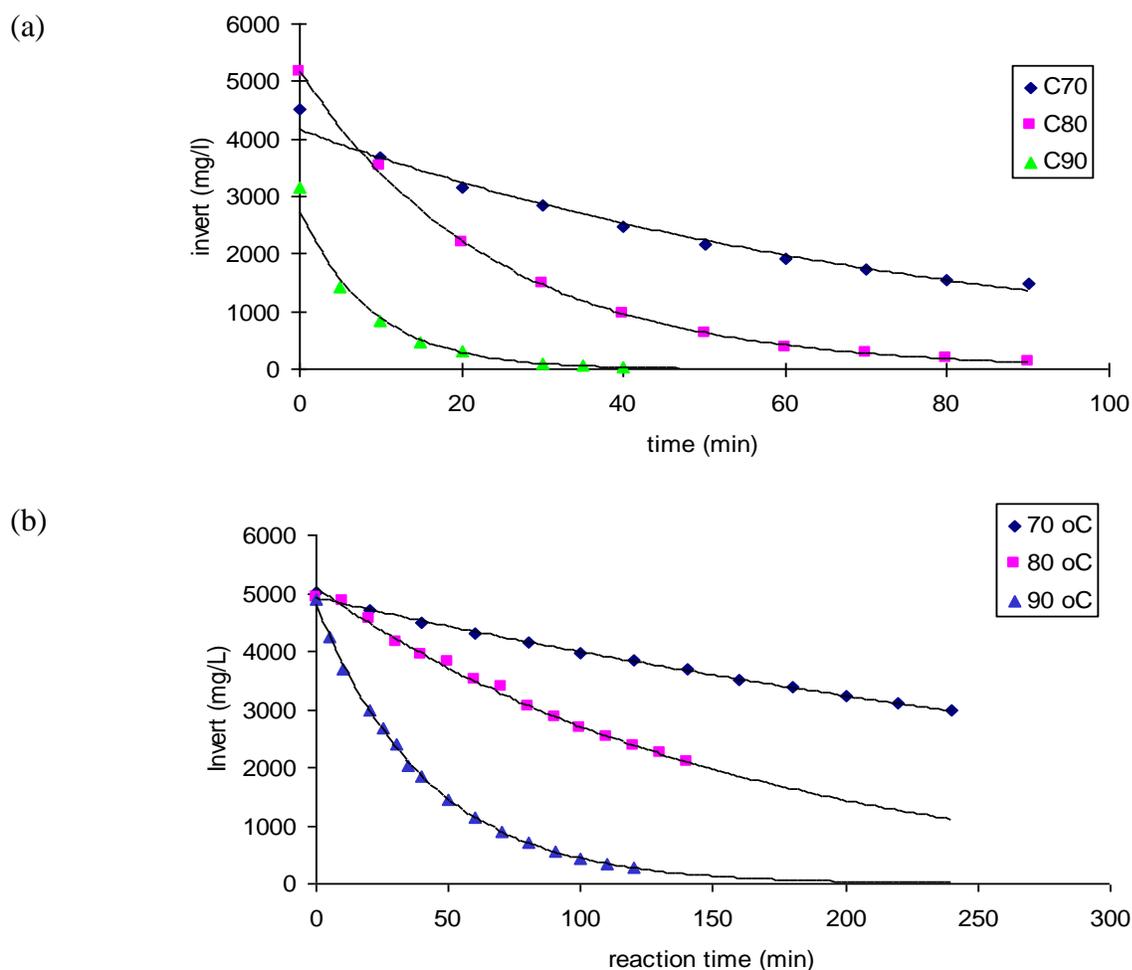
$$k = (k_g + k_f)/2 \qquad (2)$$

(a)

(b)

Figure 3. Invert degradation at different temperature. (a) pH = 11., (b) pH = 10.0

Table 1 The effect of pH on rate constants of invert degradation
(at 80°C, initial concentration of 5,000 mg/L)

pH	k_f (min^{-1})	k_g (min^{-1})	k (min^{-1})	$(k_f+k_g)/2$ (min^{-1})
pH 9.0	0.0025	0.0014	0.0019	0.0020
pH 10.0	0.0089	0.0086	0.0088	0.0088
pH 11.0	0.0667	0.0671	0.0669	0.0669

Table 2. The effect of temperature on rate constant of invert degradation
(at pH 10.0, initial concentration of invert 5,000 mg/L)

Temperature (^{o}C)	k_f (min^{-1})	k_g (min^{-1})	k (min^{-1})	$(k_f+k_g)/2$ (min^{-1})
70	0.0033	0.0012	0.0022	0.0025
80	0.0067	0.0056	0.0063	0.0062
90	0.0250	0.0230	0.0239	0.0240

Table 3 The relationship among k, k_g and k_f under the experimental condition
(pH=11.0, C_g=2,500 mg/L, C_f=2,500 mg/L)

$k \times 10^4$ (min^{-1})	CaO			KOH			NaOH		
	70°C	80°C	90°C	70°C	80°C	90°C	70°C	80°C	90°C
k_g	7.95	18.76	48.02	7.25	14.70	39.3	7.91	15.41	34.4
k_f	8.03	17.60	47.32	5.78	12.90	34.87	6.64	14.3	32.78
k	7.99	18.20	47.72	6.47	13.60	37.70	7.25	14.85	33.56
$(k_g+k_f)/2$	7.99	18.18	47.67	6.52	13.8	37.08	7.27	14.86	33.55

1.3 Activation Energy of Invert Alkaline Degradation

It is generally accepted that hydroxyl ions act as a catalyst of invert degradation, and the higher the hydroxyl ion concentration, the faster the reaction. Catalysts accelerate chemical reaction by lowering the activation energy. Vukov (1965) found that the activation energy of invert degradation was temperature independent, and the activation energy in strong alkaline solution (pH = 12 to 13) was smaller (25 kcal/mol) than that in the weak alkaline solution (pH <11, 26.71 kcal/mol). The experiment results of this study listed in Table 4 shows that increase in pH, i.e.

the concentration of OH$^-$, did not lower the activation energy. Under the experimental conditions (pH 9.0 - 11.0), the activation energy of invert degradation was pH independent and the value was 30.75 kcal/mol with a frequency factor 19.415 min^{-1}. The activation energy obtained by Sapronov (1969) at pH 10.0 is 29.55 kcal/mol, which is very close to the results in Table 4. Vukov's equation thus can be rewritten as equation (3).

$$\log k = 19.42 - 6712./T - 0.712 pOH \qquad\qquad (3)$$

Table 4 The effect of pH on activation energy of invert alkaline degradation
(pH =9.0 - 11.0, T=70 - 90ºC, Ca(OH)$_2$ used as a base)

	PH 9.0	PH 10.0	PH 11.0
E (kcal/mol)	30.69	30.81	30.28
Mean± Stdev	30.75 ± 0.278		
Log A (min-1)	19.396	19.484	19.365
Mean	19.415 ± 0.062		

To verify this equation, other experiments were performed at different pH and temperature combinations. The rate constants measured by fitting the data to exponential equation [C] = [Co]exp (-kt) were compared to the rate constants calculated by the equation (3) and Vukov's equation. The results are listed in Table 5. It is evident that rate constants from experiment data are close to that from equation (3) but far away from that calculated from Vukov's equation. The lower rate constants calculated by Vukov's equation may be due to the effects of buffering agent. Potassium dihydrogen phosphate (KH$_2$PO$_4$) and calcium lactate were used as buffers when Vukov measured the rate constants of invert alkaline degradation in Ca(OH)$_2$ suspension (Vukov, 1965). Although the effect of KH$_2$PO$_4$ is not known, calcium lactate is one of the products of invert alkaline degradation in Ca(OH)$_2$ suspension, which has inhibitory effect on the degradation of invert.

1.4 The Effect of The Nature of Base on Rate Constant and Activation Energy of Invert Alkaline Degradation

As shown Table 6, at 70 and 80ºC the rate constants were larger when KOH and NaOH were used as base than that when Ca(OH)$_2$ was used as base, the activation energies of invert

degradation in three base solutions were different, and were affected by pH of the solutions with the exception of $Ca(OH)_2$.

Table 5 The comparison of rate constants k (min^{-1}) measured by experiments and calculated by equations (3) and Vukov's equation

T (^oC)	Experimental k x 10^3 (min^{-1})			k x 10^3 (min^{-1}) by equation (3)			k x 10^3 (min^{-1}) by Vukov'sequation		
	pH 9.55	pH 10.0	pH 10.5	pH 9.55	pH 10.0	pH 10.5	pH 9.55	pH 10.0	pH 10.5
75	0.9	3.0	4.4	0.9	1.9	4.3	0.2	0.5	1.8
80	2.1	2.2	10.1	1.7	3.6	8.1	0.3	0.9	3.0
85	3.6	11.4	26.3	3.1	6.6	15	0.6	1.5	5.0

Table 6 The effect of the nature of base on rate constant and activation energy of invert alkaline degradation at pH 10.0 and pH 11.0 and Co=5,000 mg/L

	k x 10^3 (min^{-1})					
	PH 10.0			PH 11.0		
T (^oC)	$Ca(OH)_2$	KOH	NaOH	$Ca(OH)_2$	KOH	NaOH
70	0.9	1.4	1.4	5.4	6.5	7.2
80	2.5	3.5	4.2	17.6	13.5	15
90	10.4	8.8	10.9	64.1	37.7	33.6
E(kcal/mol)	30.11	26.96	25.82	30.28	21.79	18.97
LogA	20.09	18.19	17.58	20.00	16.65	14.93

The activation energy was lower when KOH and NaOH were used, and the higher the pH, the higher the concentrations of K^+ and Na^+, and the lower the activation energy. It was possible that besides the action of hydroxyl ions, cations of these bases influenced the rate constant of invert degradation, because their concentrations differed at the same pH. Generally speaking, catalysts catalyze chemical reactions by lowering the activation energy. Therefore, K^+ and Na^+ had catalytic effect on invert alkaline degradation compared with Ca^{2+}.

1.5 The effect of sucrose on the degradation rate of invert

Comparing with the rate constant of pure invert degradation at the same pH and temperature, one can see from Table 7 that the degradation rate increased with the increase in

sucrose concentration. At pH 10.5 and 85°C, the rate constant increased approximately 0.004 min^{-1} (12%) with 10% increase in sucrose concentration. According to Vukov (1965a), sucrose accelerating invert degradation was because the presence of sucrose increased the solubility of lime. However, Sheng (1992) stated that the presence of sucrose slowed down the degradation of invert sugar but gave a darker solution.

Table 7 The effect of sucrose concentration on rate constant of invert
degradation at pH10.5, 85 $^{\circ}$C, Ca(OH)$_2$ as a base

% Sucrose	0%	35%	50%	60%
Kx10^3 (min $^{-1}$)	13.9	31.4	33.2	38.6
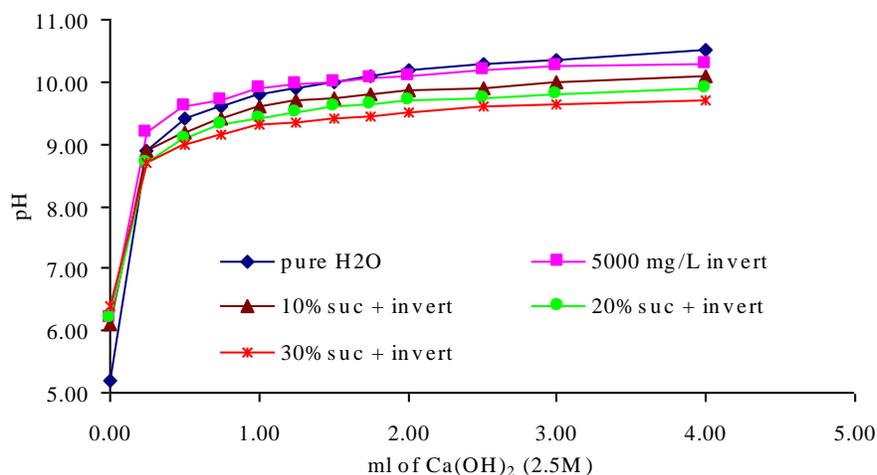
Figure 4. The effect of sucrose concentration on pH of aqueous solution.
(85°C, concentration of Ca(OH)$_2$ suspension 2.5 M)

The effect of sucrose concentration on pH measurement should not be neglected. The titration curves (Figure 4) showed that the concentration of sucrose influenced the measurement of pH. The pH reading was lower in sucrose solutions than in aqueous solutions when an equal amount of Ca(OH)$_2$ suspension was added in solutions. The higher concentration of sucrose, the lower was the pH value.

Any pH electrode behaves according to Nernst equation: $E = E_0 + 0.0591 \log[a_H]$. The usefulness and accuracy of an electrode are judged by the closeness of its response to the

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theoretical slope. When electrodes are in sucrose solutions, their response is not Nernstian: the slopes are considerably off the theoretical values (Clarke, 1971). Clarke (1971) explained the effect of sugar solutions on electrode performance by structural theories of solutions. One of the principal structural concepts related to the sucrose solution chemistry is Frank-Evans iceberg theory. The theory considered that when a non-polar molecule is dissolved in water, a microscopic "iceberg", part of a larger cluster of water molecules, would form around it, causing a loss of entropy and a decrease in the total volume of solution. This means that the dissolution of a non-polar molecule, such as sucrose, modifies the structure of water in the direction of greater crystallinity, i.e., the hydrogen-bonding is increased. The greater number of bonds associated with a water molecule, the lower is the energy level of the molecule, thus the more stable is the solution. The electrical double layer structure and the behavior of ions in this layer control the response of the pH electrode. In sucrose solution, cations are surrounded by ordered structures of sucrose and water, so that the charge and the effective radius of the cation increase. The electrode, therefore, responds to a different concentration of ions from that in aqueous solution. It can be concluded that the concentration of OH^- in the presence of sucrose is higher than that without sucrose, when pH reading is the same. The degradation of invert is, therefore, faster at the presence of sucrose.

2. LACTIC ACID FORMATION FROM INVERT ALKALINE DEGRADATION

One of the most important effects of invert degradation in alkaline solution is the formation of carboxylic acids. These acids cause undesirable pH drops in the evaporators, thus the loss of sucrose due to acidic conversion (de Bruijn, 1987; Shore, M., 1957). The formation of organic acids and aldehydes was said to be the first step of color formation through invert degradation during sugar refining (Gillett, 1953; Ramaiah et al, 1968). The most recent study (Godshall, 1997) found the strongest correlation between white sugar color and lactic acid among many organic acids such as pyrrolidone-carboxylic acid, glycolic acid and glyceric acid. Organic acids formed through invert alkaline degradation present in the form of anions. These anions can be determined by IC (de Bruijn, 1984; Tusseau and Benoit, 1987).

2.1 The Effect of Initial Concentration of Invert Sugar on the Formation of Lactic Acid

Three experiments were conducted at three different initial invert concentrations (3,600 mg/L , 5,400 mg/L, and 7,200 mg/L) but same temperature ($90^{o}C$) and pH (pH 11.0). The concentration of invert and lactate were measured, and the percentage of lactic acid on the weight of invert degraded at different reaction time were calculated and shown in Figure 5.

The figure indicates that the percentage of lactate decreased with the increase of initial invert concentration. After 50 minutes, as complete conversion reached, the percentage of lactate for initial invert concentrations of 3,600 (C_{01}), 5,400 (C_{02}) and 7,200 (C_{03}) mg/L were 26.75, 20.30 and 14.95%, respectively. The percentage of lactate was reduced about 6% with every 1800 mg/L (0.01 M) increase in the initial invert concentration. This suggests that the concentration of invert influences the pathway of invert degradation, and low invert concentration favors the formation of lactic acid. According to the mechanism of monosaccharides alkaline degradation presented by de Wit (1979), the formation of lactic acid and saccharinic acids are through the same pathway, that is, beta-elimination of enediol anions of trioses and hexoses followed by benzilic rearrangement. The increase in the invert concentration may not favor the benzilic rearrangement. This is consistent with the result of de Bruijn, et al. (1987). At certain initial concentrations, the production of lactic acid increased with reaction time because of the decrease in invert concentration.

Let ΔC_{LA} (mg/L) = $C_{t\text{-lactate}}$ − $C_{0\text{-lactate}}$, ΔC_{Invert} mg/L = $C_{0\text{-Invert}}$ - $C_{t\text{-Invert}}$

(where C_0 represents the initial concentration, and C_t represent the concentration at reaction time t) then $\Delta C_{LA}/\Delta C_{Invert}$ is the production of lactate when invert concentration decreased from C_0 to C_t. By plotting $\Delta C_{LA}/\Delta C_{Invert}$ against the invert concentration C_{Invert} (mg/L), Figure 5 was obtained. The figure indicates that the higher the C_{Invert}, the smaller the $\Delta C_{LA}/\Delta C_{Invert}$. A quantitative relationship (Equation 4) was obtained by regression analysis of data in Figure 5. This equation was valid at pH 11.0 and $Ca(OH)_2$ suspension as base. The production of lactic acid decreased in following way with the increase of invert concentration.

$$\Delta C_{LA}/\Delta C_{Invert} = 42.642 \, C_{Invert}^{-0.729} \qquad (4)$$
$$R^2 = 0.9249$$

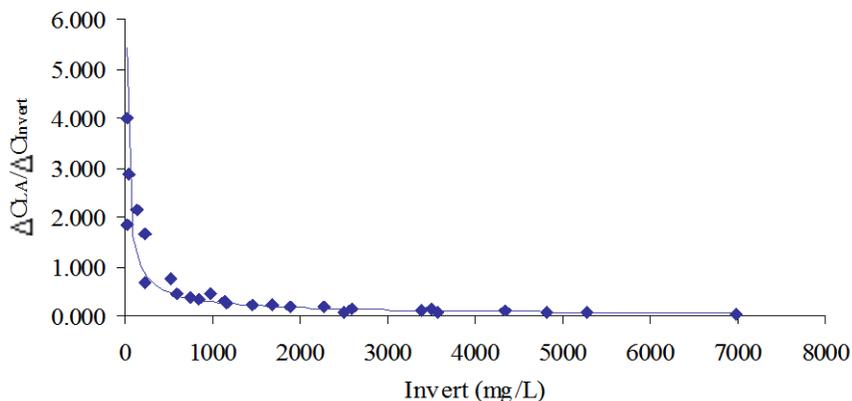Figure 5 The variation of lactic acid production with invert concentration at 90°C, pH 11.0, Ca(OH)$_2$ suspension as a base.

2.2 The Effects of Ph And Temperature on The Production of Lactic Acid in Pure Invert Solutions

Table 8 The effect of temperature and pH on the production of lactic acid in 5,000 mg/L invert solution (pH was adjusted and maintained by adding 15% Ca(OH)$_2$ suspension)

	Lactic acid % on invert degraded		
	pH 9.0	pH 10.0	pH 11.0
70°C	17.46	23.63	30.89
80°C	17.80	21.42	27.46
90°C	14.62	20.30	26.75

The results in Table 8 indicate that at a given pH, temperature did not show significant effect on the production of lactic acid; whereas, at a constant temperature, pH (the concentration of hydroxyl ions) exerted a great effect on the production of lactic acid. The percentage of lactic acid increased about 6% with one unit increase in pH. Higher production of lactic acid at higher pH suggests that higher concentration of hydroxyl ions could lead to retro-aldolization of 1,2-enediol ions of hexoses to form 1,2-enediol of triose, and favors the benzilic rearrangement of triose anions. Low temperature and high pH combination favored the formation of lactic acid.

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2.3 The effect of the nature of base on the formation of lactic acid

The effects of NaOH, KOH and Ca(OH)$_2$ on the formation of lactic acid were shown in Table 9. Among cations of these bases, Na$^+$ and K$^+$ are monovalent, but they have different size because they belong to different periods in the elemental periodic table. K$^+$ and Ca^{2+} have similar size and weight but Ca^{2+} is divalent. The comparison between NaOH and KOH reflects the effect of cation size on the production and the rate of lactic acid formation, while the comparison between KOH and Ca(OH)$_2$ reflects the effect of the number of charges.

Table 9 The effect of nature of base on the production of lactic acid at pH 10 and pH 11.0

	70ºC		80ºC		90ºC	
Base	pH 10	PH 11	pH 10	pH 11	pH 10	pH 11
Ca(OH)$_2$	15.81	25.05	16.87	28.49	18.48	20.29
KOH	11.71	24.27	10.57	21.67	14.65	23.01
NaOH	12.28	--	13.09	26.64	18.27	25.96

The fact that the catalytic effect of NaOH was larger than that of KOH may indicate the electrostatic effects of cations and steric hindrance. Small cation (Na$^+$) has stronger electrostatic effect than large cation (K$^+$) with same charge(s), but has smaller steric hindrance to form complex with sugar ions, thus exerts greater influence on the stability of C-C bond in the presence of hydroxyl ions. Early studies found that increase of the molar ration of Ca^{2+} to monosaccharide increased lactic acid formation at the cost of glycolic acid, acetic acid, saccharinic acid and >C6 acids (de Wit, 1979; de Bruijn, 1987). This phenomenum was explained by the complexation of calcium with β-fructofuranose and β-fructopyranose. The complexation promoted C$_3$-C$_4$ bond fission by retro-aldolization which ultimately leads to an enhanced formation of lactic acid (Figure 6, de Bruijn, 1987).
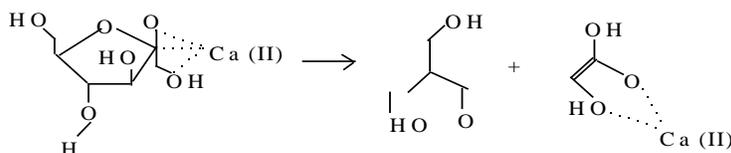

Figure 6. Promoted retro-aldolization of D-fructose by complexation

2.4 The Effect of Sucrose Concentration on Lactic Acid Production

The presence of sucrose increased the rate of invert degradation, and it was reasonable that sucrose might increase the rate of formation of various organic acids. The experiments of invert degradation in different concentrations of sucrose solutions were conducted at pH 10.5 and 85°C, invert residues and lactate formed were analyzed to investigate the impact of sucrose on the percentage of lactic acid. Results were listed in Table 10. As we can see that after 50 minutes of reaction, concentrations of invert residue were 154, 130, 105 and 58 mg/L when sucrose concentrations were 0, 35, 50 and 60%. Comparing with the initial concentration of invert, more than 97% of invert was degraded. Therefore, we could assume that at pH 10.5 and 85°C, after 50 minutes reaction, invert was completely degraded.

Table 10 The effect of sucrose concentration on the production of lactic acid at 85°C, pH 10.5, Ca(OH)$_2$ suspension as base

Time (min)	Invert Sugar (mg/L)				Production of Lactate % On Invert Degraded			
	0% sucrose	35% sucrose	50% sucrose	60% sucrose	0 % sucrose	35% sucrose	50% sucrose	60% sucrose
initial	5000	5100	5210	5291	0	0	0	0
0	4431	4113	3963	4575	18.80	9.88	12.98	21.57
10	2340	1835	2037	2017	14.06	19.17	29.30	25.34
20	1280	943	867	795	15.27	27.68	34.10	31.92
30	541	479	409	245	19.62	33.48	36.78	36.43
40	304	233	235	170	23.45	37.31	38.92	40.29
50	154	130	105	58	32.19	43.36	41.25	41.06

It was suspected that the increase of lactic acid production was due to the alkaline degradation of sucrose at high pH. Therefore, a series of experiments of sucrose alkaline degradation was conducted in 60% refined sucrose solutions at 90°C to verify this speculation. Refined sugar solutions used in the experiments contained 154.68 mg invert per liter. After 100 minutes heating at 90°C and pH 10.3, 10.6 and 10.9 (corresponding to 0.2, 0.4 and 0.8% of lime), some invert was still present in the solutions which was impossible for pure invert solution. In

addition, lactic acid produced after 100 minutes were 107.98, 146.95 and 146.96 mg/L (69.8%, 95.0%, and 120.0% of the original invert), respectively, which were close to or even higher than the initial concentration of invert in the sucrose solutions. These results prove that the increase of lactic acid production in the presence of sucrose is the degradation of sucrose at high pH and temperature and the degradation of the small amount of invert present in refined sugar. The results also imply that the degradation of sucrose in alkaline solution may be through the hydrolysis of sucrose into glucose and fructose, although the finding of Harris et al (1980) does not support this mechanism.

The effect of sucrose solution on the behavior of pH electrode also accounted for the faster formation of lactic acid in the presence of sucrose. Higher concentrations of hydroxyl ions and calcium ions in the presence of sucrose speeded up the invert degradation and favored the formation of lactic acid when the indications of pH were the same.

3. THE DEVELOPMENT OF COLOR DURING INVERT ALKALINE DEGRADATION

One of the most obvious phenomena of invert alkaline degradation is the formation of color. Color increase should be consistent with invert decrease and lactic acid increase during the reaction. Because the formation of color is associated with invert degradation and the removal of color is one of the major concerns of sugar refinery, therefore, the study of color development and removal become extremely important.

The colorants concerned in the refinery were classified into four types in 1984 according to origins and chemical structures (Clarke, et al., 1984; Clarke and Blanco, 1986). Type 1 are plant pigments including: (1) four principal types of naturally occurring colorants (chlorophylls, xanthophylls, flavones and anthocyanins) which are already colored in their original forms in the cane and are not removed completely through raw sugar processing, and (2) the substances which are normally colorless in their original forms but which can develop color during sugar processing, they are phenolics and amino compounds. Type 2 is Maillard reaction products (melanoidins). This type of colorant is formed by the reaction between reducing sugar and amino compounds in sugar solution. Type 3 is thermal degradation products (caramels) at very high temperature. Type 4 is alkaline degradation products of sugars. Alkaline degradation products of fructose, especially in refineries at pH > 7.5, can polymerize to high molecular weight substances (Clarke, 1997). It has been reported that most of the sugar colorants involved in refining have an

anionic nature, being charged negatively at higher pH values (Chou and Rizzuto, 1972). Therefore, strong positively charged absorbents such as strong base ion exchange resins and newly formed calcium carbonate are very efficient to remove colorant in refining materials.

Color formation under alkaline condition is extremely complex. It was believed that the fragmentation of invert sugars leads to the production of three-carbon compounds followed by a series of condensation and polymerization reactions between the various three-carbon aldehydes and ketonic intermediates (Bobrovnik and Rudenko, 1995; Godshall, 1997). However, until now, the mechanism of color formation due to invert alkaline degradation is not completely clear.

Assuming that three-carbon acids (C_3-acids) are the precursors of colorants and colorants are formed by condensation polymerization of these three-carbon acids, the process should occur by the mechanism in which the intermediates are ions and the processes are catalyzed by acidic or alkaline substances (Laidler and Meiser, 1982). The formation of colorants by invert alkaline degradation can be expressed as Figure 7, where k_{p1}, k_{p2}, ..., k_{pn} are the rate constants of polymerization and n is the degree of polymerization.
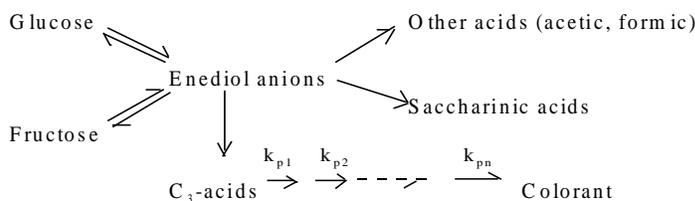

Figure 7. The mechanism of color formation from invert alkaline degradation

3.1 The Effect of Temperature on Color Development in Pure Invert Solution

Color development vs reaction time in alkaline aqueous solution at constant pH and 75, 80 and 85°C is shown in Figure 8. It can be seen that when pH was constant, the color developed faster at higher temperature because the temperature increases accelerated both the formation and the polymerization of small molecular fragments.

It was evident that color development with reaction time followed second-order polynomial relationship and was slow at the beginning of the reaction, though the degradation of invert was fast at the beginning because of

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the high concentration of invert. This implies that there is a propagation period and the formation of color precursors may be the rate-limiting stage during color development as the formation of lactic acid also follows second-order polynomial. This result is consistent with that found by Ramaiah and Kumar (1968) and Eggleston (1996). The quadratic relationship between and absorbance and reaction time is different from the linear relationship obtained by Matula, et al. (1995).
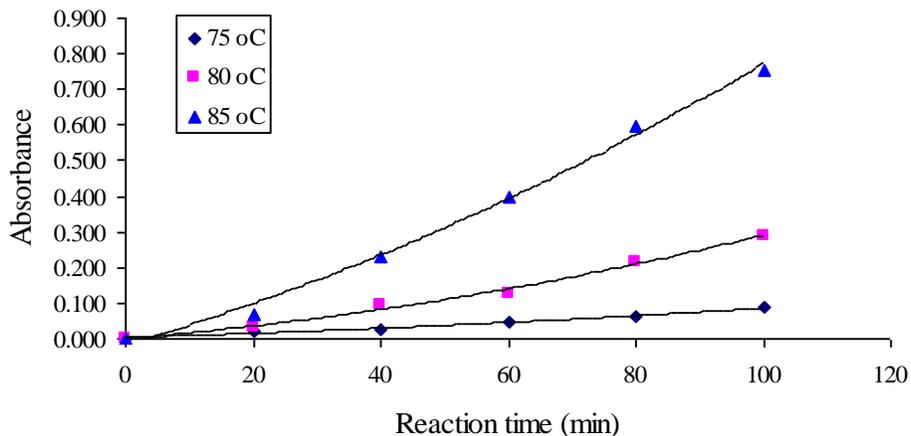

Fig. 8 The effect of temperature on color development (Absorbance was measured at 420 nm)

3.2 The Effect of pH on Color Development in Pure Invert Solution

When same amount of invert degraded at constant temperature, the absorbance of the solution decreased with the increase of pH (Figure 9). There are some possible reasons. 1) At high pH, the high concentration of calcium ions inhibited the formation of color as it was pointed out by Carpenter and Roberts (1975). Although the mechanism of this inhibition is not known yet, the relatively low solubility of inorganic calcium salt might be one of the reasons, which adsorbed part colorants. After filtration through 0.45 μm microfilter, the adsorbed colorants were removed. 2) The time to destroy the same amount of invert was shorter at higher pH, thus the time for color development was reduced. 3) $Ca(OH)_2$ does not completely dissolve in the very diluted invert solution (5,000 mg/L) because the solubility of $Ca(OH)_2$ is very low at high temperature, for example, 0.156 g/100 g water at 20^oC and 0.077 g/100 water at 100^oC (Bubnik, 1995). Undissolved $Ca(OH)_2$ may adsorb some of the colorants formed that were then removed by filtration.

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The increase of absorbance with the invert degradation at each pH can be expressed by the following regression equations

pH 9.0 $Y = 1 \times 10^{-7} x^2 + 3 \times 10^{-5} x - 0.0199$ $R^2 = 0.9915$

pH 10.0 $Y = 1 \times 10^{-7} x^2 - 6 \times 10^{-5} x + 0.0091$ $R^2 = 0.9916$

pH 11.0 $Y = 6 \times 10^{-8} x^2 - 1 \times 10^{-4} x + 0.0734$ $R^2 = 0.9959$

where, x is the invert degraded (mg/L). Since the determination of invert is very time consuming either by the traditional titration or by the modern HPLC technique, and the determination of color is much easier and faster, it is practical to develop an equation to calculate pure invert destruction from the measurement of color under certain conditions.

3.3 The Effects of the Nature of Base on the Formation of Color

It has been found that calcium hydroxide is more effective in accelerating the degradation of invert and the formation of lactic acid than KOH and NaOH (de Bruijn, 1987). On the other hand, Carpenter and Roberts (1975) have reported that calcium ions inhibited color formation at high pH, but enhanced color formation at low pH. They have also found that sodium ions were the reverse of calcium ions, and potassium ions enhanced color formation at pH8 with less color formation at either extreme of pH. Because these conclusions were drawn based on the effects of salts on color formation in alkaline sugar solution, the effects of anions were involved. The study of Ramaiah and Kumar (1968) has shown that anions such as CO_3^{2-} had a catalytic effect on the degradation of invert, whereas Cl^-, SO_4^{2-}, SO_3^{2-}, and PO_4^{3-} exhibited inhibition influences on invert degradation.

To eliminate the effects of anions on the invert degradation and the color formation, calcium hydroxide, sodium hydroxide and potassium hydroxide were used as alkaline agents and the sources of above cations in this study. The experiments were conducted at constant temperature 80^oC, but different pH (pH 10.0 and pH 11.0), i.e. different concentration of cations. The results are shown in Figure 10.

The figure indicates that color development was faster when sodium hydroxide and potassium hydroxide were used than that when calcium hydroxide was used. The effects of NaOH and KOH on color formation are almost identical at pH 11.0. According to Carpenter

(1975), it may be said that calcium ions inhibits the formation of color. The increase in pH did not change this order, and the higher the pH, the higher the concentration of cations, and the greater the inhibition effect of Ca^{2+}. At pH 11.0, the absorbance decreased after 60 minutes reaction with Ca(OH)$_2$. A simple titration indicated that to reach same pH in 5000 mg/L invert solution at 80°C, smaller volume of Ca(OH)$_2$ was needed, thus the lower concentration of Ca^{2+}. Therefore, it can be concluded that Ca^{2+} inhibits the formation of color in invert solution.
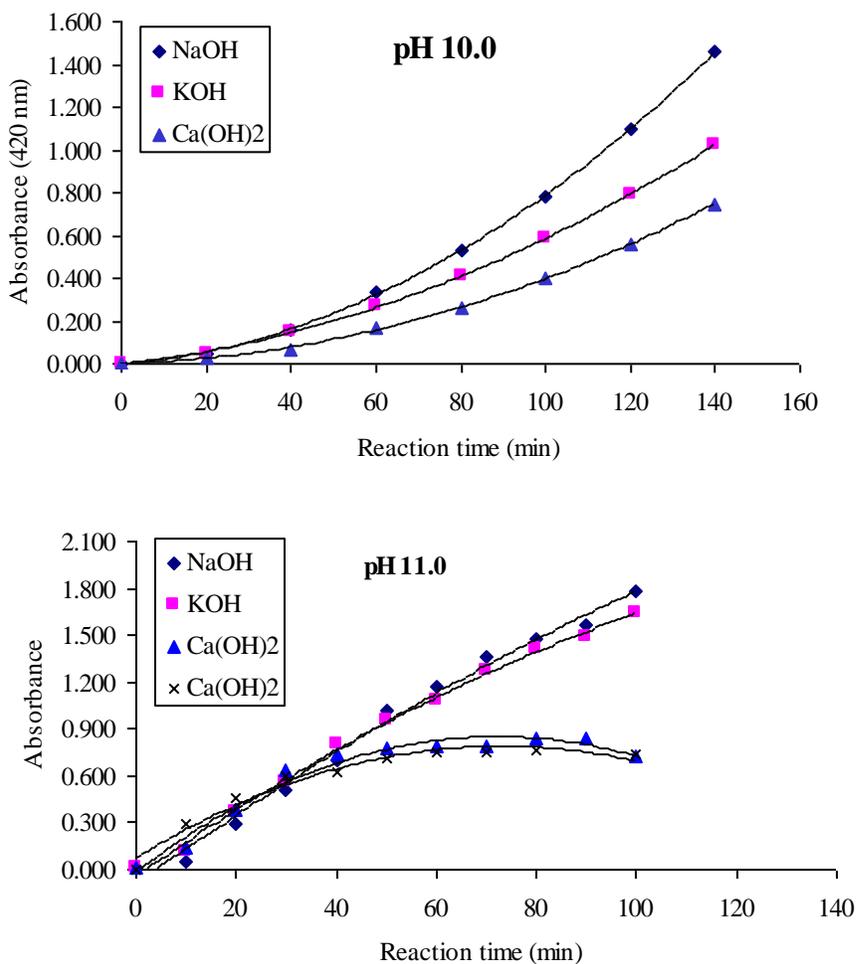
Figure 10. The effect of nature of base on the development of color in 5,000 mg/L invert solution at 80°C.

(4) The Effect of Sucrose on Color Development

In the presence of sucrose, the color of the solution may derive from both invert degradation and sucrose decomposition. A series of experiments were performed at pH 10.5 and

80°C. The results of color development are shown in Figure 11. It can be seen that the development of color was much faster at the presence of sucrose comparing with pure invert solution under the same experiment conditions. This corresponds to the faster degradation of invert, and can be explained by the effect of sucrose on the behavior of pH electrode. It was noted pH reading was lower in the sucrose solutions than in the aqueous solutions when an equal amount of $Ca(OH)_2$ suspension was added. The higher the sucrose concentration, the lower the pH value was. Alkaline decomposition of sucrose is another explanation. According to Eggleston (1996), color formation is directly related to sucrose decomposition.
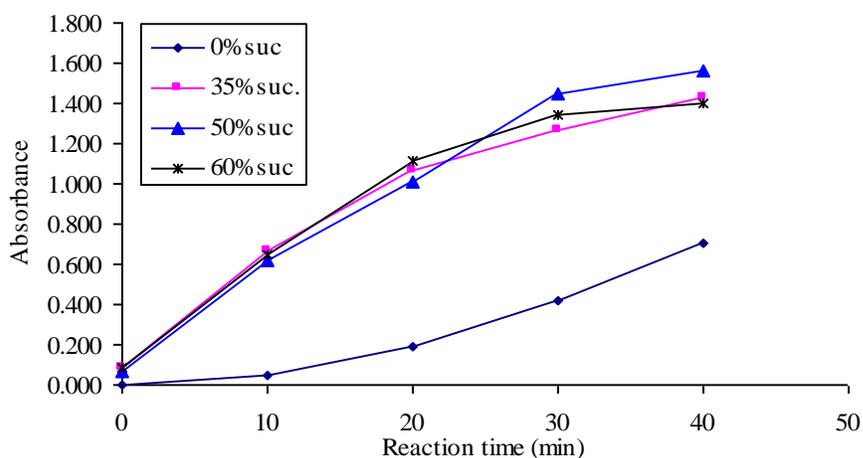

Figure 11 The effect of sucrose on color development of invert solution (5,000 mg/L)
at pH 10.5, 85°C

CONCLUSIONS

Invert degradation in alkaline solution is a multiple step reaction. Enediol ions are the key intermediates and organic acids (also called carboxylic acids) are the final products of invert alkaline degradation.

The first-order reaction kinetics of invert alkaline degradation was demonstrated for the pH range of 9.0-11.0 and temperature range of 70-90ºC. Based on the mechanism of monosaccharide degradation and experimental results, the kinetic model of invert alkaline degradation was further simplified. According to this model, the degradation rates of glucose and

fructose were proportional to the rates of enediol ions formation from glucose and fructose, respectively. Rate constants of invert alkaline degradation determined by the experiments in this study were larger than those calculated by Vukov's equation. After studying the effects of pH and temperature on rate constant and activation energy of invert degradation when $Ca(OH)_2$ suspension was used as base, the equation $\log k = 19.42 - 6712/T - 0.712pOH$ was obtained. The rate constants calculated by this equation were very close to experiment data. According to this equation, rate constant increases 3 to 4 times with a 10^oC increase in temperature at constant pH and 5 to 6 times with one unit increase in pH at constant temperature which is consistent with experimental result and de Bruijn's data.

The activation energy of invert degradation was pH independent when $Ca(OH)_2$ suspension was used as base and was 30.75 kcal/mol in the pH range 9 to 11, temperature 70 to 90^oC. The nature of base did make some differences not only on the rate constant, but also on the activation energy of invert degradation. The order of the effects of Na^+, K^+ and Ca^{2+} was found to be: $Ca^{2+} > Na^+ > K^+$. Sodium ions Na^+ had stronger catalytic effect than potassium ions K^+, calcium ions Ca^{2+} did not show catalytic effect on the degradation of invert because the activation energy of invert degradation did not change due to the increase of Ca^{2+} ions concentration.

The presence of sucrose accelerated the degradation of invert sugar, and the higher the sucrose concentration, the larger the rate constant. The effect of sucrose on the degradation rate was interpreted by the effect of sucrose concentration on the behavior of pH electrode.

Lactic acid is one of the important products of invert alkaline degradation. In the reaction mixture, lactic acid presents in the form of lactate because the salt of lactic acid is water soluble. The relationship between the percentage of lactate produced and the invert degraded was secondary polynomial. The production of lactic acid was influenced by concentration of invert, pH and the nature of base. High pH and low temperature favored the formation of lactic acid. High concentration of invert inhibited the formation of lactic acid. The percentage of lactic acid increased linearly with the increase of pH. Small and highly charged cations such as Na^+ and Ca^{2+} favored the formation lactic acid due to small steric hindrance and strong electrostatic effect to form cation-fructose complex. The effect of the presence of sucrose on the production of lactic acid also could be interpreted by the effect of sucrose on the behavior of pH electrode. The fact that concentration of lactic acid was higher than the original concentration of invert in alkaline

sucrose and invert was still detectable suggested that invert was intermediate of sucrose alkaline decomposition.

The formation of colorant is one of the most important results of invert degradation. The development of color with reaction time through invert alkaline degradation was second order polynomial (quadratic) fit. The correlation between color and invert degraded was second order polynomial fit under constant pH and temperature. The slow development of color at the beginning of reaction implies the presence of propagation period (lag phase) of color formation due to invert alkaline degradation.

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