

# Hypochlorite oxidation of cassava starch

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## Abstract

The chemical and physicochemical properties of cassava starch oxidized by sodium hypochlorite under various reaction pH of 7-11 and the reaction time between 30-300 minutes were studied. Results from the amount of hypochlorite consumed during the course of reaction revealed that the rate of starch oxidation was highest at the reaction pH of 7-8. These conditions also resulted in the highest formation of carbonyl group and the highest rate of viscosity reduction of the modified starch. However, the formation of carboxyl group was most favorable at the reaction pH of 8-9. The changes in viscosity of starch paste during storage at 50°C for 8 hours were also investigated. The results demonstrated that reaction pH drastically influenced the stability of starch paste especially for the samples prepared with the shorter reaction time (less than 120 minutes). It was found that modified starch prepared under the reaction pH of 10 and 11 showed an increase in the paste viscosity or even formed a gel during storage which indicated that the tendency for starch retrogradation prevailed in these samples. However, oxidized starch obtained under the reaction pH of 8 and 9 exhibited a dramatic decrease in the paste viscosity during storage, suggesting that these modification conditions might result in oxidized starch with less stable molecular structure, thus degradation of starch molecules under aqueous suspension could occur.

## Introduction

Oxidized starch is widely used in both food and nonfood industries where film formation and adhesion properties are desired. The major application of oxidized starch is as a surface sizing agent and a coating binder in paper industry. Oxidized starch is produced by reaction of starch with an oxidizing agent under controlled temperature and pH. Several oxidizing agents have been used; however, hypochlorite is the most common chemical used for the production of oxidized starch in an industrial scale. During the course of reaction, several reactions occur which lead to the introduction of carbonyl and carboxyl groups and the degradation of starch molecules. Hence, oxidized starch exhibits low viscosity due to depolymerization and improved stability of starch dispersion from the presence of functional groups.

Several investigations on hypochlorite oxidation of amylose, amylopectin and native starches from various origins have been reported. It has been claimed that oxidation occurred mainly in the amorphous regions since no change in the x-ray patterns and their intensity was observed in the oxidized products (Schmorak and Lewin, 1963; Kuakpetoon and Wang, 2001). Hypochlorite oxidation has been reported to depend mainly on the pH during the reaction. Whistler and Schweiger (1957) demonstrated that hypochlorite oxidation of corn amylopectin was most rapid at neutral pH while the reaction rate decreased with increasing acidity and alkalinity. Similar results were observed on wheat and corn starches (Schmorak et al., 1961; Patel et al., 1974). The type and amount of functional groups formed in the starch molecule depend on the reaction pH as well. The formation of carbonyl group was found to be higher under acidic conditions while the amount of carboxyl group increased with increasing pH (Hullinger and Whistler, 1951; Schmorak and Lewin, 1963).

Beside the reaction pH, other factors such as oxidant concentration, temperature and starch origin are known to influence hypochlorite oxidation. It has been shown that different levels of sodium hypochlorite used in the reaction yielded oxidized products with different pasting properties. The slightly oxidized starches ( $\leq 1.0\%$  chlorine for common corn starch and  $\leq 0.75\%$  chlorine for waxy corn starch) exhibited pasting profiles similar to those of slightly crosslinked starches. At higher levels of oxidant, the modified starch showed lower viscosity which is typical of commercial oxidized starch (Wang and Wang, 2003). The botanical origin of the starch used in the modification

process is of considerable importance to the reaction and properties of the resulting oxidized starch. It has been shown that tuber starch was more readily oxidized than the cereal starch. Study on the oxidation on potato, corn and rice starch under the same reaction conditions has shown that potato starch had the highest carboxyl content while rice and corn starch possessed lower values (Kuakpetoon and Wang, 2003). Potato starch was also found to be more easily oxidized than barley starch (Forsell et al., 1995).

Currently, for industrial practice, hypochlorite oxidation of starch is performed under conditions of mild to moderate alkalinity in order to favor production of carboxyl group which is a key component in stabilizing the viscosity of starch dispersion and minimizing retrogradation (Wurzburg, 1986). A range of reaction pH from 8 to 11 could be encountered in different starch modification plants. As mentioned above, reaction pH is a key factor in determining the course of hypochlorite oxidation resulting in different reaction rate and products with different chemical structures. Although several studies have been done to demonstrate the influence of reaction pH on hypochlorite oxidation, most of them performed under more drastic conditions than those normally used in the industrial practice; that was very low starch concentration and relatively high levels of oxidant. Furthermore, the studies on rate of reaction were done based on the rate of disappearance of oxidant which might not be relevant to the changes occurred to the starch molecules; and the products determined in these studies were obtained from relatively high degree of oxidation which might not represent what really encounters in the commercial products.

Relatively few data available in the literature on the physicochemical and functional properties of oxidized starch as influenced by reaction pH. The aim of this study was to determine the effect of reaction conditions (pH and time) on physicochemical properties of cassava starch. The results could provide information used to improve the manufacturing process as well as the product properties for some certain applications.

## **Materials and Methods**

### ***Preparation of oxidized starch***

A cassava starch slurry containing 40% dry solid was prepared and the pH was adjusted to 7, 8, 9, 10, and 11 with sodium hydroxide solution. The temperature of the slurry was brought to 30°C and sodium hypochlorite (3% active chlorine based on starch) was added dropwise over a period of 20 min with stirring. During the addition of the reagent and the course of reaction, the pH of the slurry was maintained at the desired value with sodium hydroxide or hydrochloric solution. Then the mixture was stirred under the defined conditions and the sample was then collected at 30, 60, 120 and 300 min later. The reaction in collected samples was terminated by addition of sodium bisulphite. The sample was then filtered and suspended in distilled water and the pH was adjusted to 6.5-7.0. The sample was then filtered and thoroughly washed and oven dried at 50°C.

The decrease in hypochlorite concentration due to oxidation of starch was also determined from the difference between the concentration of blank solution and the solution containing the starch at the time of sample removal from the reaction mixture. Hypochlorite concentration was determined by sodium thiosulfate titration of acidified aliquots containing excess potassium iodide (Hebeish et al., 1989).

### ***Determination of carbonyl content***

The carbonyl content was determined as described by Kuakpetoon and Wang (2001). Starch sample (4 g) was slurried in 100 mL of distilled water. The slurry was gelatinized in a boiling water bath for 20 min, cooled to 40°C, adjusted to pH 3.2 with 0.1 M HCl, and 15 mL of hydroxylamine reagent was added. The flask was stoppered and agitated in a water bath at 40°C. After 4 hour, the sample was rapidly titrated to pH 3.2 with 0.1 M HCl. The hydroxylamine reagent was prepared by dissolving 25 g hydroxylamine hydrochloride in 100 mL of 0.5 N NaOH. The final volume was then adjusted to 500 mL with distilled water.

### ***Determination of carboxyl content***

The carboxyl content of hypochlorite-modified cassava starch was determined by the ISO method (ISO-11214, 1996). Starch sample (5 g) was stirred in 0.1 M HCl for 30 min. The slurry was then filtered and washed with distilled water until free of chloride ions. The filtered cake was transferred to 300 mL water and the starch slurry was heated in a boiling water bath with continuous stirring until gelatinized and continue stirring at that temperature for another 15 min. The hot sample was titrated with 0.1 M NaOH using phenolphthalein as an indicator.

### ***Viscosity and viscosity stability of starch dispersion***

A suspension of starch (15%, w/w) in distilled water was heated in a water bath at 95°C for 15 minutes with continuous stirring in the first 5 minutes. The starch paste was then maintained at 50°C in a temperature-controlled water bath. When a temperature of starch paste reach 50°C, the initial viscosity of the sample was measured with a rotational Physica MCR 300 rheometer (Physica Messtechnik GmbH, Stuttgart, Germany) using a concentric cylinder or a double gap cylindrical geometry. The temperature was regulated by a Paar Physica circulating bath and a controlled peltier system (TEZ 150P/MCR) with an accuracy of  $\pm 0.1^\circ\text{C}$ . Measurements were performed in the shear rate range 1-500  $\text{s}^{-1}$ . The viscosity value at 20  $\text{s}^{-1}$  was used for comparison between different samples. The viscosity stability of starch paste was determined by measuring the viscosity of the sample after being maintained at 50°C for 1, 2, 4 and 8 hours.

### ***Molecular weight distribution***

The molecular weight distribution was determined by High Performance Size Exclusion Chromatography (HPSEC) using one Ultrahydrogel linear and two Ultrahydrogel 120 columns connected in series (Waters Corporation, MS), and columns were maintained at 55°C with a column heater. The mobile phase was an aqueous solution of 0.1 M NaCl with a flow rate of 0.3mL/min.

## **Results and Discussion**

The reduction of available chlorine content in the reaction medium is a preliminary measure of the course of the oxidation process. In the present study, cassava starch at 40% concentration in water was treated with sodium hypochlorite (3% active chlorine based on starch) at various pHs for different period of time from 30 to 300 min. The extent and rate of hypochlorite oxidation are shown in Figure 1. In general, the rate of oxidant consumption decreased with increasing pH. For oxidation conducted at pH 7 and 8, the disappearance of oxidant was rapid. After 60 min, less than 0.1% of oxidant was found in the reaction mixture. At pH 9, the rate of oxidant consumption was intermediate; after 120 min of reaction time about 1% of oxidant was remained. The rate was much slower when the reaction was conducted at pH 10 and 11. The results was in agreement with the studies reported on other starches (Whistler and Schweiger, 1957; Schmorak et al., 1961).

Carbonyl and carboxyl contents of oxidized starch are persented in Figure 2 and 3. Both carbonyl and carboxyl contents of all oxidized samples were hgiher than those of the native starch. The highest formation of carbonyl group was found at pH 7 and decreased as the reaction pH increased, which agrees with previous reports. Reaction time seemed to have no effect on the amount of carbonyl group; irrespective of reaction pH, carbonyl contents remained constant after only 30 min of reaction time. The maximum amount of carboxyl groups was obtained at the reaction pH 8 and 9; at these reaction pHs, the rate of carboxyl group formation was very fast, it remained constant since only 30 min of reaction time. On the other hand, at pH 10 and 11 the amount of carboxyl group gradually increased with reaction time.

The viscosity of oxidized starch modified under various reaction pHs and time is shown in Figure 4. Data on native starch are not presented since the experimental design was not appropriate for native starch due to the high viscosity (starch paste with 15% solid content was used for viscosity

determination). In general, longer reaction time yielded starch with lower viscosity. Oxidation conducted at pH 7 gave starch with the lowest viscosity indicating that oxidative degradation of starch molecule would prevail at this pH. At pH 8, 10 and 11, the oxidation yielded starch with intermediate viscosity values while starch oxidized at pH 9 exhibited the highest viscosity implying that depolymerization would take place at a much slower rate.

The HPSEC chromatograms of native and oxidized cassava starches are shown in Figure 5. The first fraction of the native cassava starch with a shorter retention time consisted of high molecular weight component which was presumed to be amylopectin, and the second fraction appeared as a shoulder of the first peak composed of lower molecular weight component, presumably amylose. Starch oxidized at pH 7 and 8 exhibited the most reduction in the starch molecular size whereas starch oxidized at pH 9 showed the least molecular degradation compared to native starch; which agreed with the results on viscosity of starch dispersion. The reaction pH at 10 and 11 showed intermediate results; depolymerization seemed to be lower than at pH 7 and 8 but higher than at pH 9. For starch modified at pH 7, 8 and 9, the reaction time seemed to have no effect on the HPSEC profiles. However, at pH 10 and 11, the reaction time showed significant influence on chromatogram patterns; the chromatograms of starch oxidized with the longer reaction time shifted to a slightly longer retention time and contained higher proportion of lower molecular weight fraction.

In addition to low viscosity, improved stability of starch dispersion is another main characteristic desired from the oxidation of starch. In general, a dispersion of native starch is prone to intermolecular association or retrogradation showing an increase of paste viscosity during storage. The introduction of functional group, especially carboxyl group, has been reported to minimize retrogradation resulting in improves stability of starch dispersion. In the present study, the change of the viscosity of starch dispersion during storage at 50°C was followed for 8 hr and the results are presented in Table 1. Oxidized starch obtained at reaction pH 7 showed the lowest initial viscosity and the best stability of paste viscosity among all starches tested. Unexpectedly, a considerable reduction in viscosity of stored paste was observed in the oxidized starch modified at pH 9. The viscosity of starch paste after 8 hr storage was dropped to only 16, 22 and 75% of the initial viscosity for starch modified with a reaction time of 30, 60 and 120 min. The stability of starch paste was much improved when starch was modified with 300 min. Similar observations were also found in the oxidized starch obtained with the reaction pH at 8, but with a lesser extent. The stability was also improved with prolonged reaction time.

A reduction in viscosity of stored paste was not observed for starch oxidized at pH 10 and 11. Contrarily, the starch paste showed an increase in viscosity with increasing storage time, especially for starch modified with a shorter reaction time. For instance, after 8 hr storage, starch oxidized at pH 10 for 30 min showed an increase in viscosity to about 30% of the initial viscosity. The extent was more pronounced for starch oxidized at pH 11 where a dispersion of the sample with 30 min reaction time set a soft gel after storage at 50°C for only 4 hr. An increase in the viscosity of the stored paste in these samples could be attributed to the intermolecular association of starch molecules. When the degree of oxidation was low, the molecular weight was still high and relatively few carboxyl groups were introduced to the molecules, starch had a higher tendency for intermolecular reassociation; the paste viscosity during storage thus increased. Viscosity of starch paste became more stable when the degree of hydrolysis and the carboxyl content increased, as seen in the oxidized starch obtained at a longer reaction time.

A reduction in the viscosity of stored paste observed for starch oxidized at pH 8 and 9 was considered to be attributed to depolymerization of starch molecules. The fact that such behavior was not observed for samples modified at other pHs implied that oxidation at various pHs might yield oxidized products with different molecular structures. Modification at pH 8 and 9 might lead to oxidized starch with molecular structure that is more labile to chemical degradation resulting in depolymerization of starch molecules and decreased viscosity of starch dispersion during storage. The transformation of dialdehyde starch in aqueous dispersion under slightly acidic and neutral conditions causing a reduction in dispersion viscosity and the lower molecular weight of starch has

been reported (Veelaert et al., 1997). These authors explained that the structure of dialdehyde starch with two aldehyde groups on C<sub>2</sub> and C<sub>3</sub> carbons of glucose unit and the glycol bond between these 2 carbons cleaved was susceptible to  $\beta$ -elimination leading to depolymerization of starch molecules. The oxidized starch obtained at pH 8 and 9 in the present study might possess similar structure as in dialdehyde starch; that is these conditions might favor the formation of dicarboxyl groups on C<sub>2</sub> and C<sub>3</sub> carbons leading to molecular structure that is more readily hydrolyzed. This speculation is further supported by a study of Whistler et al. (1956). This group of researchers studied the action of hypochlorite at pH 9 and 11 on corn amylose and found that hydrolysis products of oxidized amylose obtained at pH 9 contained large amount of D-erythronic acid and glyoxylic acid. The appearance of these complementary 2- and 4-carbon fragments suggests that extensive oxidation occurred at carbon atoms C<sub>2</sub> and C<sub>3</sub> of anhydro-glucose units. They suggested that at pH 9 the oxidation action of hypochlorite did not random but was extensively specific for positions C<sub>2</sub> and C<sub>3</sub> causing bond cleavage between these two carbons and carboxyl groups were formed at positions C<sub>2</sub> and C<sub>3</sub>. The presence of carbonyl group at C<sub>2</sub> or C<sub>3</sub> of sugar units would weaken the bond at C<sub>1</sub> position; thus, depolymerization might be expected.

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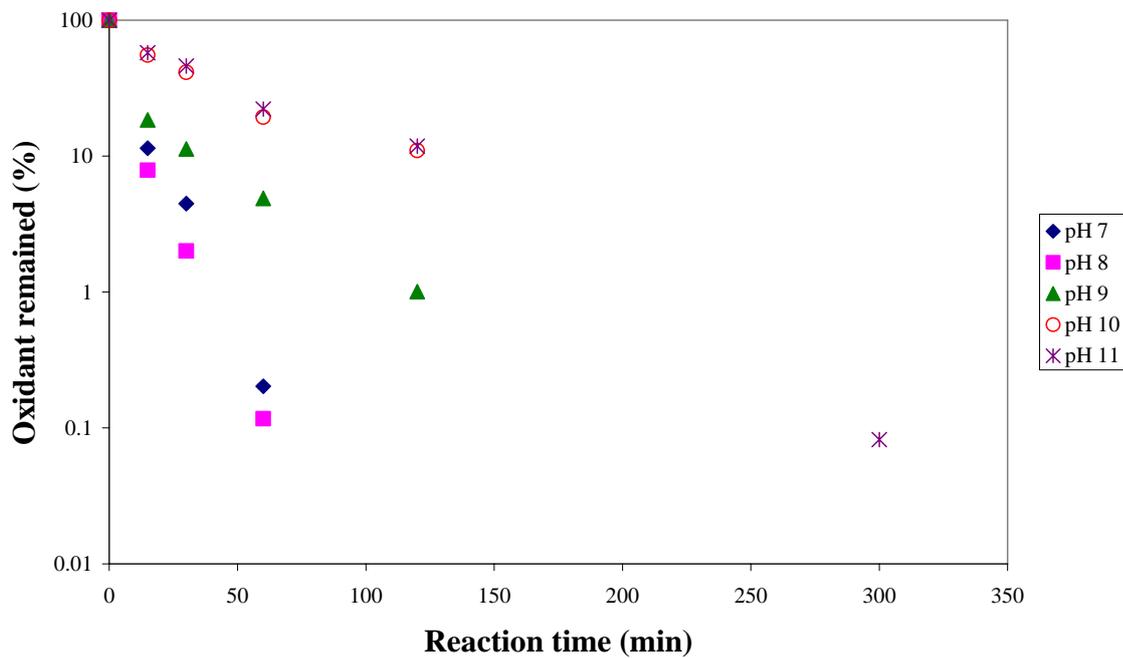


Figure 1 Hypochlorite consumption during oxidation of cassava starch

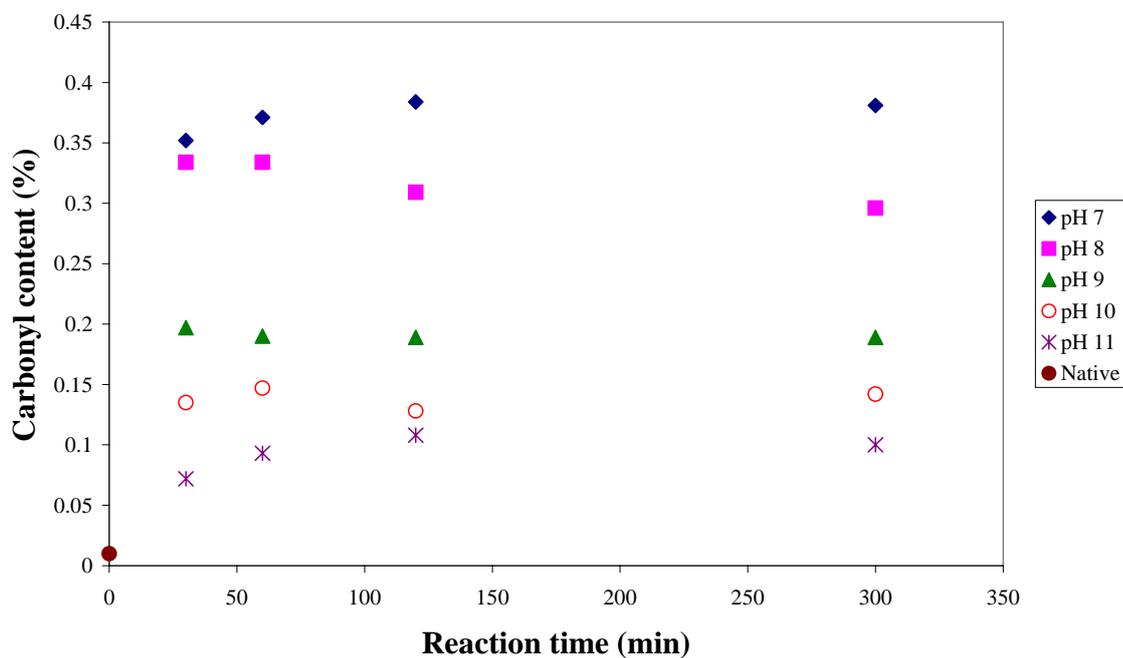


Figure 2 Carbonyl content of cassava starch oxidized by hypochlorite under various reaction pH and time

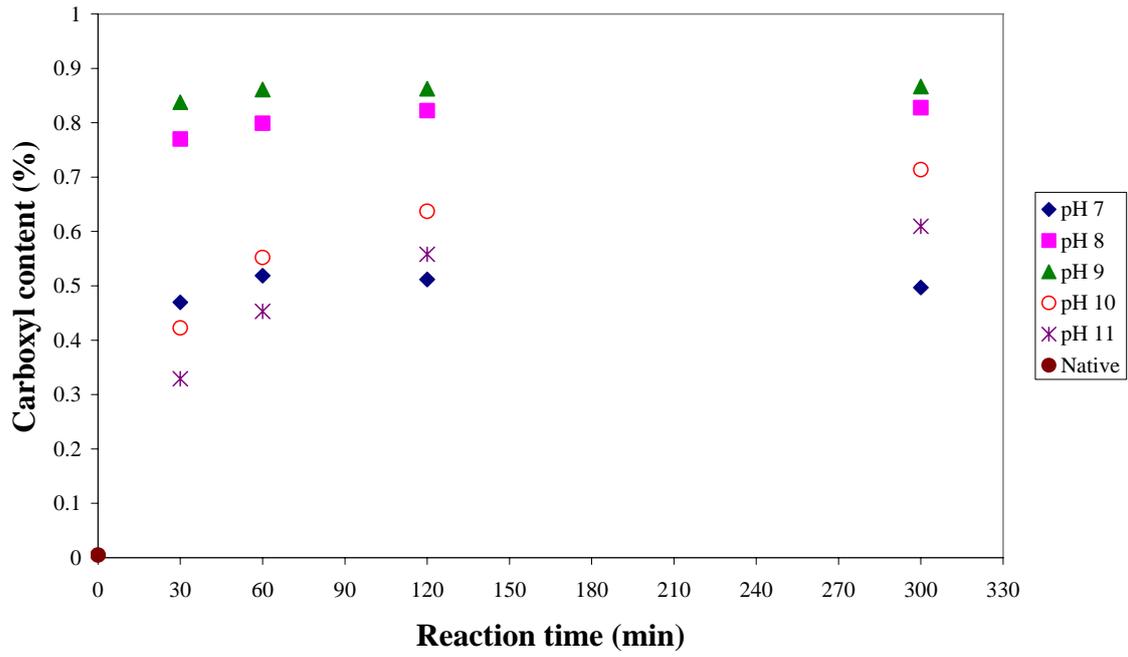


Figure 3 Carboxyl content of cassava starch oxidized by hypochlorite under various reaction pH and time

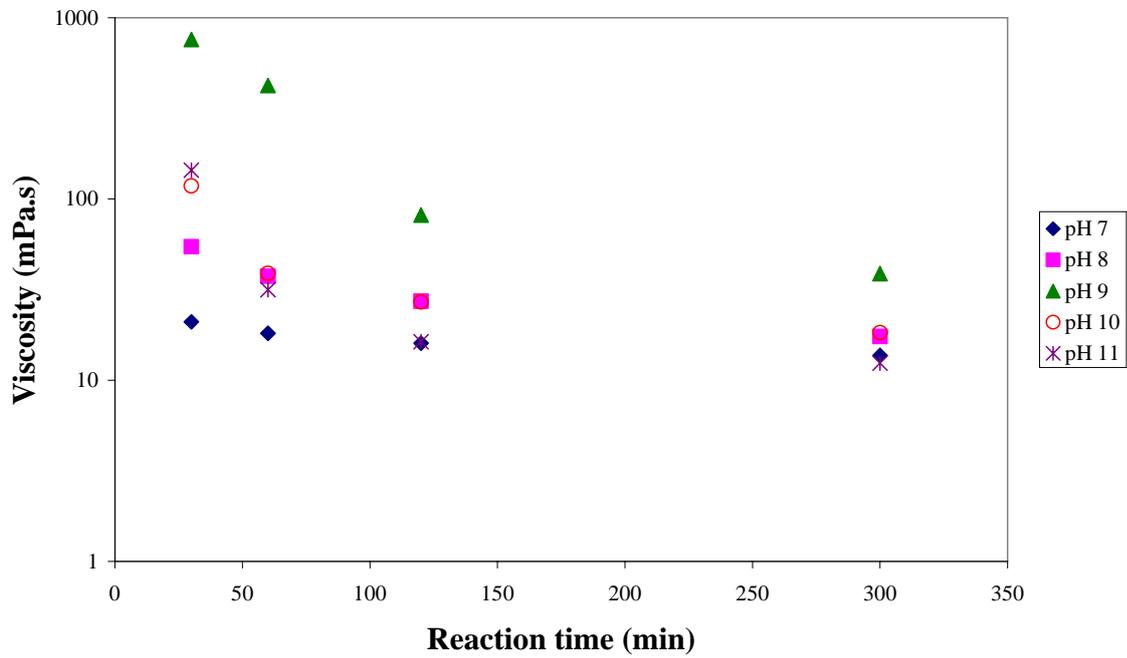


Figure 4 Viscosity (at shear rate  $20 \text{ s}^{-1}$ ) of cassava starch oxidized by hypochlorite under various reaction pH and time

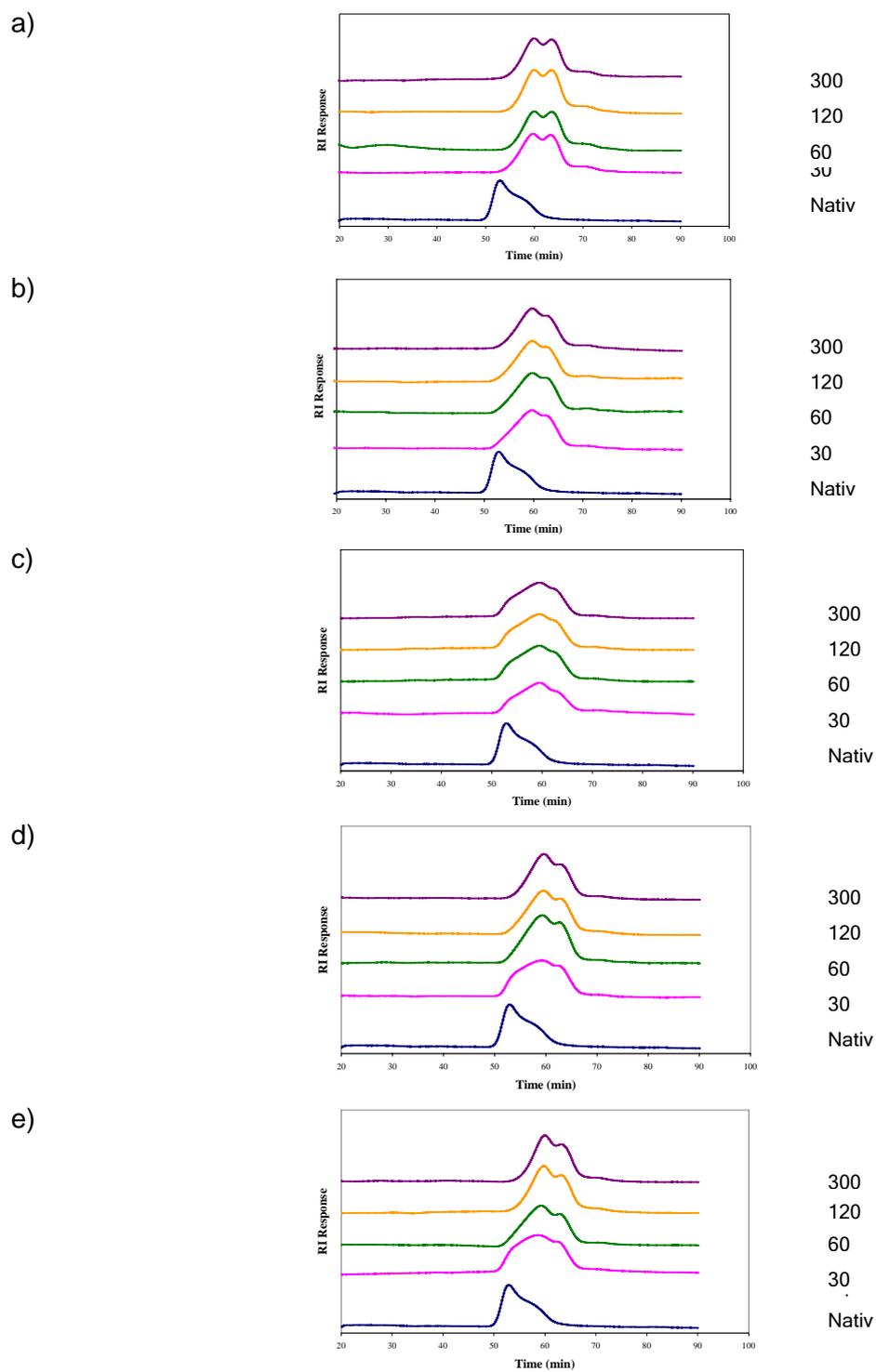


Figure 5 Molecular weight distribution of cassava starch oxidized by hypochlorite under various reaction pH and time. a) pH 7; b) pH 8; c) pH 9; d) pH 10; e) pH 11

Table 1 Viscosity (mPa.s; at shear rate  $20 \text{ s}^{-1}$ ) of dispersion of hypochlorite oxidized cassava starch during storage at  $50^\circ\text{C}$  for 8 hr

Reaction pH	Reaction time (min)	Storage time (hr)				
		0	1	2	4	8
7	30	21	20	20.6	19.8	18.5
	60	18.2	18.1	18.2	17.7	16.1
	120	16	16	16	15.8	14.6
	300	13.7	13.9	13.9	13.6	13.3
8	30	54.5	46.8	44.4	38.8	33.8
	60	37.5	34.4	32.8	30.5	28.3
	120	27.3	26.6	26.2	25.3	23.4
	300	17.4	18.3	18.1	17.8	17.2
9	30	758	473	286	215	118
	60	422	263	217	107	93
	120	81.5	75	71.9	69.3	61.1
	300	38.7	39.9	37.8	38	37.7
10	30	118	121	123	137	154
	60	38.8	41.3	42.6	43.6	43.6
	120	27	26.8	26.8	27.7	27.2
	300	18.3	19	19	18.8	19.1
11	30	144	640	1189	weak gel	weak gel
	60	31.5	34.6	38.8	43.8	52.4
	120	16.3	16.5	16.8	17.2	18.1
	300	12.4	12.7	13.1	13.1	13.5