EFFECT OF ACETYLATION AND CARBOXYLATION ON SOME PHYSICOCHEMICAL PROPERTIES OF CASSAVA STARCHES

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ABSTRACT
Starch from five cassava varieties; 97/4414, 97/3982, 97/4489, 97/4962, and Afisiafi was modified with acetic anhydride and sodium hypochlorite solutions. The study was carried out to determine the effect of acetylation and carboxylation on some physicochemical properties of the native starches. The physicochemical properties studied were moisture content, pH, solubility, swelling power, granule size and water binding capacity. The moisture content, pH, water binding capacity, and swelling power of the native starches were generally higher than those of the modified starches. Native 97/4414 had the highest moisture content of 11.31% and acetylated 97/3982, the lowest value of 7.21%. The pH ranged from 4.23 for carboxylated 97/4414 to 6.96 for native 97/3982. The solubility of the carboxylated starches was comparatively lower than those of the native and acetylated starches. The native 97/4414 had the lowest solubility of 11.77% whereas acetylated 97/4414 and Afisiasi, the highest value of 31.91%. The native Afisiafi had the highest swelling power of 40.42% and carboxylated 97/4489 the lowest value of 21.58%. The water binding capacity ranged from 93.79% for native 97/4414 to 51.46% for 97/4489. The granule size ranged between 4.33µm and 6.67µm. Statistical analysis revealed significant effect ($p < 0.05$) of acetylation and carboxylation on the physicochemical properties of the native starch.

INTRODUCTION
Starches are made from many different starchy raw materials, such as wheat, barley, maize, sweet potato and cassava. Although they have similar chemical reactions and are usually interchangeable, starches from different sources have different granular structures that affect their physical properties. Starch functionality depends on the average molecular weight of amylose and amylopectin, as well as of their molecular organization within the granule. Native starches are used in foods because they regulate and stabilize the texture and because of their good thickening and gelling properties (Cousidine, 1982). Native (unmodified) starches are generally not very useful for many applications. This is because the native starches are unstable and tend to retrogradate after gelatinization. The native starch granules take up water very easily, swell rapidly, rupture, lose viscosity and produce stringy and cohesive pastes. These characteristics are not very useful

in food or other products. Native starches are also not very stable to acids, high temperatures or mechanical action. These shortcomings can be overcome by modifying starch native structure by chemical, physical and enzymatic methods (Fleche, 1984).

Modifications may involve altering the form of the granules or changing the shape and composition of the constituent amylose and amylopectin molecules. Modifications are therefore carried out on the native starches to confer them with properties needed for specific uses. Modified starches usually show better paste clarity and stability, smaller tendency to retrogradation and increase in freeze-thaw stability (Agboola *et al.*, 1991).

The increasing industrial and food applications of modified starches and the production of newly improved varieties of cassava require study into the physicochemical properties of their native structure and that of the modified starches. Thus the objective of this research was to determine the effect of acetylation and carboxylation on some physicochemical properties of cassava starch.

MATERIALS AND METHODS

Source of cassava varieties

Four new varieties of cassava coded 97/4414, 97/4489, 97/3982, and 97/4962 and already existing variety Afisiafi were obtained from the Crop Research Institute, Fumesua, Kumasi, Ghana.

Starch extraction

The extraction of starch was carried out using the wet extraction method described by Barimah *et al., (*1999).

Modification of starch samples

The starch samples were modified with acetic anhydride (acetylation) and sodium hypochlorite (carboxylation) based on the modification of the method adopted by Paton (1974).

For the acetylation of the starch, five hundred grams of each starch sample were made into slurry using 5 l of distilled water. Fifty millilitres (50 ml) of 10% v/v of acetic anhydride solution was added to the starch slurry dropwise over a period of two hours while a Griffin stirrer at speed ranging between 25-35 rpm continuously agitated the starch slurry. The temperature of the slurry was maintained at 45° C and the pH between 8-10 using 3% sodium hydroxide solution. The reaction was stopped by addition of 0.5 M HCl, which adjusted the pH to 6.5. The slurry was allowed to sediment for about 6 hrs and the supernatant liquid decanted. The starch was washed with several changes of distilled water until all the chlorides were removed. This was confirmed by testing with 0.1 M $AgNO_3$. The acetylated starches were dried in a solar tent dryer. The dried starches were stored in air tight-containers for analysis.

For the carboxylation of starch, 0% (w/v) sodium hypochlorite solution was added dropwise into 10% (w/v) starch slurry held at room temperature. The slurry was agitated at a constant speed of 25-35 rpm for 4 hrs and pH was maintained at 9-10 with 3% sodium hydroxide solution. The reaction was halted with 0.5 M HCl which adjusted the pH to 6.5. The slurries were allowed to incubate for 6 hrs after addition of antichlor ($NaHSO_3$) which destroyed excess chlorine. The sedimented starch pastes were filtered and then washed several times with distilled water until washings were free from chlorine. The wet starch cakes were dried in solar tent dryer and stored in air-tight containers for analysis.

Analysis of starch samples

The moisture content of the starch samples was determined using official method of AOAC (1990) .The pH was determined directly using Corning pH meter (model 240). The solubility and swelling power were determined based on the method of Leach *et al.* (1959). The starch granule size was determined by light microscopy using the method described by MacMaster (1964). The water binding capacity was determined by the method of Medcalf and Gilles (1965).

Statistical analysis

The data obtained were analyzed for significant treatment effect and variety effect and also least significant difference calculated using Costat Version 6.024. The significance was established at $p \leq 0.05$.

RESULTS AND DISCUSSION

The physicochemical properties of the native, acetylated and carboxylated starches are presented in Tables 1-6. The moisture content of the starch samples ranged between 7.21% for acetylated 97/3982 and 11.31% for native 97/4414. Generally, all the starch samples had low moisture content indicative of better shelf life. There were significant differences ($p < 0.05$) in the moisture content of the starch samples. The carboxylated 97/4414 had the lowest pH of 4.23 and native 97/3982 the highest value of 6.96. The pH of the native starch samples was within the standard pH specification for native starch which ranges from 4.5-7.0 (Ingram, 1975). According to Sriroth (1998), the pH values of good quality native starch

Table 1: Moisture content of native, acetylated and carboxylated cassava starches

Cassava varieties	Native	Acetylated	Carboxylated	LSD (0.05)
97/4414	11.31_a^1 (0.02)	8.80_b^2 (0.05)	8.53_c^3 (0.05)	0.19
97/3982	9.73_a^4 (0.03)	7.21_c^4 (0.16)	9.27_b^2 (0.03)	0.43
97/4489	9.73_a^4 (0.04)	8.27_c^3 (0.12)	8.64_b^3 (0.06)	0.36
97/4962	10.69_a^2 (0.04)	8.07_c^3 (0.03)	9.30_b^2 (0.03)	0.12
Afisiafi	10.40_a^3 (0.05)	10.21_a^1 (0.04)	9.89_b^1 (0.06)	0.23
LSD (0.05)	0.12	0.34	0.17	

a - c values within each row with the same subscripts are not significantly different (p>0.05)
1- 4 values in the same column with the same superscripts are not significantly different (p>0.05)
( ) values in brackets are standard deviations of the mean

Table 2: pH of native, acetylated and carboxylated cassava starches

Cassava varieties	Native	Acetylated	Carboxylated	LSD (0.05)
97/4414	5.42_a^5 (0.02)	4.41_b^2 (0.02)	4.23_c^3 (0.04)	0.13
97/3982	6.96_a^1 (0.02)	4.43_b^2 (0.03)	4.25_c^3 (0.03)	0.12
97/4489	5.53_a^4 (0.05)	4.27_c^3 (0.03)	4.49_b^2 (0.02)	0.16
97/4962	5.86_a2 ((0.02)	4.29_b^3 (0.04)	$4{,}31_b^3$ (0.02)	0.12
Afisiafi	5.75_a^3 (0.03)	4.62_c^1 (0.02)	4.88_b^1 (0.03)	0.12
LSD (0.05)	0.11	0.11	0.11	

a - c values within each row with the same subscripts are not significantly different (p>0.05)
1- 4 values in the same column with the same superscripts are not significantly different (p>0.05)
( ) values in brackets are standard deviations of the mean

range between 5 and 7, and upon acetylation the pH is reduced to 4.7. However, the pH of the acetylated as well as the carboxylated starches was slightly lower than the reported value (4.7). This may be due to different varieties of cassava used and experimental conditions.

The swelling power of the acetylated and carboxylated starches was generally lower than that of native starch. Native Afisiafi gave the highest swelling power (40.42 g/g) whilst 97/4489 the lowest (21.58 g/g). The higher swelling power of the native starch compared to that of the modified starches may be attributed to the higher proportions of hydroxyl groups in the native starch which allow it to

Table 3: Swelling Power of native, acetylated and carboxylated cassava starches

Cassava varieties	Native	Acetylated	Carboxylated	LSD (0.05)
97/4414	$37.42_{a}^{1,2}$ (1.19)	32.02_{a}^{1} (1.94)	25.20_{b}^{1} (0.60)	6.11
97/3982	$36.72_{a}^{1,2}$ (1.15)	28.45_{b}^{2} (0.56)	$23.46_{b}^{1,2}$ (1.16)	5.24
97/4489	$37.20_{a}^{1,2}$ (1.51)	33.91^{a1} (0.86)	21.58_{b}^{2} (0.34)	4.60
97/4962	34.91_{a}^{2} (0.81)	29.42_{b}^{2} (0.59)	$23.44_{c}^{1,2}$ (0.46)	2.87
Afisiafi	40.42_{a}^{1} (0.69)	33.23_{b}^{1} (0.27)	24.22_{c}^{1} (0.53)	2.37
LSD (0.05)	4.37	3.72	2.47	

a - c values within each row with the same subscripts are not significantly different (p>0.05)
1- 4 values in the same column with the same superscripts are not significantly different (p>0.05)
( ) values in brackets are standard deviations of the mean

Table 4: Solubility of native, acetylated and carboxylated cassava starches

Cassava varieties	Native	Acetylated	Carboxylated	LSD (0.05)
97/4414	11.77_{c}^{4} ((0.25)	31.91_{a}^{1} (0.19)	17.37_{b}^{3} (0.05)	0.83
97/3982	25.68_{a}^{1} (0.71)	25.98_{a}^{3} (0.91)	25.28_{a}^{1} (0.29)	3.09
97/4489	20.98_{b}^{2} (0.23)	28.66_{a}^{2} (0.29)	15.62_{c}^{4} (0.31)	1.25
97/4962	17.41_{b}^{3} (0.07)	31.41_{a}^{1} (0.21)	17.18_{b}^{3} (0.21)	0.79
Afisiafi	26.57_{b}^{1} (0.07)	31.91_{a}^{1} (0.19)	19.97_{c}^{2} (0.04)	0.93
LSD (0.05)	1.37	1.65	0.78	

a - c values within each row with the same subscripts are not significantly different (p>0.05)
1- 4 values in the same column with the same superscripts are not significantly different (p>0.05)
( ) values in brackets are standard deviations of the mean

bind more water and swell. Oxidative treatment of starch has been found to decrease the swelling power of the native starch (Leach, 1959). Hashim *et al.*, (1992) also observed that acetic anhydride treatment of the native starch decreased its swelling power. The solubility of the acetylated starch was generally higher than that of the caboxylated and native starches. Acetylated 97/4414 and Afisiafi had the highest solubility (31.91%) whereas native 97/4414 the lowest (11.77%). The introduction of acetyl groups into the native starch structure weakened the associative forces (hydrogen bonding) between the molecules in the starch granules resulting in higher solubility of the acetylated starch. There were significant differences ($p<0.05$) in the swelling power as well as the solubility of the starch samples.

The water binding capacity (WBC) of the starch samples was generally high ranging between 51.46% and 93.79% with acetylated

Table 5: Granule size of native, acetylated and carboxylated cassava starches

Cassava varieties	Native	Acetylated	Carboxylated	LSD (0.05)
974414	5.67_a^{2} (0.02)	5.33_b^{2} (0.01)	4.80_c^{2} (0.15)	0.14
97/3982	5.00_a^{3} (0.08)	5.33_a^{2} (0.04)	4.33_b^{3} (0.02)	0.48
97/4489	6.33_a^{1} (0.21)	5.00_b^{3} (0.04)	4.33_c^{3} (0.01)	0.56
97/4982	$5.33_a^{2,3}$ (0.19)	5.33_a^{2} (0.02)	5.33_a^{1} (0.01)	0.50
Afisiafi	$5.33_a^{2,3}$ (0.17)	6.67_a^{1} (0.04)	5.33_b^{1} (0.03)	0.46
LSD (0.05)	0.16	0.12	0.10	

a - c values within each row with the same subscripts are not significantly different (p>0.05)
1- 4 values in the same column with the same superscripts are not significantly different (p>0.05)
( ) values in brackets are standard deviations of the mean

Table 6: Water binding capacity of native, acetylated and carboxylated cassava starches

Cassava varieties	Native	Acetylated	Carboxylated	LSD (0.05)
974414	93.79_a^{1} (1.44)	80.29_b^{1} (2.30)	69.60_c^{2} (0.82)	6.76
97/3982	$89.92_a^{1,2}$ (1.73)	59.98_c^{4} (0.73)	77.17_b^{1} (1.88)	9.83
97/4489	82.04_a^{2} (1.73)	51.46_b^{5} (0.73)	59.40_b^{3} (1.32)	12.90
97/4982	70.85_a^{3}	74.61_a^{2} (1.32)	75.40_a^{1} (0.78)	7.25
Afisiafi	$84.68_a^{1,2}$ (1.77)	66.79_b^{3} (0.48)	69.94_b^{2} (1.19)	4.05
LSD (0.05)	9.99	5.19	4.59	

a - c values within each row with the same subscripts are not significantly different (p>0.05)
1- 4 values in the same column with the same superscripts are not significantly different (p>0.05)
( ) values in brackets are standard deviations of the mean

97/4489 having the lowest and the native 97/4414 the highest. Water binding capacity reflects the ability of a starch molecule to form, non-covalent bonds with water (Rickard *et al.,* 1991). The water binding capacity of commercial starches is important to the quality and texture of some food products, because it stabilizes them against effects such as syneresis, which sometimes occur during retorting and freezing (Baker *et al.*, 1994). The water binding capacity of the starches were significantly different ($p < 0.05$) indicating the effects of modification on the native starch structure. Wooton and Bamunuarachi (1978) reported that differences in WBC of starches could be due to different degrees of available water binding sites which are considered to be hydroxyl groups and oxygen atoms. The water binding capacity of the native starch was generally higher compared to that of the modified starches. The hydroxyl groups which are required to bind water molecules are all intact in the native starch while some are substituted in the modified starches.

CONCLUSION

The moisture content, pH, water binding capacity, and swelling power of the native starches were generally higher than those of the modified starches. The physicochemical properties of the native starches were significantly affected by the modification processes. The acetylated and carboxylated starches had good physicochemical properties indicating that they are good quality starches with potential industrial use.

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