EFFECT OF AUTOCLAVING TREATMENT ON INCREASING OF RICE AND CORN RESISTANT STARCHES

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ABSTRACT

The present study was conducted at laboratory of food technology department, faculty of agriculture, Kafrelsheish University. This research was carried out study the most suitable conditions for increasing precent of resistant starch for both of rice and corn starches as they considered the main sources of starch in food industry in Egypt. The following experiments and tests were done: preparing resistant starch (RS3) through overlapping among several factors that is number of cooking cycles in autoclave, cooling, cooking time and the percentage of suspended starch of rice and corn starch in water. The resistant starch was estimated and the laboratory digestion of starch, and amylose and amylopectin content were estimated, the most appropriate conditions to get the highest percent of resistant starch (RS3) from the rice and corn starch was known. The physical properties of resistant starch was studied in comparison with native starch. The data showed that the best conditions to obtained the highest yield of resistant starch (RS3) from corn starch was after16 autoclaving- cooling cycles with 60 min autoclaving time at 1:4 starch /water ratio, this yield was 28.50 % this equal to 8.14 fold of that present in native rice starch (3.50 %). On the other hand, the best conditions to obtained the highest yield of resistant starch (RS3) from rice starch was after 20 autoclaving-cooling cycles with 30 min autoclaving time at 1:5 starch /water ratio. At these conditions, RS equal to 6.12 fold of that present in native starch (4.90 %).

INTRODUCTION

Starch is classified as readily digestible starch, slowly digestible starch or resistant starch (RS) based on *in vitro* enzymatic hydrolysis which is assumed to represent the rate of glucose release and its absorption in the gastrointestinal tract. Readily digestible starch and slowly digestible starch represent the amount of starch digested *in vitro* within 20 minutes and between 20-100 minutes, respectively; while RS is not digested *in vitro* within 120 minutes and is assumed to pass into the large intestine undigested **(Englyst *et al*., 1992)**. The term ‘resistant starch’ (RS) was coined and used to describe these starches. Although the term ‘resistant starch’ is not defined by any government agency**,** a group of scientists funded by the European Union (EU) in a concerted action known as EURESTA defined RS as the ‘total amount of starch, and the products of starch degradation that resists digestion in the small intestine of healthy people’ **(Asp 1992; Goldring, 2004).**

RS that reaches the large intestine can act as a substrate for microbial fermentation, the end-products being hydrogen, carbon dioxide, methane and short chain fatty acids (SCFA)**.** In intestine, the indigested starch is fermented by gut microflora, producing short chain fatty acids (SCFA), with high proportion of butyrate. The butyrate stimulated the immunogenicity of cancer cell. As a substrate for colonocytes, it determines the rate of ATP production, and as a single metabolite, it activates proliferation and differentiation. These effects may lead to decreased incidence of colon cancer, atherosclerosis, and obesity related complication in human. Furthermore, various studies demonstrated that consumption of RS can reduce postprandial blood-glucose level and may be play a role in providing improved metabolic control in type 2 diabetes (non-insulin dependent). These also may be benefit for diabetic by lower lipid levels, as well as hypocholesterolemic effect **(Englyst *et al*., 1996; Haralumpu, 2000; Schwiertz *et al*., 2002; Kim andKwak 2009).**

The development of new products is a strategic area of food industry. Consumers are demanding foods that shoe two main properties: the first –one deals with the traditional nutritional aspects of food, whereas, as a second feature, additional health benefits are expected from its regular ingestion. These kinds of food products are often called nutraceutical foods in a rapidly changing world, with altered food habits and stressful lifestyles, it is more and more recognized that a healthy digestive system is essential for the overall quality of life. One of the current tendencies in nutrition and health is to consume low-carbohydrate food products **(Brouns *et al*., 2002 and Saguilan *et al*., 2007).**

Scientific evaluation of the benefits of RS has also generated commercial interest. RSs are a good source of dietary fiber with neutral characteristics and have no effect on the sensory properties of processed foods. Therefore, RSs are now widely used as ingredients to increase the fiber content of products**.** RS has been define as the starch and products of starch digestion not digested in the human small intestine **(Englyst *et al.,* 1992; Food Innovation, 2014).** The objective of the present study is to characterize the autoclaving resistant starch (RS) as an important component produced from corn and rice starches. RS is chemically and physically were also evaluated.

MATERIALS AND METHODS

Rice samples (Sakha 101) were obtained from Rice Research Center, Sakha, Kafr El-Sheikh governorate, Egypt. Corn starch was kindly provided by Starch, Yeast and Detergent Co., Alexandria, Egypt. Amyloglucosidase (EC.3.2.1.3, from Aspergillus nigr, 10,863U/ml), α-amylase (EC.3.2.1.1 type VI-B from porcine pancreas, 19.6 U/mg), glucose oxidase peroxidase reagent (G3360) was purchased from El-Gomhoria Co. for Drugs and Chemicals, Cairo, Egypt. Other chemicals used in this study were of analytical grade.

Proximate chemical composition:

Moisture, crude protein, ether extract, ash and crude fibers contents were determined according to the methods outlined in **A. O. A. C. (2005)**. Total carbohydrates were determined using phenol–sulfuric acid according to the methods described by **Dubois *et al.* (1956).** Caloric value was calculated from the sum of the percentages of crude protein and available carbohydrates multiplied by a factor of 4 (kcal/g) plus the crude fat content multiplied by 9 (kcal/g), according to **Zambrano *et al*. (2004).**

Isolation of rice starch:

Rice starch was isolated from rice flour using the alkaline deproteination method of **Lim *et al*. (1999)** with some modification as follows: Rice grains were dehulled and grounded to powder then, the milled flour passed through 1 mm sieve screen. 200g ground samples were mixed with 500 ml of 0.1% NaOH. The mixture was stirred at room temperature for 3hrs, and stored at 4°C overnight. The supernatant was decanted, and fresh volume of sodium hydroxide was added to the solid phase and stirred for another 3hrs at ambient temperature. The procedure was repeated twice after which the solid phase was washed with 0.1% NaOH, blended and filtered. 500 ml of distilled water was added to the filtrate and allowed to stand for 3 hrs. The supernatant was decanted and 500 ml of distilled water was added again. The procedure was repeated several times until the pH of the filtrate was between 6.0 and 6.5. The starch residue was collected and dried in a vacuum air oven at 40°C for 48 hrs.

Total starch content:

Starch was determined by the direct acid hydrolysis method using concentrated HCL for 2.5 hrs. The sugar of resulted hydrolyzate was determined by Lane and Eynon method **(A. O. A. C., 2005)**. Starch was calculated by multiplying the obtained sugars content by a 0.9 conversion factor.

***In- vitro* starch digestibility (SD):**

In vitro starch digestibility was analyzed using method of **Goni *et al*. (1997)**.

Amylose and amylopectin contents:

The amylose and amylopectin contents of native and treated starches were determined by the method outlined by **Williams *et al*. (1970)**.

<u>Determination of resistant starch (RS):</u>

Resistant starch content in all samples was determined according to the method described by **Goñia *et al*. (1996)** with some modifications as follows: 100 mg of sample was dispersed in 9mL distilled water and incubated with 1ml α-amylase solution (enzyme activity 2,500 U/mL) at 37 °C for 24 h with constant shaking to hydrolyze digestible starch, then deposited with 95% ethanol (ethanol volume was 4 times of the residue) for 12hrs. The obtained residue was washed with 95% ethanol twice, air dried and then treated with 3ml potassium hydroxide solution (4 mol/L) to solubilize RS. RS solution was adjusted to pH 4.75 with 2 mol L-1 hydrochloric acid and 0.4 mol L-1 sodium acetate buffer, incubated with 1 mL amyloglucosidase solution (enzyme activity 1,500 U mL-1) at 60 °C for 45 min with constant shaking, and then heated at boiling bath for 5 min to inactivate enzyme. Glucose formed in solution was determined by titration of Fehling reagent. RS was calculated as glucose (g) × 0.9. RS content was expressed as percent of RS in sample analyzed.

<u>Preparation of resistant starch (RS):</u>

Autoclaving–cooling cycle:

Many trials resulted from combination of time and suspension ratio treatments have been applied to form RS_3 from various sources of native starch, following the method of **Sievert and pomeranz (1989).**

<u>Functional properties of native and treated starches:</u>

Water and oil absorption capacity: Water and oil absorption capacity were determined according to the method described by **Ashwar *et al.* (2016).**

Swelling power (SP) and water solubility: Swelling power and solubility in water were measured according to the method described by **Schoch (1964).**

<u>RESULTS AND DISCUSSIONS</u>

Chemical composition of rice varieties

Gross chemical composition of some rice varieties under study was determined and the results are recorded in Table (1). Data showed that, the moisture content of rice varieties ranged between 12.37 to 13.80 %. These values are in line with those of **Perez, (1993) and Dharmaputra, (1997).** Moisture content plays a great role during the storage for rice **(Amorim *et al*., 2004).** From the same table, it could be observed that Giza 178 variety contains a relatively high level of crude protein content (6.90%)**,** while Egyptian. Jasmine rice variety had the lowest level of Protein content (6.38%).

Table (1): Gross chemical composition (%) of some rice varieties

Constituents (%)	Rice varieties			
	Sakha 101	Jasmine	Giza 178	Black
Moisture	12.64[c]	13.80[a]	13.23[b]	12.37[d]
Dry matter	87.36[b]	86.20[d]	86.77[c]	87.63[a]
Crude protein	6.65[c]	6.38[d]	7.20[a]	6.83[b]
Ether extract	1.06[b]	0.65[d]	0.87[c]	1.93[a]
Ash	0.54[c]	0.49[d]	0.82[b]	1.26[a]
Crude fibers	0.38[d]	0.91[b]	0.74[c]	1.36[a]
Total Carbohydrate	91.75[b]	92.48[a]	91.01[c]	89.98[d]
Available Carbohydrate.	91.37[b]	91.57[a]	90.27[c]	88.62[d]
Amylose	21.28[b]	20.40[c]	19.50[d]	32.57[a]

* Values followed by the same letters in row are not significantly different at ($P \leq 0.05$).
**Each value was an average of three determinations.

Results of the same table also showed that, these were high significant differ in ether extract between the different varieties. The black rice variety had the highest ether extract content (1.93%). **Siebenmorgen and Sun (1994)** reported that surface fat content was inversely related to the degree of milling. A high significant difference in ash content was recorded between the varieties. The black rice variety contained the highest ash content (1.26%). **Amorim *et al*. (2004**) found that, the ash content in the rice was 0.4%. Furthermore, the ash content indicated the amount of minerals. As shown in Table (1), these were significant differences in total carbohydrates content between the varieties, where the Black rice had lowest content of carbohydrates (89.98%) and Jasmine rice had highest value (92.48%). As for the fiber content, it could be noticed that there are high significant differences between the varieties. The highest fiber content was in Black (1.36%), while the lowest values were Sakha 101 (0.38%).

El-Bana *et al*. (2016) found that, black rice contains high content of ether extract, ash and crude fibers, but lower content of total carbohydrate, compared with other varieties. The results in Table (1) show also that, amylose content of Giza 178 rice variety significantly low content 19.50% compared to other varieties. The highest contents for amylose were found in Black rice variety (32.57%. While Sakha 101 and Jasmine recorded 21.28% and 20.40%; respectively. These results are in agreement with those reported by **El-Bana (2010)**.

Kadan *et al*., (1997) found that amylose content of short-, medium- and long-grain rice ranged from 12% to 21%, but the Waxy rice is a short grain cultivar with almost 100% amylopectin starch. Both the amylose content and amylopectin branch chain-length distributions affected starch pasting properties **(Jane *et al*., 1999)**. For all the previous reasons the variety which content a large amount of amylose is very suitable for preparation of resistant starch because amylose content is consider the most important factor for preparation of the resistant starch, also amylose is the principle starch fraction required for formation RS3.

As the results in Table (1) showed that, black rice variety had the highest amylose content between all the varieties under study it was not chosen to use in preparing resistant starch (RS) because it produced with a little amount in Egypt specially in Research center in the same time sakha 101 variety recorded the second content of amylose (20.40%) between the rice varieties and also it was produced by a large quantity in Kafr El-sheikh Government it was chosen to use for preparation of the resistant starch in this study.

Chemical composition of rice flour, isolated rice starch and native corn starches

Rice flour of sakha 101, isolated rice starch and native corn starch were analyzed for their chemical composition. The obtained results in indicated that, rice flour contained 12.64 % moisture, 87.36% dry matter, 6.65% crude protein, 1.06 % ether extract, 0.54% ash, 91.75% total carbohydrate, 91.37% available carbohydrate and 038% crude fibers While isolated rice starch contained 11.80% moisture, 88.12% dry matter, 0.39% crude protein, 0.17% ether extract, 0.20% ash, 99.24% total carbohydrate, 99.24 % available carbohydrate and 0% crude fibers. Corn starch contained 11.50% moisture, 88.50% dry matter, 0.34% crude protein, 0.40% ether extract, 0.23% ash, 99.03% total carbohydrate and 99.03% available carbohydrate. These results are in agreement with those of **(BeMiller *et al*., 2007).**

Table (2): Chemical composition of rice flour, isolated rice starch and native corn starch (calculated on dry weight basis)

Sample / Component	Rice flour	Isolated rice starch	Native corn starch
Moisture	12.64[a]	11.88[ab]	11.50[b]
Dry matter	87.36[b]	88.12[ab]	88.50[a]
Crude protein	6.65[a]	0.39[b]	0.34[b]
Ether extract	1.06[a]	0.17[c]	0.40[b]
Ash	0.54[a]	0.20[b]	0.23[b]
Crude fibers	0.38	0.00	0.00
Total carbohydrate	91.75[b]	99.24[a]	99.03[a]
Available carbohydrate	91.37[b]	99.24[a]	99.03[a]

* Values followed by the same letters in row are not significantly different at (P≤ 0.05).
**Each value was an average of three determinations.

Total starch, amylose, amylopectin and resistant starch (RS) contents in rice and corn starches:

Total starch, amylose, amylopectin and resistant starch contents of rice and corn starches were determined. The results are presented in table (3), it could be observed that total starch was 98.07, 96.61% for rice and corn starch; respectively. Amylose and amylopectin content of rice starch were 21.28 and 78.72% of total starch, respectively. According to **Ashwar *et al*. (2015)** the amylose content of native rice starches ranged between 8.56 and 24.78 % of starch. Amylose and amylopectin contents of corn starch were 24.30, 75.70% of total starch, respectively as shown in table (3).

Table (3): Total starch, amylose, amylopectin and resistant starch (RS) contents in rice and corn starches

sample / component	Rice starch	Corn starch	Significant
Total starch	98.07 ± 0.100	96.61 ± 0.260	**
Amylose	21.28 ± 0.590	24.30 ± 0.250	**
Amylopectin	78.72 ± 0.270	75.70 ± 0.250	**
Resistant starch	3.50 ± 0.500	4.90 ± 0.400	**

NS, * and ** indicate not significant and significant at $p < 0.05$, $p < 0.01$; respectively.

They were in a normal range of 20- 30 % for amylose and 70-80% for amylopectin according to **Baldwin, (2001).** Resistant starch content of native corn and rice starches were 4.90 and 3.50, respectively.

Effect of autoclaving –cooling cycles and time on formation of resistant starch (RS), digestibility (SD), amylose and amylopectin contents in rice and corn starches:

Many combinations of autoclaving time, number of autoclaving-cooling cycles and starch/water ratio treatments have been carried out to RS3 formation from various sources of native starch.

Rice starch at starch /water ratios 1:2, 1:3, 1:4 and 1:5.

The RS, SD, amylose and amylopectin contents of rice starch that was autoclaved at 121°C for 30, 60 and 90 min in suspension of starch /water ratios 1:2, 1:3, 1:4 and 1:5 then cooled up to twenty cycles and air-dried are presented in Figures (1-4).

Data in Figure (1) indicated that significant differences ($p \leq 0.05$) were found between the percentages of RS of rice starch in different combinations of autoclaving time, starch /water ratios and number of autoclaving-cooling cycles treatments.

The highest RS yield treatments (17.90 %) was obtained after 20 autoclaving- cooling cycles for 90 min. The lowest value (4.20 %) was obtained after 4 autoclaving- cooling cycles for 90 min for starch /water ratios 1:2. While, the RS increased from 4.50 % after 4 autoclaving-cooling cycles at 30 min to 22.50 % after 16 autoclvaing –cooling cycles at 60 min for starch /water ratios 1:3. Also, autoclaving resulted in an increase in RS. Autoclaved rice starch at 1:4 starch /water contained 5.75 % RS (of dry wet basis) after 4 futher autoclaving- cooling cycles at 60 min, this figure increased to 28.50 % after16 further autoclaving – cooling cycles at 60 min. Meanwhile, the highest RS yield treatments was obtained after 20 autoclaving- cooling cycles for 30 min. The lowest value was obtained after 4 autoclaving- cooling cycles for 60 min for starch /water ratios 1:5. These results were similar to the study of **Szczodrak and Pomeranz (1991).**

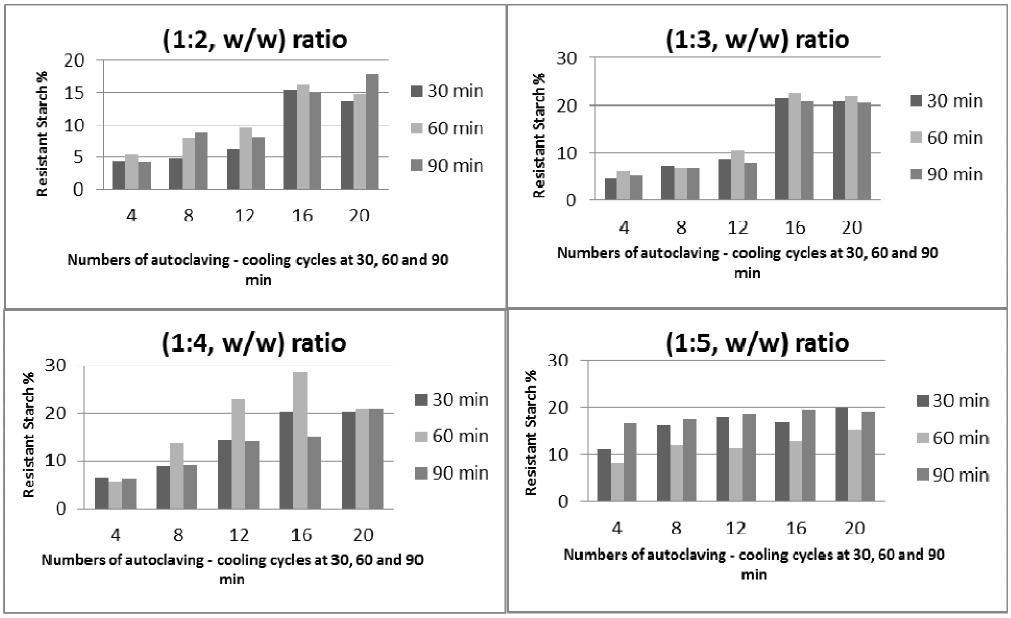

Fig. 1: Effect of autoclaving –cooling cycles and time at yields of resistant rice starch at starch: water (1:2, 1:3, 1:4 and 1:5 w/w) ratios

Data also showed that the SD ranged from 80.17 % after 20 autoclaving - cooling cycles for 90 min to 93.90% after 4 autoclaving- cooling cycles for 90 min at 1:2 starch /water contained. While, SD decrease from 93.57% after 4 autoclaving- cooling cycles at 30 min to 75.57 % after 16 autoclaving – cooling cycles at 60 min for starch/ water ratio 1:3. On the other hand, The value of SD decreased from 92.32 % after 4 autoclaving – cooling cycles at 60 min to 69.57 % after 16 autoclaving – cooling cycles at 60 min for starch/ water ratio 1:4. For starch/ water ratio 1:5 the SD showed decrease from 90.07% for 4 autoclaving–cooling cycles at 60 min to 78.07% for 20 autoclaving–cooling cycles at 30 min autoclaving times, respectively.

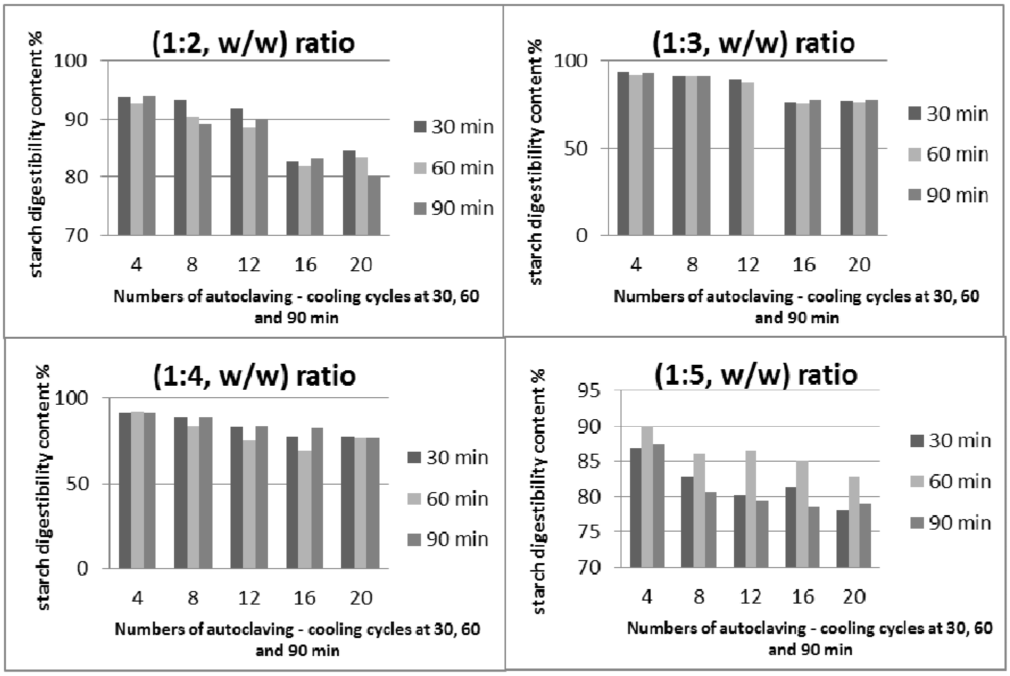

Fig. 2: Effect of autoclaving –cooling cycles and time at yields of starch rice digestibility at starch: water (1:2, 1:3, 1:4 and 1:5 w/w) ratios

Amylose content varied from 16.55 % (20 autoclaving- cooling cycles for 30 min) to 20.90 % (4 autoclaving- cooling cycles for 90 min) at starch/water ratio 1:2. Also, amylose content ranged between15.10 and 20.00 % the lowest value was obtained after 20 autoclaving – cooling cycles at 90 min, while highest value obtained for 4 autoclaving at 30 min for starch/water ratio 1:3. Amylose content decreased from 20.00 % after 4 autoclaving – cooling cycles at 60 min to14.90 % after 20 autoclaving – cooling cycles at 30 min for starch/water ratio 1:4. Meanwhile, the amylose contents decreased from 19.50 for 4 autoclaving–cooling cycles at 60 min to14.00 % for 20 autoclaving–cooling cycles at 30 min autoclaving time.

Corn starch at starch /water ratios 1:2, 1:3, 1:4 and 1:5

The RS, SD, amylose and amylopectin contents of corn starch that was autoclaved at 121°C for 30, 60 and 90 min in suspension of starch /water ratios 1:2, 1:3, 1:4 and 1:5 then cooled up to twenty cycles and air-dried are presented in Figures (5-8). The data presented in figure (5) showed that the highest yield of RS content was obtained after twenty autoclaving-cooling cycles at 90 min at starch/water ratio 1:2.

Meanwhile, the highest value of RS content for starch/water ratio 1:3, 1:4 and 1:5 recorded after twenty autoclaving-cooling cycles at 30 min with values 17.80%, 22.30% and 30.00%, respectively. It is clear that increasing the number of cycles increased yield of RS. These results were line with **Sievert and pomeranz (1989**).

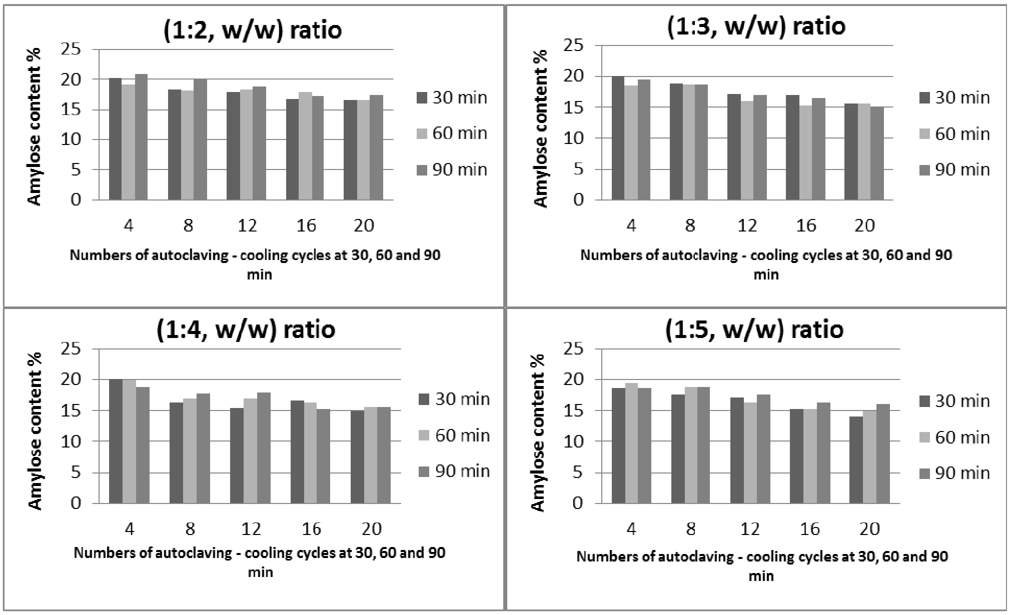

Fig. 3: Effect of autoclaving –cooling cycles and time at yields of amylose at starch: water (1:2, 1:3, 1:4 and 1:5 w/w) ratios

Data in Figure (4) indicated that, the amylopectin content at starch/water ratio 1:2 ranged between 81.52% and 77.17 % for 4 autoclaving–cooling cycles at 90 min and 20 autoclaving–cooling cycles at 30 min, respectively. For the starch/water ratio 1:3, the highest content of amylopectin was showed after 4 autoclaving – cooling cycles at 30 min while the lowest content was after 16 autoclaving – cooling cycles at 60 min. Whereas, amylopectin decreased from 83.17 to 78.07 % after 20 autoclaving – cooling cycles at 30 min and 4 autoclaving – cooling cycles at 60 min, respectively for starch/water ratio 1:4. Meanwhile, amylopectin increase after 20 autoclaving – cooling cycles at 30 min and decrease after 4 autoclaving – cooling cycles at 60 min autoclaving time at starch/water ratio 1:5.

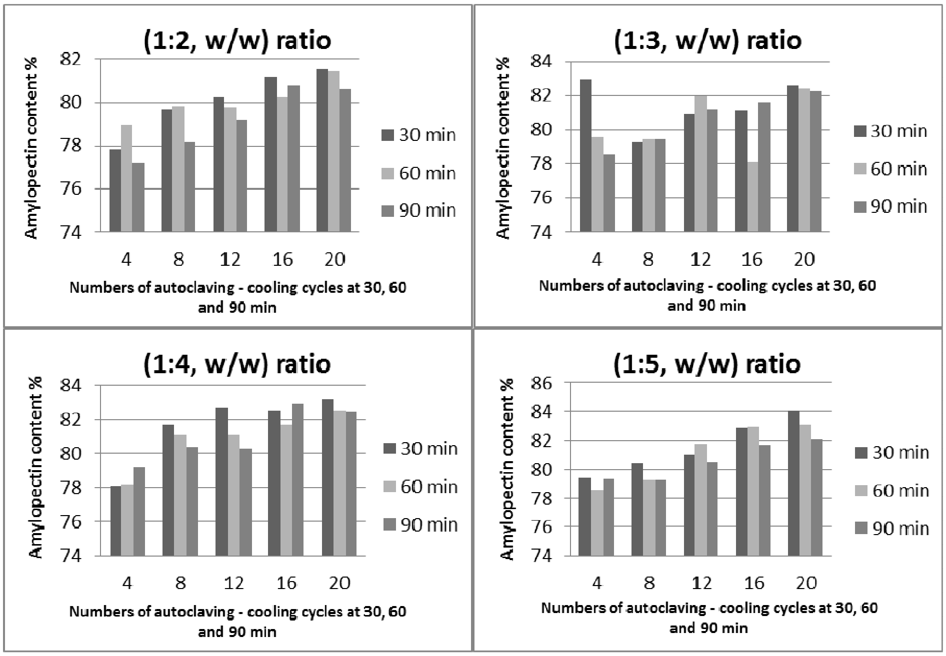

Fig. 4: Effect of autoclaving –cooling cycles and time at yields of amylopectin at starch: water (1:2, 1:3, 1:4 and 1:5 w/w) ratios

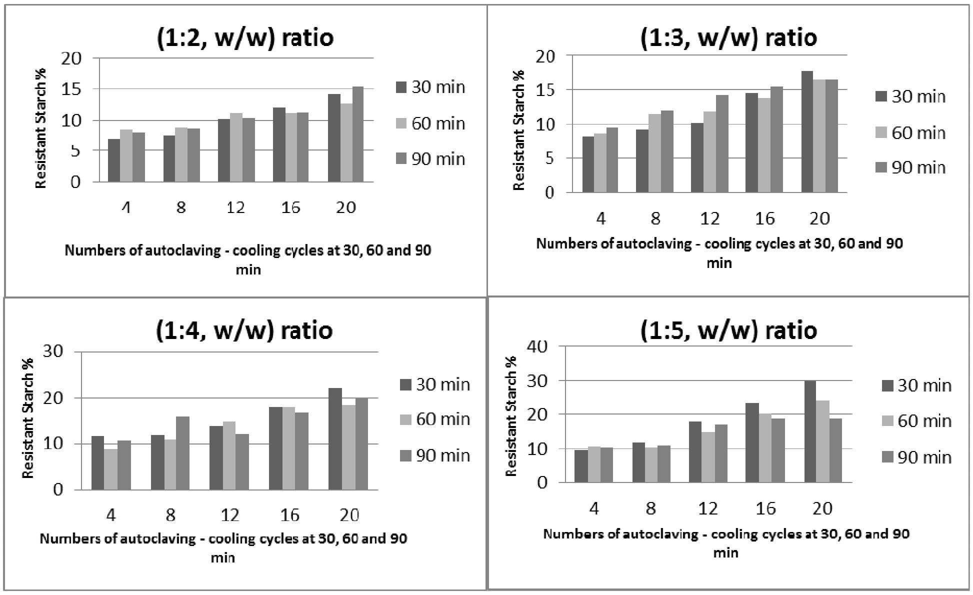

Fig. 5: Effect of autoclaving –cooling cycles and time at yields of resistant corn starch at starch: water (1:2, 1:3, 1:4 and 1:5 w/w) ratios

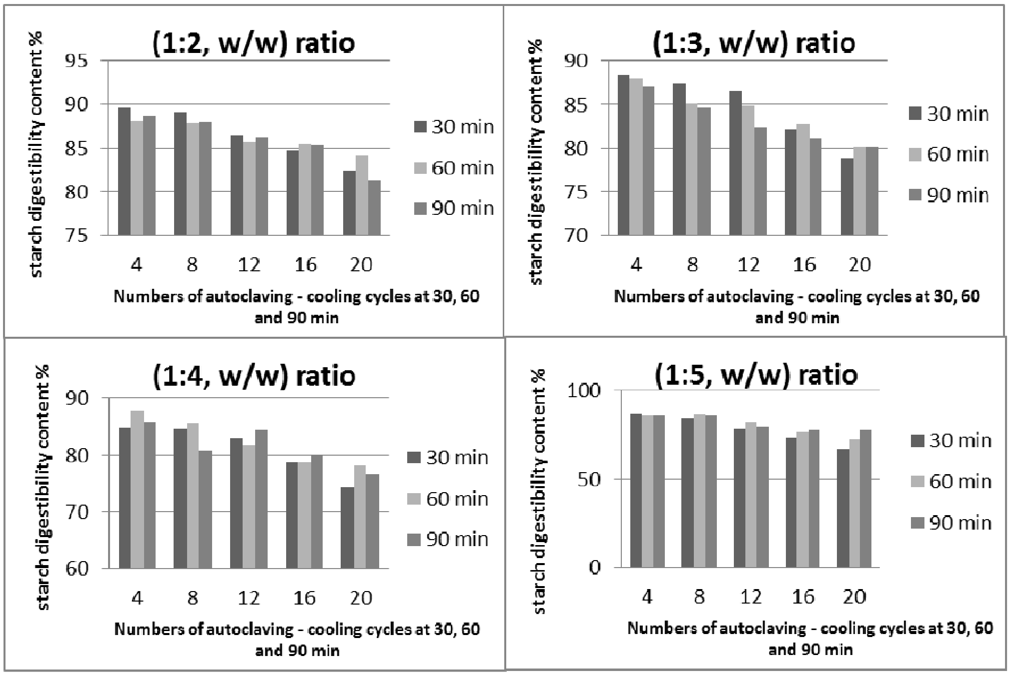

Fig. 6: Effect of autoclaving –cooling cycles and time at yields of starch corn digestibility at starch: water (1:2, 1:3, 1:4 and 1:5 w/w) ratios

From the data in figure (6), the highest yield of starch digestibility content was obtained after four autoclaving-cooling cycles at 30 min for starch/water ratios 1:2, 1:3 and 1:5. Meanwhile, the highest value of starch digestibility content for starch/water ratio 1:4 recorded after twenty autoclaving-cooling cycles at 60 min. on the other hand, the lowest yield of starch digestibility content was obtained after twenty autoclaving-cooling cycles under different combinations of starch /water ratio treatments. These results indicated that starch digestibility content decreased by increasing autoclaving- cooling cycles. With respect to amylose content, the highest content of amylose was obtained after four autoclaving-cooling cycles at 30 min, 60 min, for starch/water ratios 1:2, 1:3 and 1:5. Also, the highest value of amylose content for starch/water ratio 1:4 recorded after twenty autoclaving-cooling cycles at 60 min. On the other side, the lowest value for amylose were founded after twenty cycles of autoclaving at 90 min, 30 min, 90 min and 30 min for starch/water ratios 1:2, 1:3, 1:4 and 1:5, respectively. The data are in a good agreement with that found by **Kim and kwak (2009)**.

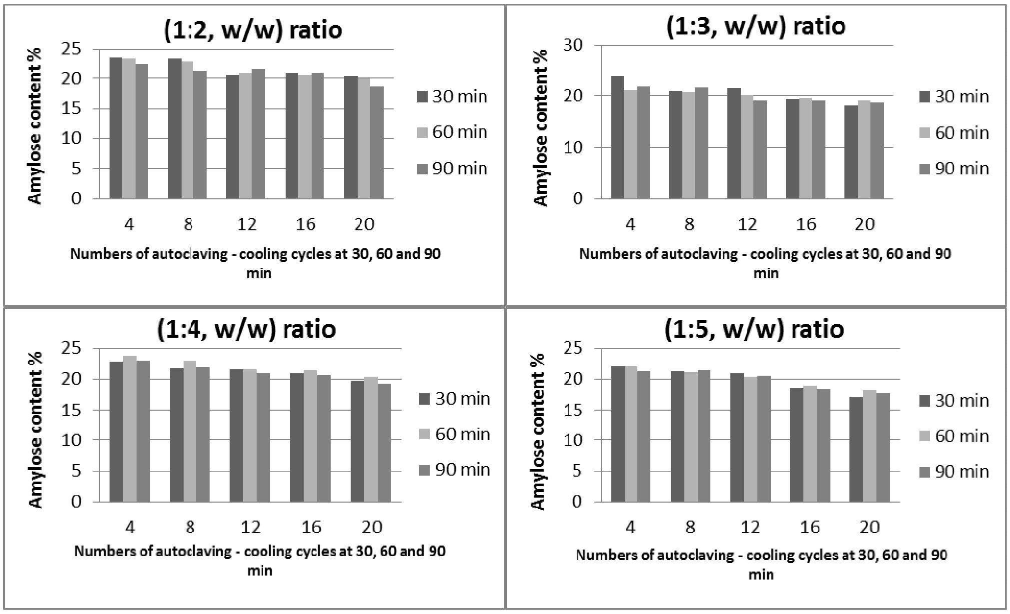

Fig. 7: Effect of autoclaving –cooling cycles and time at yields of amylose at starch: water (1:2, 1:3, 1:4 and 1:5 w/w) ratios

For amylopectin content Figure (8), the highest content of amylopectin was obtained after twenty autoclaving-cooling cycles at 30 min for starch/water ratios 1:2 and 1:4 also, at 90 min for starch/water ratios 1:3 and 1:5. However, the lowest content of amylopectin was obtained after 4 cycles autoclaving-cooling cycles under different combinations of autoclaving time and starch/water ratio treatments.

Data indicated that amylose content decreased and amylopectin content was increased by increasing autoclaving- cooling cycles and reduction of amylose content was more pronounced. This may be due to amylopectin retrogrades very slowly according to **(Rashmi and urooj, 2003)**. According to **(Kawabata *et al*., 1994)** heat moisture treatment (HMT) may increase RS levels in starch by increased interaction between amylose and amylopectin and / or transformation of single amylose- chain into its double helical crystals **(Lorenz and Kulp, 1982).**

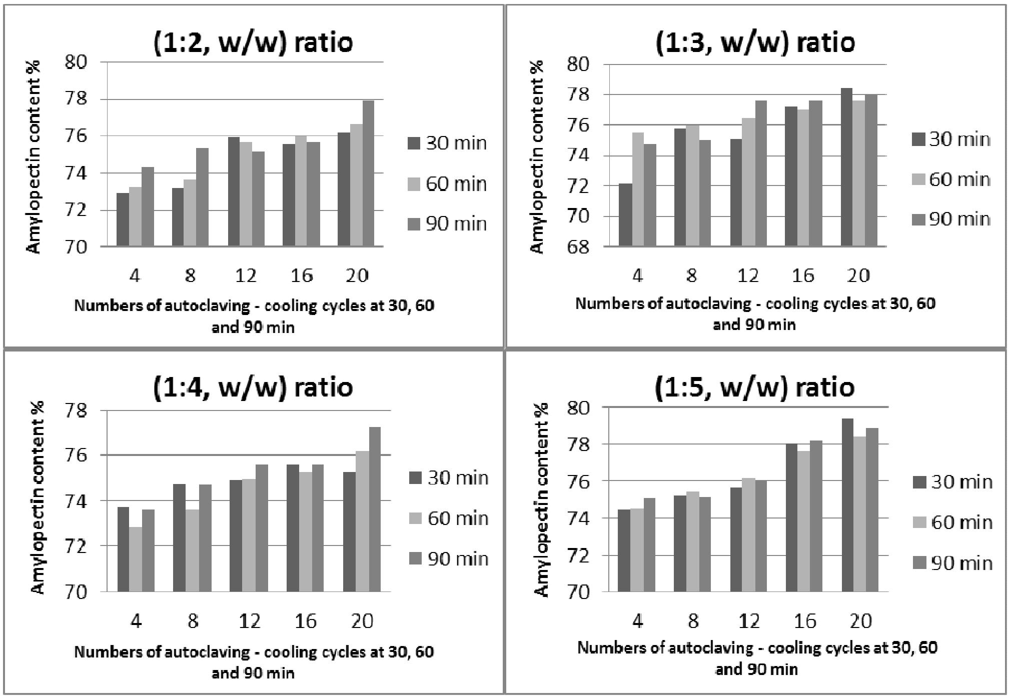

Fig. 8: Effect of autoclaving –cooling cycles and time at yields of amylopectin at starch: water (1:2, 1:3, 1:4 and 1:5 w/w) ratios

The optimum conditions for resistant starch (RS) formation from corn and rice starches

As shown in Table (4), for the best conditions to obtained the highest yield of resistant starch (RS_3) from corn and rice starches, the highest resistant corn starch (30 %) was obtained after 20 autoclaving-cooling cycles with 30 min autoclaving time at 1:5 starch /water ratio. At these conditions, RS equal to 6.12 fold of that present in native starch (4.90 %). Whereas highest yield of rice resistant starch obtained 16 autoclaving- cooling cycles with 60 min autoclaving time at 1:4 starch /water ratio, this yield was 28.50 % this equal to 8.14 fold of that present in native rice starch (2.50 %). **Onyango, *et al*., (2006)** found that the amount of RS_3 formed increased with increasing autoclaving time, attaining a maximum value at 45 min autoclaving time at 121°C. The RS_3 yields declined when the autoclaving time exceeded 45 min indicating the effect of thermal hydrolysis of starch. Shorter autoclaving times, giving high RS_3 yields **(Goni *et al*., 1996),** which significantly better than the 1h treatment **(Eerlingen *et al*., 1993a)** or 2h **(Shamai *et al*., 2003)** as autoclaving times. The formation of Rs_3 is a two- stage process that involves starch hydrolysis followed by recrystallization of amylose polymers. Whereas recrystallization takes place during incubation, starch hydrolysis occurs during autoclaving (**Eerlingen *et al*., 1993b)**.

Starch hydrolysis should preferably result in amylose polymers with degrees of polymerization of 100-300 glucose units. Amylose is the principle starch fraction required for the formation of RS3 because its polymers have a high degree of polymerization that form enzyme-resistant double helices stabilized by hydrogen bonds. When the degree of polymerization is less than 100 glucose units, the RS3 yield is low because the polymers are not long enough to form enzyme-resistant crystallites.

Table (4): Optimum conditions for resistant starch (RS) formation from rice and corn starches

	Resistant starch		Sig.
	Rice	**Corn**	
Native starch	4.90 ± 0.250	3.50 ± 0.260	**
Treated starch	30.00 ± 0.350	28.50 ± 0.250	**
Optimum conditions:			
-Autoclaving-cooling cycles	20	60	
- Time (min)	30	60	
- Starch : water ratio (w/w)	1:05	1:04	

NS, * and ** indicate not significant and significant at $p < 0.05$, $p < 0.01$; respectively.

On the other hand, insufficient hydrolysis of amylose is undesirable since amylose polymers with degrees of polymerization higher than 300 cannot effectively align to form enzyme- resistance crystallites (**Eerlingen *et al*., 1993b)**. In general, the starch digestibility negatively correlated with resistant starch formed. About the relationship between formation of resistant starch and amylose and amylopectin contents, data showed that the amylose and amylopectin negatively correlated with resistant starch formed.

Physical properties native and treated rice and corn starches

Swelling power, solubility and water and oil absorption capacities

When the starch granules, heated in water, being the swelling power and solubility phenomenon. Swelling depends on the starch granule size and on the OH- groups that can interact with molecules of water favoring swelling of starch granules. The values of swelling power and solubility of native and resistant rice and corn starches are presented in Table (5). It is clear that the differences between native corn and rice starches concerning of swelling power and solubility were significant ($p \leq 0.05$) moreover the HMT understudy affect significantly of different starches. As shown from the result in Table (5), a parallel relationship was observed between swelling power and solubility of starch.

Table (5): Swelling power, solubility, water and oil absorption capacity of native and treated rice and corn starches

Sample Property	Rice starch		Corn starch	
	Native	**Resistant**	**Native**	**Resistant**
Swelling Power (%)	16.66[b] ± 0.200	6.15[d] ± 0.150	19.35[a] ± 0.050	7.67[c]± 0.230
Solubility (%)	12.40[b] ± 0.140	5.89[c] ± 0.190	14.74[a] ± 0.200	5.93[c] ± 0.03
Water Absorption capacity (g/g)	0.80[b] ± 0.200	1.60[a] ± 0.280	0.95[b] ± 0.120	1.93[a] ± 0.200
Oil Absorption capacity (g/g)	0.66[b] ± 0.070	1.05[a] ± 0.080	0.75[b] ± 0.200	1.20[a] ± 0.100

Means of values having the same case letters within a row are not significantly different (P<0.05).

The starches having high swelling power were also high in solubility. Data a show that the swelling power and solubility decreased from (16.66 to6.15 g/g) and (12.40 to 5.89 %) for native and resistant corn starch, respectively. Whereas, swelling power and solubility decreased from (19.35 to7.67 g/g) and (14.74 to 5.93 %) for native and resistant rice starch, respectively. **Hormdok and Noomhorm, 2007** reported that the HMT (20% moisture, 92.5°C/ 30 min) of rice starch (27% amylose) reduced the swelling power from 14.11 g/g of native starch to 10.29 g/g. Several authors have observed a reduction in the swelling power of the heat moisture treatment in potato, cassava, rice, sorghum and corn starches. The reduction in swelling power following hydrothermal modification has been attributed to internal rearrangement of the starch granules, which causes further interaction amongst the starch functional groups making it form more ordered double helical amylopectin side chain clusters. This accounts for increased starch crystallinity according to **Hoover and Manuel, 1996)**,**(Adebowale and Lawal, 2002) (Gunaratne and Hoover, 2002)**, **(Hormdok and Noomhorm, 2007), (Olayinka, *et al*., 2008)** and **(Chung, *et al*., 2009)**.

According to **Aparicio- Saguilan *et al*. (2005)** and **Kim and Kwak (2009)**, the number of autoclaving-cooling cycles significantly affected swelling power. It significantly decreased as the number of cooling cycles increased. Previous studies have also reported significant difference in the swelling index of different rice varieties **(Ashwar *et al*., 2014a).** Autoclaving- retrogradation treatment decreased the swelling index of rice starch samples significantly (P ≤ 0.05). The decrease in swelling index of treated starches could be attributed to increased crystallite perfection and to additional interaction between amylose amylose and/or amylose-amylopectin chains **(Lan *et al*., 2008).** Autoclaving- retrogradation treatment reduced the solubility index of all starch samples. The decrease in starch solubility with treatment can be attributed to an internal rearrangement of starch granules that provides higher interactions between starch functional groups, the formation of more ordered amylopectin clusters, and the formation of amylose-lipid complexes within starch granules (**Dias *et al*., 2010).** The increase in water absorption capacity was mainly due to the gelatinization of starch caused by autoclaving. Previous studies have also reported significant increase in the water absorption capacity in autoclaved, HMT and annealed starches **(Chung, liu and Hoover, 2009).**

The values of Water and oil absorption capacities of native and resistant corn and rice starches are presented in table (5). It is clear that the differences between native corn and rice starches concerning of swelling power and solubility were significant ($p \leq 0.05$) moreover the HMT understudy affect significantly of different starches. As shown from the result in table (5). It is clear that the differences between native corn and rice starches concerning of water absorption capacity and oil absorption capacity were significant ($p \leq 0.05$). Data showed that water absorption capacity and oil absorption capacity increased from (0.80 to 1.60 g/g) and (0.66 to 1.05g/g) for native and resistant corn starch, respectively. Whereas, water absorption capacity and oil absorption capacity increased from (0.95 to 1.93 g/g) and (0.75 to 1.20 g/g) for native and resistant rice starch, respectively. Oil absorption capacity of starch might be due to the entrapment of oil in a porous starch matrix by capillary forces or inside helical structures of amylose or amylopectin due to the formations of amylose-lipid complexes. Increased oil absorption capacity of treated starches might be due to the exposure of more hydrophobic moieties of amylose-lipid complexes due to high temperature. **Olayinka, Adebowale and Olu-Owolabi (2008)** they also reported that increase in oil absorption capacity in sorghum starch with HMT.

REFERENCES

Adebowale, K. O. and Lawal, O. S. (2002). Effect of annealing and heat moisture conditioning on the physicochemical characteristics of Bambara groundnut (*Voandzeia Subterranea*) starch. *Nahrung-Food*, 46: 311- 316.

Amorim, J. A.;Eliziário, S. A .;Gouveia, D. S.; Simões, A. S. M.; Santos, J. C. O.;Marta M. C.; Souza, A. G. and Maria F. S. T. (2004). Thermal analysis of the rice and by-products. *J. of Thermal Analysis and Calorimetry*, 75, 393–399.

Aparicio-Saguil´an A, Flores-Huicochea E, Tovar J, Garc´ıa-Su´arez F, Guti´errez-Meraz F, Bello-P´erez LA.(2005). Resistant starch-rich powders prepared by autoclaving of native and linearized banana starch: partial characterization. Starch/Stärke 57:405–12.

Ashwar, B. A., Gani, I. A.; Asima, S.; Farooq, A. M.; Dharmesh, C. S. (2016). Production of resistant starch from rice by dual autoclaving-retrogradation treatment. In vitro digestibility, thermal and structural characterization, *Food Hydrocolloids* (2016), doi: 10.1016/ j.foodhyd.2015.12.004.

Ashwar, B. A., Gani, I.A., Wani, I. A., and Masoodi, F. A. (2015). Preparation, health benefits and applications of resistant starch-a review. Starch/Stärke, 67, 1–15.

Ashwar, B. A., Shah, A., Gani, A., Rather, S. A., Wani, S. M., Wani, I. A., Masoodi, F. A. (2014a). Effect of gamma irradiation on the physicochemical properties of alkali extracted rice starch. *Radiation Physics and Chemistry*, 99, 37–44.

Asp. (1992) Preface: Resistant Starch, proceedings of the 2nd plenary meeting of EURESTA: European Flair Concerted Action No. 11 on physiological implications of the consumption of resistant starch in man. *Eur J Clin Nutr* 46:S1.

Association of Official Agricultural Chemists (AOAC). (2005). Official Methods of Analysis, 17th Ed. W. Horwitz, ed. AOAC Int.: Gaithersburg, MD.

Baldwin, P. M. (2001). Starch- granule associate proteins and polypeptides: a review. Starch/ Stärch: 475- 503.

BeMiller, J.N. (2007). Carbohydrate chemistry for food scientists, 2nd ed. AACC International: St. Paul, MN. : 389.

Brons, F; Kettliz, B., and Arrigoni, E., (2002). Resistant starch and the butyrate revolution. *Trends in Food Science and Technology* 13: 251-261.

Chung, H.; Hoover, R.; Liu, Q. (2009a). The impact of single and dual hydrothermal modifications on the molecular structure and physicochemical properties of normal corn starch. *Int J Biol Macromol* 44:203–10.

Dhamaputra, O. (1997). The effects of milling degree and type of bag on fungal infection and some chemical contents of stored milled rice. Biotropia., 10: 42-62.

Dias, A.; R. G.; Zavareze, E; Spier, F; de Castro, L. A. S.; and Gutkoski, L. C. (2010). Effects of annealing on the physicochemical properties and enzymatic susceptibility of rice starches with different amylose contents. *Food Chemistry*, 123(3), 711–719.

Eerlingen, R. C.; Crombez, M.; and Declour, J. A. (1993a). Enzyme – resistant starch. I. Quantative and qualitative influence of incubation time and temperature of autoclaved starch on resistant starch formation. *Cereal Chemistry* 70: 339- 344.

Eerlingen, R. C.; Deceunin, M.; and Declour, J. A. (1993b). Enzyme – resistant starch. II. Influence of amylose chain length on resistant starch formation. *Cereal Chemistry* 70 (3): 345 – 350.

El- Bana, M. A., W.K. Galal and S.T.El- Hadidie. (2010). Physico- and technological studies on some Egyptain rice varieties. *J.Food and Dairy Sci.* Mansora Univ., 1(4): 161- 172.

El- Bana, M. A.,Abd Elsattar, A.S., Somaya,M.M.(2016). Physical properties ,chemical and technological evaluation of waxy and non waxy rice. *Minnufiya J. Agri.Res*.Vol. 41 No. 1(1): 53 – 64.

Food Innovation. (2014). Why Hi-maize® resistant starch. Received August26,2014fromhttp://www.foodinnovation.com/foodinnovation/en-us/Hi-maize/Pages/Why-Hi-maize.aspx

Goñi, I., Manas, E., Garcia-Diz, F., Saura-Calixto, F. (1996). Analysis of resistant starch: a method for food and food products. *Food Chemistry*, 56, 445-449.

Goñi, I.; Garcia-Alonso, A.; and Saura-Calixto, F. (1997) A starch hydrolysis procedure to estimate glycemic index. *Nutrition Research*, 17(3):427-437.

Gunaratne, A.; and Hoover, R. (2002). Effect of heat- moisture treatment on structure and physicochemical of tuber and root starches. *Carbohydrate Polymers* 49: 425- 437.

Haralampu, S.G. (2000). Resistant starch-a review of the physical properties and biological impact of RS3. *Carbohydrate Polymers*, 41:285-292.

Hoover, R.; and Manuel, H. (1996). Effect of heat- moisture treatment on the structure and physicochemical properties on normal maize, waxy maize, dull waxy maize and amylomaize V Starches. *Journal of Cereal Science*, 23: 153- 162.

Hormdok, R .and Noomhorm, A. (2007). Hydrothermal treatment of rice starch for improvement of rice noodle quality. LWT. *Food Science and Technology*, 40: 1723- 1731.

Jane, J.; Chen, Y.Y.; Lee, L.F.; McPherson, A.E.; Wong, K.S.; Radosavljevic, M.; Kasemsuwan, T. (1999). Effects of amylopectin branch chain length and amylose content on the gelatinization and pasting properties of starch. *Cereal Chem* 76(5):629–37.

Kadan, R.S.; Champagne, E. T.;Ziegler, G. M.; and Richard, O. A. (1997). Amylose and protein contents of rice cultivars as related to texture of rice- based fries. *J. of Food Sci*., 62(4): 701-703.

Kawabata, A.; Takase, N.; Miyoshi, E.; Sawayama, S.; Kimura, T. and Kudo, K. (1994). Microscopic observation and X- ray diffractometry of heat/ moisture- treated starch granules. Starch, 46: 463- 469.

Kim, S.K., and Kwak, J.E. (2009) Formation of resistant starch in corn starch and estimation of its content from physicochemical properties. Starch-Starke, 61(9):514-519.

Lan, H.; Hoover, R.; Jayakody, L.; Liu, Q.; Donner, E.; Baga, M. (2008). Impact of annealing on the molecular structure and physicochemical properties of normal, waxy and high amylose bread wheat starches. *Food Chemistry*, 111(3), 663–675.

Lim, S. T.; Kasemsuwan, T.; and Jane, J. L. (1999). Characterization of Phosphorus in Starch by P- 31-Nuclear Magnetic-Resonance Spectroscopy. *Cereal Chemistry* 71:488-493.

Lorenz, K., and Kulp, K. (1982). Cereal and root starch modification by heat-moisture treatment. I. Physico- Chemical properties. Starch, 34: 50-54.

Olayinka, O.O.; Adebowale, K. O.; and Olu-Owolabi, B.I. (2008). Effect 486 of heat-moisture treatment on physicochemical properties of white sorghum starch. *Food Hydrocolloids*, 22, 225- 230.

Onyango, T.; Bley, A.; Jacob, T.; Henle , L.; and Rohm, H. (2006). Influence of incubation temperature and time on resistant starch type III formation from autoclaved and acid- hydrolyzed cassava starch. *Journal of Carbohydrate Polymers*. 66: 494–499.

Perez, C.M. (1993). Gel consistency and viscosity of rice, International Rice Research Institute (IRRI). pp. 292.

Rashmi, S.and Urooj, A. (2003). Effect of processing on nutritionally important starch fraction in rice varieties. *Interanational Journal of Food Sciences and Nutrition* 54: 27- 36.

Saguilan,A.A; Sayago- Ayerdis, S.G.; Vargas- Torres, A.; Tovar,J.; Ascencio-Oter, T. E. and Bello- Perez, L . A. (2007). Slowly digestible cookies prepared form resistant starch- rich linearized banana starch. *Journal of food composition and analysis* 20: 175- 181.

Schoch, T. J. (1964). Swelling Power and Solubility of Starch Granules, in Methods in Carbohydrate Chemistry, Vol. IV, (Ed. R. L. Whistler) Academic Press, Orlando, F1: 106- 108.

Schwiertz, a.; Lehman, U.; Jacobasch, G.; Blaut, M. (2002). Influence of resistant starch on the SCFA production and cell counts of butyrate-producing Eubacterium spp. in the human intestine. *J Appl Microbial* 93:157–62.

Siebenmorgen, T.J.; and Sun, H. (1994). Relationship between milled rice surface fat concentration and degree of milling as measured with commercial degree of milling as measured with commercial milling meter. *Cereal Chem*. 71(4): 327-329.

Sievert, D.; and Pomeranz, Y. (1989). Enzyme-resistant starch 1. Characterization and evaluation by enzymatic, thermo analytical and microscopic methods. *Cereal Chemistry* 66(4):342-347.

Szczdrak, J.; And Pomeranz, Y. (1991). Starch and enzyme- resistant starch from high- amylose barely. *Cereal Chem* 68 (6), pp. 589- 596.

Zambrano, F.; Despinoy, P.; Ormenese, R.C.; and Faria, E.V. (2004). The use of guar and xanthan gums in the production of light low fat cakes. International Journal of Food Science and Technology, 39:959-966.

الملخص العربى

تأثير المعاملة بالأوتوكلاف على زيادة نشا الأرز والذرة المقاوم

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اجريت هذه الدراسة فى معمل قسم تكنولوجيا الاغذية – كلية الزراعة – جامعة كفرالشيخ. تم إجراء هذا البحث لدراسة أنسب الظروف لزياده نسبة النشا المقاوم لكل من نشا الأرز والذرة بوصفهما المصادر الرئيسية فى صناعة المواد الغذائية فى مصر. وقد أجريت التجارب والاختبارات التالية : إعداد النشا المقاوم (RS3)من خلال التداخل بين عدة عوامل هي عدد دورات الطبخ فى الأوتوكلاف والتبريد وزمن الطبخ ونسبة معلق النشا والماء على نشا الأرز والذرة. تم تقدير النشا المقاوم ، والهضمية المعملية للنشا ، ومحتوى الأميلوز والأميلوبكتين وتم معرفة أنسب الظروف للحصول على أعلى نسبة نشا مقاوم (RS3) من نشا الأرز والذرة ودراسة الخواص الفيزيائية للنشا المقاوم الناتج بالمقارنة بالنشا الخام. اظهرت النتائج أنه يمكن الحصول على نسبة للنشا المقاوم من نشا الأرز والذى يحتوى على نسبة 3.50% والوصول الى نسبة 28.50% آى ما يعادل 8.14 ضعف النشا المقاوم الموجود من نشا الأرز الخام وكانت أفضل الظروف للحصول على تلك النسبة بعد 16 دورة طبخ وتبريد وكان زمن الطبخ 60 دقيقة ونسبة معلق النشا للماء 1: 4 بينما أوضحت البيانات أن أفضل الظروف للحصول على أعلى نسبة من النشا المقاوم للذرة كانت 30% بعد 20 دورة طبخ وتبريد عند زمن طبخ 30 دقيقة ونسبة معلق النشا للماء 1: 5 وهذه النسبة تعادل 6.12 ضعف قدر ما تحتويه النشا الخام (4.90%) من النشا المقاوم.