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RESEARCH ARTICLE

EFFECTS OF STORAGE AND WATER ACTIVITY (a_w) ON THE CHEMICAL AND NUTRITIONAL COMPOSITION OF COCOYAM (*Colocasia exculenta*), SOYBEAN (*Glycine max*) FLOURS AND THEIR VARIOUS BLENDS

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ABSTRACT

This study evaluates the effects of storage and water activity (a_w) on the chemical and nutritional composition of cocoyam, soybean flours and their various blends. Reports on effects of water activity (a_w) which can be used to evaluate the nutritional quality of these products are at best, scanty, and in some case, apparently non-existence. The method used involved preparing three saturated solutions that gave water activities of 0.11 (LiCl), 0.33 (MgCl₂), and 0.75 (NaCl), respectively. The *Colocasia exculenta* and *Glycine max* samples were thus stored under the various water activities for 3-months and changes in the nutritional and chemical quality indices were monitored during the 3-months storage using the AOAC (2004) methods. Significant changes occurs in the chemical constituents of *Colocasia exculenta* and *Glycine max* flours (including their blends) which are traceable to factors such as a_w , blending proportions, length of storage. Important chemical changes occurred during the 3-month storage.

KEYWORDS

Chemical, *Colocasia exculenta*, *Glycine max*, Nutrients, Water activity

1. INTRODUCTION

Food nutrients are utilized for growth and development by man and provide the bulk energy, while micronutrients provide the co-factors for metabolism (Sizer and Whitney, 2013). Micronutrients are used to build and repair tissues and regulate body processes, while macronutrients are converted to smaller sugars (glucose, fructose and galactose) from carbohydrate, protein to amino acids and fats to fatty acids and glycerol (Salami, 2018). Animals have specialized digestive systems that work to break down macronutrients to provide energy and utilize micronutrients for both metabolism and anabolism (constructive synthesis) in the body. Organic nutrients consist of carbohydrates, fats, proteins (or other building blocks, amino acids) and vitamins (Eleazu et al., 2013). Inorganic chemical compounds such as dietary minerals elements and water may also be considered nutrients (Anon, 1994).

Cocoyam is rich in vitamin B6 and potassium. It contains dietary fibre and high protein contents than the majority of the tropical root crops (Braide and Nwaoguikpe, 2011).

The key benefits of soybean are its high protein content, vitamins, mineral elements and insoluble fibre. Soya bean contains vitamin k, riboflavin, foliate, vitamin B6, thiamine and folic acid (Braide and Nwaoguikpe, 2011).

Storage studies provide data base for remediation and ameliorative purposes and in diet formulations for institutional and home uses (Monday and Ukhun, 2020).

Water activity of a foodstuff refer to the degree to which the water is "bound" in the food and hence its accessibility to act as a solvent and

contribute in chemical/biological reactions and growth of micro-organism. It is a vital asset that can be used to predict the stability and safety of food with regard to rates of deteriorative reactions, microbial growth, and chemical/physical properties (DRINC, 2013).

Temperatures, pH and water activity are the factors that most control rates of spoilage and the development of micro-organisms in foods. The parameters are often referred to as hurdles. Water activity is measured as equilibrium relative humidity (ERH). The water activity (a_w) signifies the ratio of the water vapour pressure of the food to the water vapour pressure of pure water under the same equilibrium conditions and it is expressed as a fraction. If we multiply this ratio by 100, we obtain the equilibrium relative humidity (ERH) that the food stuff would produce if enclosed with air in a sealed container at constant temperature. Thus, a food with water activity (a_w) of 7.0 would produce an ERH of 70%. Water activity (a_w) has its suitability in predicting the growth of bacteria, yeasts and molds. For a food to have a suitable shelf life without relying on refrigerated storage, it is necessary to control either its acidity (pH) or the level of water activity (a_w) or a suitable combination of the two. This can successfully increase the product stability and make it possible to forecast its shelf-life under known ambient storage conditions. Food can be made safe to store by lowering the water activity to a point that will not allow dangerous pathogens such as *Clostridium botulinum* and *Staphylococcus aureus* to grow in it (DRINC, 2013).

$$a_w = \frac{P_w}{P_{w0}}$$

ERH = equilibrium relativity humidity

ERH = $a_w \times 100\%$ (Labuza et al., 1971).

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The water activity of a food is not the same thing as its moisture content. While moist foods are expected to have greater water activity than dry foods, this is not permanently so. However, a variety of foods may have precisely the same moisture content and yet have somewhat dissimilar water activities (Labuza et al., 1971).

Foods stored can be made safe by lowering the water activity to a point that will not permit hazardous pathogens such as botulinum, Clostridium and Staphylococcus aureus to grow in it. The suggested safe levels of moisture content for the period of storage of flours or food powder is between 12 – 14% (FAO, 1993). Various biochemical, besides physiological changes occur during storage of foods that could influence or affect food quality (Teede, 2004). The changes can be ameliorated by proper control of factors such as the a_w of the food.

2. MATERIALS AND METHODS

Analytical-grade Sulphuric Acid, Potassium Sulphate, Copper Sulphate, Perchloric Acid, Lithium Chloride, Magnesium Chloride and Sodium Chloride.

2.1 Instruments

Oven, Muffle furnace, Soxhlet apparatus, Kjeldahl flask, Spectrophotometer, Flame photometer, Buck scientific atomic absorption spectrophotometer (AAS) and Distillation machine and Three glass desiccators.

2.2 Preparation of Samples

Colocasia exculenta were peeled and cut into parts, and dried under sun for five days. *Glycine max* bought from New Benin Market, were also dried under the sun for five days. Both the dried *Colocasia exculenta* and *Glycine max* were milled. Wire whisk sieve of 25 μ m aperture size was used to sieve the flours to obtain smooth flours and packaged in appropriate containers (glass containers) and labelled for analyses.

3. EVALUATION OF THE EFFECTS OF WATER ACTIVITY (a_w) OF 0.11, 0.33 AND 0.75

The processed *Colocasia exculenta* and *Glycine max* products were placed inside the desiccators labelled A, B and C, where the respective a_{ws} of 0.11, 0.33 and 0.75, were established. Saturated solutions gave water activities of 0.11 (LiCl), 0.33 (MgCl₂), and 0.75 (NaCl), respectively. A 2-week equilibration period was observed. The *Colocasia exculenta* and *Glycine max* samples were thus stored under the various water activities for 3-months and changes in the nutritional and chemical quality indices were monitored during the 3-months storage using the AOAC (2014) methods. The Methodology for Proximate, Phytochemical and Mineral elements analyses conducted are as quoted in the earlier journal title Evaluation of Chemical and Nutrient Constituents of Cocoyam and Soybean Flours and their Blends in Walisongo Journal of Chemistry Vol. 7 Issue 2 (2024), 193-202 (Monday et al., 2024).

4. RESULTS AND DISCUSSION

Table 1: Effects of water Activity (a_w) on the Proximate Composition of Soybean and Cocoyam Flours.

Samples	Moisture content (%)			Crude Protein (%)			Ether Extract (%)			Ash (%)			Crude Fibre (%)			Nitrogen - Free Extract (%)			a_w
	months in storage			months in storage			months in storage			months in storage			months in storage			Months in storage			
	1	2	3	1	2	3	1	2	3	1	2	3	1	2	3	1	2	3	
Cocoyam flour	7.23 ± 1.02	7.40 ± 1.04	7.57 ± 1.12	5.30 ± 1.03	5.25 ± 1.13	4.65 ± 1.06	0.19 ± 0.02	0.18 ± 0.03	0.17 ± 0.04	2.50 ± 1.12	2.50 ± 1.21	2.00 ± 1.10	2.50 ± 1.00	2.50 ± 1.07	2.3 ± 1.20	82.28 ± 2.00	82.17 ± 2.10	83.29 ± 2.05	0.11
	$\Delta m = 0.34$			$\Delta cp = 0.65$			$\Delta EE = 0.02$			$\Delta ash = 0.11$			$\Delta cf = 05.00$			$\Delta NFE = 1.12$			
Cocoyam flour	8.95 ± 1.11	9.50 ± 1.03	14.50 ± 1.08	5.00 ± 1.04	4.25 ± 1.12	4.25 ± 1.21	0.18 ± 0.01	0.18 ± 0.11	0.17 ± 1.01	1.90 ± 1.20	2.50 ± 1.03	1.50 ± 1.01	2.50 ± 1.00	2.50 ± 1.00	2.2 ± 1.00	81.37 ± 0.06	81.07 ± 0.00	77.3 ± 1.21	0.33
	$\Delta m = 5.55$			$\Delta cp = 0.75$			$\Delta EE = 0.01$			$\Delta ash = 1.00$			$\Delta cf = 0.27$			$\Delta NFE = 4.02$			
Cocoyam flour	10.54 ± 1.21	11.25 ± 1.06	16.35 ± 1.12	4.50 ± 0.02	4.00 ± 1.20	3.50 ± 1.12	0.17 ± 0.21	0.17 ± 0.01	0.16 ± 0.04	1.80 ± 1.04	1.50 ± 0.03	1.50 ± 0.02	2.50 ± 1.01	2.50 ± 1.20	2.1 ± 1.10	80.49 ± 0.02	80.58 ± 2.10	76.43 ± 2.03	0.75
	$\Delta m = 5.81$			$\Delta cp = 1.00$			$\Delta EE = 0.01$			$\Delta ash = 0.30$			$\Delta cf = 0.35$			$\Delta NFE = 4.15$			
Soybean flour	4.65 ± 1.12	5.50 ± 1.03	7.00 ± 1.05	42.18 ± 1.12	42.0 ± 1.00	41.75 ± 1.08	22.85 ± 1.15	22.50 ± 1.23	20.50 ± 1.22	8.50 ± 1.10	8.00 ± 1.22	8.00 ± 2.01	8.03 ± 0.07	8.01 ± 1.50	8.0 ± 2.08	13.82 ± 1.09	13.91 ± 1.10	14.7 ± 1.60	0.11
	$\Delta m = 4.65$			$\Delta cp = 0.43$			$\Delta EE = 2.35$			$\Delta ash = 0.50$			$\Delta cf = 0.03$			$\Delta NFE = 0.93$			
Soybean flour	9.00 ± 1.05	13.00 ± 1.02	14.50 ± 1.07	42.05 ± 1.00	42.0 ± 1.06	41.65 ± 1.12	22.00 ± 1.13	21.50 ± 1.05	20.05 ± 1.21	7.00 ± 1.23	5.00 ± 1.30	5.50 ± 1.23	7.90 ± 1.40	7.81 ± 1.60	7.6 ± 1.80	13.16 ± 1.13	10.69 ± 1.03	10.7 ± 1.40	0.33
	$\Delta m = 5.50$			$\Delta cp = 0.40$			$\Delta EE = 1.95$			$\Delta ash = 2.00$			$\Delta cf = 0.30$			$\Delta NFE = 2.37$			
Soybean flour	9.50 ± 1.10	14.00 ± 1.30	16.43 ± 1.20	42.00 ± 1.40	41.8 ± 1.03	41.50 ± 1.06	21.00 ± 1.05	21.00 ± 1.20	19.85 ± 1.12	6.50 ± 1.14	5.00 ± 1.40	5.00 ± 1.32	7.84 ± 1.42	7.60 ± 1.09	7.3 ± 0.13	12.05 ± 1.09	10.60 ± 1.10	9.86 ± 1.20	0.75
	$\Delta m = 6.93$			$\Delta cp = 0.35$			$\Delta EE = 1.15$			$\Delta ash = 1.50$			$\Delta cf = 0.48$			$\Delta NFE = 2.19$			

Δm = Highest moisture content – lowest moisture cont.

Δcp = Highest crude protein content – lowest crude protein conts.

Δash = Highest ash content – lowest ash content crude

Δcf = highest crude – lowest crude

ΔNFE = Highest NFE – Lowest NFE

ΔEE = either extract – lowest either extract

5. WATER ACTIVITY AND MOISTURE

The trend of moisture content with respect to a_w was 0.75 > 0.33 > 0.11 a_w , as showed in Table 1. The importance of these results lies in the fact that

low water activity which in this study was associated with low moisture contents is related to reduced, hydrolysis, enzyme activity, and proteolysis. Browning reactions, some forms of oxidation in agreement with previous work done by (Labuza, 1971). Microbes also tend to thrive less at low water activity and low moisture content conditions.

5.1 Water Activity and Protein Content.

The highest crude protein value of 5.30% was obtained in the cocoyam flour kept at water activity of 0.11. Similarly, the highest protein content of 42.18% was obtained in soybean kept at water activity of 0.11. The trends of protein content was 0.11 > 0.33 > 0.75 a_w . It is discernible from the result that low a_w was more protective of the crude protein content than the higher a_w .

5.2 Water activity and Crude Fat Content

The crude fats contents of the food products did not show appreciable changes during storage at the different water activities. However, as in Table1, oxidative rancidity of fats could have occurred because of the detection of off-odour and off-flavour in samples stored at the high a_w of 0.75. The intensity of the offensive odour increased with increasing water activity. There was no offensive odour detected at the lowest water activity of 0.11. The response of lipids to changes in a_w is not as straight forward and simple as is observed with other chemical entities such as protein and carbohydrate in foods. There is usually a biphasic response. At low a_w of say 0.11, lipid oxidation is high. At of 0.33, the oxidation rate decreases and at high a_w of about 0.75, the rate spikes in agreements with previous work done by Labuza *et al.*, 1971. The trend in crude fat contents followed the order: $0.11 > 0.33 > 0.75 a_w$. While the trend in off-odour

development was: $0.75 > 0.33 > 0.11 a_w$. The implication of the results in Table1 is that the food products can best be preserved by using lower water activity storage conditions.

5.3 Water Activity and Ash, Crude fibre and NFE Values

The values for ash, crude fibre and NFE obtained as presented in Table1, show unimpressive changes. As a_w s increased, the values decreased unimpressively in the order: $0.11 > 0.33 > 0.75$. It is discernible from the results that the low a_w was more protective of the ash, crude fibre and NFE contents than the higher a_w s in the food samples.

The reduction of a_w in foods prevents the growth of vegetative microbial cells, germination of spores and toxic production by molds and bacterial as reported in literature by (Ekwe *et al.*, 2009), this is in agreement with the results presented in this study

Table 2: Effects of water activity (a_w) on the contents of phytochemicals in soybean and cocoyam flours

Samples	Total saponins content (%)			Total Phenols content ($\mu\text{g/ml}$)			Total Flavonols content ($\mu\text{g/ml}$)			Alkaloids content (%)			Total Antioxidants content ($\mu\text{g/ml}$)			Water Activities (a_w)
	Month			Month			Month			Month			Month			
	1	2	3	1	2	3	1	2	3	1	2	3	1	2	3	
Cocoyam flour	35.11± 0.21	33.73± 0.43	29.43± 0.43	70.67± 0.24	68.10± 0.35	62.57± 0.43	113.24± 0.25	72.70± 0.12	42.58± 0.43	22.20± 0.34	21.00± 0.24	20.00± 0.13	7.54± 0.12	7.27± 0.241	7.56± 0.22	0.11
Cocoyam flour	34.08± 0.11	31.26± 0.42	25.85± 0.34	31.94± 0.33	44.41± 0.24	44.38± 0.11	22.56± 0.44	32.01± 0.32	30.97± 0.21	20.00± 0.13	13.00± 0.22	15.00± 0.43	8.05± 0.34	8.09± 0.33	9.81± 0.43	0.33
Cocoyam flour	31.70± 0.23	31.17± 0.14	23.19± 0.42	28.30± 0.32	35.15± 0.23	31.19± 0.12	20.15± 0.42	20.03± 0.13	16.62± 0.42	17.60± 0.21	3.00± 0.21	12.00± 0.42	20.80± 0.43	8.97± 0.11	10.23± 0.32	0.75
Soybean flour	49.81± 0.33	47.25± 0.42	36.32± 0.34	69.52± 0.13	68.65± 0.25	90.31± 0.34	114.41± 0.43	49.62± 0.44	25.12± 0.35	21.40± 0.33	16.00± 0.34	17.00± 0.23	5.70± 0.43	7.01± 0.32	7.00± 0.42	0.11
Soybean flour	38.42± 0.25	37.42± 0.32	26.32± 0.34	45.64± 0.14	43.53± 0.42	73.38± 0.11	22.06± 0.24	21.12± 0.33	21.68± 0.22	20.56± 0.32	7.00± 0.31	9.00± 0.22	8.46± 0.34	7.70± 0.33	7.80± 0.41	0.33
Soybean flour	33.40± 0.42	33.48± 0.12	6.16± 0.43	31.80± 0.13	26.32± 0.41	24.63± 0.21	20.51± 0.34	18.60± 0.41	18.68± 0.43	17.00± 0.23	5.00± 0.12	6.00± 0.14	8.77± 0.22	7.97± 0.12	10.23± 0.23	0.75

Table 2 shows the analyses for phytochemicals during storage of the food samples, as influenced by different water activities during 3- months' storage. The results indicate that the contents of phenols, saponins and alkaloids decreased as a_w decrease in the order $0.11 > 0.33 > 0.75$. In other words, the quality and biological functions of phenols, saponins, and alkaloids are less protected at a higher a_w . This could be attributed to high moisture content available for reactive chemical reactions involving, hydrolysis, proteolysis, oxidative rancidity, browning or Maillard

reactions, and decarboxylation that may occurs in the food samples at higher a_w . Which would lead to reduction of the contents of phenols, saponins and alkaloids at the higher a_w level of 0.75. The quality and biological functions of the food samples will be less preserved at higher a_w s than at low a_w .

Correspondingly, the content of flavonoid was high at low a_w but due, obviously to degradation, it decreased at a_w of 0.33 and decreased further at 0.75.

Table 3: Effect of water activity (a_w) on the contents of mineral elements in the soybean and cocoyam flours (mg/kg)

Samples	Na			Mg			Ca			K			Fe			a_w
	Month			Month			Month			Month			Month			
	1	2	3	1	2	3	1	2	3	1	2	3	1	2	3	
Cocoyam flour	2.30± 0.21	1.40± 0.41	1.39± 0.51	3.25± 0.21	3.10± 0.30	3.18± 0.11	2.43± 0.30	1.17± 0.31	1.15± 0.61	40.50± 0.70	40.30± 0.20	26.60± 0.19	2.00± 0.76	1.60± 0.62	1.90± 0.23	0.11
Cocoyam flour	2.26± 0.51	1.20± 0.51	0.90± 0.31	3.13± 0.70	3.00± 0.50	2.14± 0.81	1.60± 0.41	1.14± 0.71	0.30± 0.21	40.00± 0.30	39.00± 0.50	31.70± 0.21	2.30± 0.68	1.21± 0.88	1.20± 0.16	0.33
Cocoyam flour	2.50± 0.61	1.15± 0.31	0.20± 0.31	3.00± 0.80	2.43± 1.50	1.12± 0.61	1.50± 0.31	1.13± 0.11	1.06± 0.01	38.00± 0.60	36.00± 0.30	28.60± 0.21	2.10± 0.23	1.00± 0.67	0.18± 0.05	0.75
Soybean flour	1.40± 0.32	1.36± 0.41	1.17± 0.12	12.00± 0.76	6.58± 1.43	5.61± 0.64	6.10± 1.63	4.89± 0.56	3.50± 0.32	110.00± 0.5	86.4± 0.31	65.20± 0.21	2.80± 0.03	2.30± 0.25	1.80± 0.01	0.11
Soybean flour	1.32± 0.61	1.17± 0.65	1.13± 0.16	11.00± 0.34	6.58± 0.70	3.56± 0.65	5.59± 0.64	3.26± 0.28	2.25± 0.36	100.00± 0.34	65.20± 0.35	62.10± 0.26	1.90± 0.50	1.52± 0.23	0.31± 0.02	0.33
Soybean flour	1.30± 0.51	1.15± 0.41	1.12± 0.43	8.00± 0.52	3.78± 0.54	2.48± 0.65	3.10± 1.63	2.14± 0.61	1.90± 0.31	95.00± 0.67	56.4± 0.31	45.20± 0.21	1.32± 0.51	1.40± 0.41	0.03± 0.01	0.75

Table 3 (cont): Effect of water activity (a_w) on the contents of mineral elements in the soybean and cocoyam flours (mg/kg)

Samples	Zn			Cu			P			a_w
	Month			Month			Month			
	1	2	3	1	2	3	1	2	3	
Cocoyam flour	0.59±0.02	0.27±0.05	0.11±0.03	1.60±0.09	0.12±0.07	0.05±0.01	23.70±1.04	8.57±0.03	4.03±0.01	0.11
Cocoyam flour	0.20±0.06	0.09±0.03	0.30±0.05	0.06±0.08	0.04±0.03	0.03±0.02	10.70±1.16	4.19±0.06	3.03±0.06	0.33
Cocoyam flour	0.18±0.07	0.08±0.03	0.30±0.04	0.08±0.02	0.02±0.01	0.01±0.46	9.26±1.03	2.10±0.23	2.02±0.01	0.75
Soybean flour	0.46±0.06	0.40±0.02	0.38±0.05	0.10±0.05	0.06±0.01	0.04±0.03	19.30±1.02	9.34±1.12	8.05±0.02	0.11
Soybean flour	0.31±0.08	0.09±0.03	0.05±0.06	0.05±0.02	0.02±0.02	0.20±0.01	17.70±1.06	8.21±1.03	7.04±0.01	0.33
Soybean flour	0.05±0.01	0.04±0.02	0.02±0.08	0.03±0.02	0.01±0.01	0.01±0.01	15.86±1.04	6.15±1.01	5.02±0.01	0.75

The result depicted in Table 3 shows that the values of mineral elements decreased as a_w levels increased. This means that, the food products were relatively stable at low a_w levels, but decreased as the water activity increased. The reductions of the mineral element contents during storage followed the order: 0.75 > 0.33 > 0.11. This may be ascribed to the fact that, at higher a_w and moisture content, various chemical reactions such as, hydrolysis, deamination, proteolysis, decarboxylation and oxidation reactions could have led to decreases in Na, Mg, Ca, K, Fe, Zn, Cu and Phosphorus contents at higher water activities as reflected in Table 3. Free-radical producing substances and transition metal ions and light, may catalyse lipid oxidation, thereby resulting to reduction of mineral elements functions as the a_w levels increases, which is in agreement with previous work done by (Labuza, 1971).

6. CONCLUSION

Statistically, significant changes occur in the chemical constituents of soybean and cocoyam flours (including their blends) which are traceable to factors such as water activity (a_w), blending proportions, and length of storage, may be used as additional methods of assessing protein qualities of cocoyam and soybean flours and their blends. It was observed that the sample kept in desiccator tagged A containing water activity 0.11(LiCl) was fresh throughout the experimental period, indicating foodstuff can be preserved for longer period using water activity 0.11(LiCl).

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AUTHOR'S CONTRIBUTION

1st author Monday I. IMAFIDON Conceived and design of the study, collected the data, performed the analysis/interpretation of data, while the 2nd author, Mark E. UKHUN contributed in the Drafting the manuscript/revisins for important intellectual context and Approval of the final version of the manuscript.

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