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Auto-hydrolytic effect on the nutrient profile of corn cob flakes treated feed with different additives

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Abstract

Corn cob, an abundant agricultural residue, can be efficiently valorized through auto-hydrolysis with additives such as enzymes or chemical solutions, which break down its complex lignocellulosic structure and enhance the accessibility of chemical and phytochemical components. This study was conducted to assess the impact of auto-hydrolysis on the nutrient profile of corn cob flakes treated with various feed additives. The feed materials were divided into pretreated groups (non-auto hydrolysis and auto hydrolysis) and subjected to six treatments using solutions of potassium phosphate and bicarbonate of soda and cellulase enzyme (Fullzyme®). The treatments were designated as follows: T0 (2 kg of corn cob flakes soaked in 1 liter of water (control), T1 (2 kg of corn cob flakes treated with 20 g of potassium phosphate per liter of water), T2 (2 kg of corn cob flakes treated with a mixture of 10 g potassium phosphate and 10 g sodium bicarbonate per liter of water), T3 (2 kg of corn cob flakes treated with 20 g of sodium bicarbonate per liter of water), T4 (2 kg of corn cob flakes treated with a mixture of 10 g sodium bicarbonate and 500 units of cellulase enzyme per liter of water), T5 (2 kg of corn cob flakes treated with a mixture of 10 g potassium phosphate and 500 units of cellulase enzyme per liter of water), and T6 2 kg of corn cob flakes treated with 500 units of cellulase enzyme per liter of water). The experiment followed a completely randomized design. The proximate analysis showed a significant increase ($P>0.05$) in CP, EE, and ash content in T6, while T4 recorded the lowest values. Conversely, a notable reduction was observed in CF, fiber fractions, and phytochemical properties in T6 compared to other treatments. It was concluded that auto-hydrolysis enhances the effectiveness of feed additives by facilitating the breakdown of lignified bonds in corn cob flakes. This process might improve nutrient availability when offered to ruminant animals particularly at an optimal combination of saline solutions (potassium phosphate or bicarbonate of soda) and enzymes.

Keywords: Auto-hydrolysis, corn cob, feed additives, chemical, phytochemical profile
Running title: Auto-hydrolytic Effect on corn cob feed

Effet de l'Auto-Hydrolyse sur le Profil Nutritionnel d'un Aliment à Base de Flocons de Rafle de Maïs Traités avec Différents Additifs

Résumé

La rafle de maïs, un résidu agricole abondant, peut être efficacement valorisée par auto-hydrolyse avec des additifs tels que des enzymes ou des solutions chimiques, qui dégradent sa structure lignocellulosique complexe et améliorent l'accessibilité des composés chimiques et phytochimiques. Cette étude a été menée pour évaluer l'impact de l'auto-hydrolyse sur le profil nutritionnel de flocons de rafle de maïs traités avec divers additifs alimentaires. Les matières premières ont été divisées en groupes prétraités (non auto-hydrolyse et auto-hydrolyse) et soumises à six traitements utilisant des solutions de phosphate de potassium et de bicarbonate de soude ainsi qu'une enzyme cellulasique (Fullzyme®). Les traitements étaient désignés comme suit : T0 (2 kg de flocons de rafle de maïs trempés dans 1 litre d'eau (témoin)), T1 (2 kg de flocons traités avec 20 g de phosphate de potassium par litre d'eau), T2 (2 kg de flocons traités avec un mélange de 10 g de phosphate de potassium et 10 g de bicarbonate de sodium par litre d'eau), T3 (2 kg de flocons traités avec 20 g de bicarbonate de sodium par litre d'eau), T4 (2 kg de flocons traités avec un mélange de 10 g de bicarbonate de sodium et 500 unités de cellulas par litre d'eau), T5 (2 kg de flocons traités avec un mélange de 10 g de phosphate de potassium et 500 unités de cellulas par litre d'eau), et T6 (2 kg de flocons traités avec 500 unités de cellulas par litre d'eau). L'expérience a suivi un dispositif complètement *aléatoire. L'analyse proximale a montré une augmentation significative (P<0,05) des teneurs en MP, EE et cendres pour T6, tandis que T4 enregistrait les valeurs les plus basses. À l'inverse, une réduction notable a été observée pour la CF, les fractions fibreuses et les propriétés phytochimiques dans T6 par rapport aux autres traitements. Il a été conclu que l'auto-hydrolyse améliore l'efficacité des additifs alimentaires en facilitant la rupture des liaisons lignifiées dans les flocons de rafle de maïs. Ce processus pourrait améliorer la disponibilité des nutriments lorsqu'ils sont offerts aux ruminants, en particulier avec une combinaison optimale de solutions salines (phosphate de potassium ou bicarbonate de soude) et d'enzymes.*

Mot-clé : Auto-hydrolyse, rafle de maïs, additifs alimentaires, profil chimique et phytochimique

Introduction

The dry season poses significant challenges for livestock due to limited feed availability, marked by a scarcity of forage and the prevalence of fibrous standing hays (Habte *et al.,* 2022; Cooke *et al.,* 2024). These dry season conditions lead to adverse effects such as weight loss, reduced milk production, and increased animal mortality. Additionally, the incidence of disease outbreaks rises during this period, as low-quality and insufficient feed weakens ruminants' immunity (Cooke et al., 2024). According to Lamidi and Ologbose, (2014) many animals succumb to starvation during the dry season due to the unavailability of year-round feed resources and the high cost of conventional feeds. This underscores the need to explore alternative, affordable, and readily available local feed ingredients. Corn waste, particularly corn cobs, emerges as a potential solution. These by-products of maize processing, whether from industrial or household sources, are often considered environmental nuisances in Nigeria, especially during harvest seasons when heaps of husks and cobs accumulate in refuse dumps or litter the surroundings (Kalio *et al.,* 2013). Despite their abundance, these residues remain highly underutilized across Africa, particularly in Nigeria (Olamilusi and Oboite, 2024), where they are frequently left on farmlands to decompose naturally through microbial activity (Okafor, 1988). However, converting these wastes into livestock feed could provide a valuable resource for ruminant production (Olamilusi *et al.,* 2019). The main limitation, however, lies in the high lignin content, the crystallinity of cellulose, and their low nutritional value and degradation rate (Sun and Cheng, 2002).

The treatment of crop residues to enhance their nutritional value has been explored since the early 20th century (Doyle *et al.,* 1990). Over time, significant efforts have focused on various treatment methods, including physical, physicochemical, chemical, and biological approaches. Faniyi (2006) emphasized the importance of pre-treating corn cobs to facilitate their utilization by ruminants. Among these approaches, combining physical and chemical methods has been noted for there effectiveness (Eniolorunda *et al.,* 2023). Auto-hydrolysis, is a widely recognized method for pretreating lingo-cellulosic materials (McMillan, 1994). This process involves treating chipped biomass with high-pressure saturated steam, followed by a rapid pressure release, causing explosive decompression. The high temperatures in this process lead to hemicellulose degradation and lignin transformation, thereby enhancing cellulose hydrolysis potential. Chemical treatments, on the other hand, involve soaking materials in acidic or alkaline solutions, such as sodium hydroxide or calcium hydroxide. For example, Sahare *et al.* (2012) demonstrated that treating corn cobs with an alkaline saline solution weakens their fiber structure. Sodium bicarbonate, commonly used as a rumen buffer supplement, has also been identified as an economical and efficient chemical pretreatment for improving anaerobic digestion processes (Paton *et al.,* 2006). Despite these advancements, the combined effect of auto-hydrolysis and chemical solutions on the nutritional value of corn cobs has not been extensively studied. This study aims to explore the auto-hydrolytic effects combined with salts and enzyme on the nutritional properties of corn cob flakes.

Materials and methods

Experimental location

The experiment was conducted at the Training and Research Farm and Animal Nutritional Laboratory of the College of Agricultural Sciences, Olabisi Onabanjo University, Ayetoro Campus, , Ogun State, Nigeria located in a deciduous-derived savannah zone of Nigeria with latitude $7^0 0^0 12$'N and longitude of $30^0 3$'E (Onakomaiya *et al.* 1992). Climate is a sub-humid tropical with an annual rainfall of 1909.30mm; it has an area of land 2087km^2 and a population of 181,826. The rainy season commences between early April and late October, maximum temperature varies during the wet season between 29^0C season and 34^0c at onset of the dry season.

Sample collections and preparation

Maize cobs were collected from the Crop Production Processing site at the Teaching and Research Farm, College of Agricultural Sciences, Olabisi Onabanjo University, Ayetoro, Ogun State, during the dry season between October and February, 2025, following harvesting of late maize . After the shelling of maize, the cobs were sun-dried to make them more brittle for grinding. The corn cobs were prepared by mechanical crushing with a thickness ranging from approximately 0.5-1.0mm and lateral dimensions of 5-8 to form a flake. The soft media (enzyme, potassium phosphate, and bicarbonate of soda) were purchased from a farmers' shop in Abeokuta, Ogun State, Nigeria. The cellulase® enzyme was measured according to the manufacturer's specifications, with 1.00g each of potassium phosphate, bicarbonate of soda, and cellulase® dissolved in 1 liter of water to create saline solutions, which were used to treat (soak) 2kg of corn cob flakes for 24hrs. The moistened corn cob flakes were transferred into airtight heated-resistant containers. Auto-hydrolysis was carried out by heating the samples in a laboratory oven at 100-121^0C for 30-60minutes. After the heating period, the containers were removed and allowed to cool naturally to room temperature. The treated samples were then carefully opened to release pressure gradually, preventing material loss or structural damage. The Auto-hydrolysed corn cob flakes were oven-dried at 60-650C until a constant weight was achieved. Drying was done to stabilize the treated material and prevent microbial deterioration during storage. The treated corn cob flakes were further stored in airtight polyethylene bags at room temperature until further chemical and phytochemical analysis.

Experimentation

The experimental feedstuff was organized into two pre-treatment groups (Non-auto-hydrolysis and Auto-hydrolysis) with six treatments prepared using solutions of inorganic salts (potassium phosphate and bicarbonate) and cellulase (Fullzyme® enzyme). These treatments were labeled as T0, T1, T2, T3, T4, T5, and T6, following a completely randomized design. The designations were as follows:

Table 1: Composition of aqueous additive solutions (g/L) used for auto-hydrolytic pretreatment of corn cob flakes

Ingredient (g/L)	T0 (control)	T1	T2	T3	T4	T5	T6
K_3PO_4	0.00	100	50.00	0.00	0.00	50.00	0.00
$NaHCO_3$	0.00	0.00	50.00	100.00	50.00	0.00	0.00
Cellulase ®	0.00	0.00	0.00	0.00	50.00	50.00	1.00
H_2O (litre)	1.00	1.00	1.00	1.00	1.00	1.00	1.00

Data collection and laboratory analysis

Proximate Composition

The ground samples were analyzed for dry matter (DM), crude protein (CP), crude fibre (CF), ash, ether extract (EE) and nitrogen free extract (NFE) contents following standard procedures as outlined by AOAC (2005).

Fibre fractions

The Neutral Detergent Fibre (NDF), Acid Detergent Fibre (ADF), Acid Detergent Lignin (ADL) was determined by the method of Van Soest *et al.* (1991).

Phytochemical Analysis

The phytochemicals present - tannin, saponin, phytate and oxalate) were determined by using the standard procedure of (Harborne, 1998; Houghton and Raman, 1998).

Statistical analysis

Data were subjected to analysis of variance and significant means were separated by Duncan multiple range tests (DMRT) using the procedure of SAS (2002).

Results and Discussion

The effect of auto-hydrolysis on the proximate composition of corn cob flakes subjected to different additive treatments is presented in Table 2. CP and ash contents were significantly influenced by the auto-hydrolytic process, whereas CF and EE were significantly affected only in the steamed samples ($p < 0.05$). Among the non-hydrolyzed samples, corn cob flakes treated with sodium bicarbonate exhibited the highest CP value, followed by those treated with a combination of potassium phosphate and sodium bicarbonate, which did not differ significantly ($p > 0.05$) from samples treated with sodium bicarbonate and enzyme. Corn cob flakes treated with potassium phosphate alone recorded the lowest CP value. With respect to pretreatment effects, enzyme-treated corn cob flakes showed the highest CP content ($p < 0.05$), followed by potassium phosphate-treated samples, while the untreated control recorded the lowest value. Crude fibre content was highest ($p > 0.05$) in the untreated control samples, followed by samples treated with potassium phosphate, potassium phosphate sodium bicarbonate combination, and sodium bicarbonate alone. A marked reduction in crude fibre was observed in corn cob flakes treated with feed additives, particularly inorganic salts and enzymes, with enzyme-treated samples exhibiting the lowest CF values. Ether extract content was highest ($p < 0.05$) in enzyme-treated corn cob flakes, followed in descending order by samples treated with potassium phosphate plus enzyme, sodium bicarbonate plus enzyme, sodium bicarbonate alone, potassium phosphate–sodium bicarbonate combination, potassium phosphate alone, and the untreated control. For ash content, enzyme-treated corn cob flakes recorded the highest values compared with other treatments, whereas the lowest ash values ($p < 0.05$) were observed in samples treated with potassium phosphate–sodium bicarbonate combination, potassium phosphate alone, and the untreated control.

The study demonstrated that auto-hydrolysis, combined with salt or enzymatic pretreatments, is a viable method for preparing corn cob flakes as ruminant feed resources, with significant effects on their nutritional composition. The present study revealed that applying auto-hydrolysis to corn cob flakes with pretreatments (either with salt solutions or enzymes) significantly impacts their nutritional properties. This aligns with findings by Liu *et al.* (2011), who reported that steam treatment in a high-pressure vessel at varying pressures enhances the nutritional quality of fibrous materials. Similarly, Patel *et al.* (2007) and Verma *et al.* (2011) emphasized that pretreating lignified feed materials induces various biochemical and nutritional transformations, including the breakdown of certain components and structural alterations in cellulosic biomass, making it more accessible to enzymes that convert fibrous material into simple carbohydrate polymers. These observations highlight the potential of using corn cob as a basal diet for ruminants, provided it undergoes appropriate treatments (physical, chemical, or a combination of both). Arieli (1997) noted that salt solution methods improve the utilization of crop residues for ruminant feeding. Furthermore, Vadiveloo (2000) identified key benefits of sodium hydroxide solution treatments, such as increased degradability and palatability of treated straw compared to untreated straw. Zimbardi *et al.* (2002) stressed the necessity of pretreating raw materials to enhance enzymatic accessibility, particularly through auto-hydrolysis. This is consistent with Owen and Jayasuriya (2009), who reported that various treatment methods for crop residues improve dry matter intake and digestibility, resulting in significant weight gain in ruminants. However, this contrast with findings by Mihiretu *et al.* (2019), who noted that using auto-hydrolysis with NaOH or ammonium hydroxide, has been developed to enhance the nutritive value of such materials.

Table 2: Effect of auto hydrolysis on the proximate analysis of corn cob flakes treated feed additives

Parameters (%)	Auto-hydrolysis	Treatment T0	T1	T2	T3	T4	T5	T6	SEM
DM	NS	56.43	57.77	62.00	59.00	61.33	59.12	59.55	0.78
	S	70.10	72.41	75.71	71.09	70.81	74.55	84.44	0.36
CP	NS	2.47^{c}	2.07^{d}	2.63^{b}	2.93^{a}	2.63^{b}	2.36^{c}	2.64^{c}	0.02
	S	2.18^{d}	2.60^{cd}	2.86^{cd}	3.18^{c}	3.57^{b}	3.88^{ab}	4.00^{a}	0.01
CF	NS	61.67	65.66	57.00	56.18	55.33	56.67	56.67	0.71
	S	46.77^{a}	41.08^{ab}	39.99^{b}	33.20^{bc}	29.21^{c}	29.08^{c}	25.18^{d}	0.75
EE	NS	0.33	0.30	0.33	0.30	0.33	0.33	0.36	0.06
	S	0.30^{d}	0.56^{cd}	0.67^{bc}	0.60^{c}	0.73^{b}	0.78^{ab}	0.84^{a}	0.05
Ash	NS	0.15^{d}	0.19^{d}	0.38^{c}	0.40^{b}	0.40^{b}	0.53^{a}	0.44^{ab}	0.03
	S	0.76^{d}	1.00^{cd}	0.95^{c}	1.40^{b}	1.56^{b}	1.68^{a}	1.67^{a}	0.05
NFE	NS	34.78	31.18	39.73	40.26	41.61	40.24	40.59	0.65
	S	49.99	55.19	54.53	55.62	57.93	61.58	63.31	0.78

^{abcd} **means alone rows with different superscript are significantly different (p<0.05)**
NS; Non-steamed, S: steamed

The effect of auto-hydrolysis on the fibre fractions of corn cob flakes subjected to different additive treatments is presented in Table 3. All fibre components evaluated were significantly influenced by the auto-hydrolytic pretreatments ($p < 0.05$). NDF content was highest in the untreated control samples, followed by corn cob flakes treated with potassium phosphate alone and those treated with a combination of potassium phosphate and sodium bicarbonate, which did not differ significantly from each other. The lowest NDF values were observed in corn cob flakes treated with sodium bicarbonate plus enzyme, potassium phosphate plus enzyme, and enzyme alone, respectively. A similar pattern was observed for ADF and ADL with higher values recorded in the untreated control and in samples treated with inorganic salts either singly or in combination. In contrast, corn cob flakes treated with combinations of inorganic salts and enzyme, as well as those treated with enzyme alone under auto-hydrolytic conditions, recorded the lowest fibre fraction values ($p < 0.05$).

The findings indicated that auto-hydrolysis as a pretreatment significantly enhances the ability of the treatment to penetrate the cellulose matrix, enabling effective degradation. This aligns with the observations of Chen *et al.* (2007), who noted that one benefit of hydrolysis is its capacity to weaken the cell walls of fibrous materials, facilitating easier access for anaerobic microbes. Similarly, Arwinsyah *et al.* (2019) reported that combining physical and chemical treatments is more effective for breaking down the lignified structure of corn cobs, simplifying their composition. Additionally, the inclusion of feed additives (50% potassium phosphate or bicarbonate of soda combined with 50% enzyme) during processing promoted fibre degradation and improved the nutritional attributes of corn cobs, thereby enhancing their suitability as a prepared feed resource. These findings are consistent with Tamminga (2011), who emphasized the role of enzymatic treatment in fibre breakdown of corn cobs. Furthermore, Qing *et al.* (2016) demonstrated that alkali-based pretreatments effectively remove lignin with minimal release of sugar degradation products and furan derivatives. Research has also confirmed that alkaline salt-based catalysts dissolve lignin and hemicellulose, de-esterify intermolecular ester bonds (Kim *et al.,* 2016), restructure lignin, and alter the crystalline state of cellulose (Geng *et al.,* 2014). These findings suggest that treating corn cobs with organic salt-based pretreatments is among the most efficient chemical approaches, offering low-pollution, non-corrosive characteristics and requiring milder chemical conditions compared to other methods.

Table 3. Effect of auto hydrolysis on fibre fraction of corn cob flakes treated feed additives

Parameters (%)	Auto - hydrolysis	T0	T1	T2	T3	T4	T5	T6	SEM
NDF	NS	68.33	66.67	66.67	65.00	64.01	62.10	60.60	0.78
	S	51.20^{a}	48.58^{ab}	49.73^{ab}	44.22^{b}	38.00^{c}	37.13^{c}	33.74^{d}	0.45
ADF	NS	63.00	63.00	62.67	57.00	55.00	55.00	48.76	0.56
	N	38.73^{a}	36.66^{ab}	31.47^{b}	31.01^{b}	30.00^{bc}	27.61^{c}	24.01^{d}	0.89
ADL	NS	57.00	59.00	56.33	48.71	47.90	46.90	46.00	0.56
	S	37.01^{a}	34.21^{ab}	32.90^{ab}	31.71^{b}	25.15^{c}	24.44^{cd}	20.10^{d}	0.60

^{abcd} means with different superscript are significantly different ($p<0.05$)
NS; Non-steamed, S; steamed

The effect of auto-hydrolysis on the phytochemical profile of corn cob flakes subjected to different additive treatments is presented in Table 4. The concentrations of saponins, tannins, and flavonoids in the treated corn cob flakes differed significantly among the treatments ($p < 0.05$). Higher concentrations of saponins, tannins, and flavonoids were observed in the untreated control samples, followed by corn cob flakes treated with potassium phosphate alone, those treated with a combination of potassium phosphate and sodium bicarbonate, and samples treated with sodium bicarbonate alone. However, corn cob flakes treated with combinations of inorganic salts and enzyme exhibited significantly lower concentrations of these phytochemicals ($p < 0.05$), with samples treated with enzyme alone under auto-hydrolytic conditions recording the lowest values.

The observed reduction in phytochemical content in corn cob flakes treated with combinations of inorganic salts and enzyme, as well as in flakes treated with enzyme alone, can be attributed to the pretreatment process, which effectively decreases the levels of saponins, tannins, and flavonoids. The results show that the combination of auto-hydrolysis with a saline solution (potassium phosphate) and enzymes enhances the breakdown of anti-nutritional compounds in corn cob. This is consistent with Ogunbayo *et al.* (2016), who found that pretreatment is crucial for altering the structure of cellulosic biomass, thereby increasing the rate and yield of enzymatic cellulose bio-conversion. To achieve this, various pretreatment methods have been developed. The reduction in phytochemical content observed in corn cob flakes treated with combinations of inorganic salts and enzyme, as well as those treated with enzyme alone, can be attributed to the pretreatment process, which effectively decreases levels of saponins, tannins, and flavonoids. This supports the findings of Lawther *et al.* (2010), who noted that hydrolysis combined with enzymes breaks down cellulosic bonds and reduces the anti-nutritional factors in corn cob. Additionally, Haydersah (2010) emphasized that mechanical, thermal, chemical, and biological processes are used to reduce anti-nutritional factors and improve nutrient bioavailability. Therefore, the significant reduction in phytochemical levels following the hydrolysis treatment of corn cob offers considerable benefits.

Table 4 Effect of auto hydrolysis on phytochemical profiles of corn cob flakes treated feed additives

Parameters (%)	Auto - hydrolysis	T0	T1	T2	T3	T4	T5	T6	SEM
Saponin	NS	93.61	88.71	86.34	84.45	84.41	81.22	74.80	0.64
	S	49.46^{a}	49.01^{a}	45.03^{b}	44.76^{b}	42.44^{c}	42.19^{c}	41.19^{c}	0.56
Tannin	NS	1.90	1.81	1.81	1.62	1.51	1.51	1.44	0.01
	S	1.85^{ab}	1.90^{a}	1.70^{b}	1.60^{bc}	1.51^{c}	1.50^{c}	1.43^{d}	0.01
Flavonoid	NS	96.81	89.61	88.82	81.67	80.81	78.81	76.81	0.33
	S	87.61^{a}	86.91^{a}	78.10^{ab}	72.11^{b}	60.11^{cd}	57.77^{d}	65.00^{c}	0.38
Phytate	NS	0.28	0.26	0.25	0.23	0.23	0.23	0.22	0.02
	S	0. 25	0.23	0.24	0.23	0.22	0.21	0.22	0.01

^{abcd} = Means on the same row with different superscripts differ significantly (P <0.05).

NS; Non-steamed, S: steamed

Conclusion and Recommendation

Corn cob, an abundant agricultural residue, constitutes a valuable lignocellulosic resource with significant potential for value addition through appropriate processing. This study shows that auto-hydrolysis effectively modifies the structural matrix of corn cob, enhancing additive accessibility and improving its chemical and phytochemical profile. The combined application of saline solutions (potassium phosphate or bicarbonate of soda) and enzymes in equal proportions further promotes the disruption of lignified bonds, supporting efficient resource upgrading while contributing to environmental waste reduction. It is therefore recommended that auto-hydrolysis tends to be an effective pre-treatment strategy for corn cob processing. The use of 50% enzyme in combination with 50% chemical treatment (salt solutions) is suggested to optimise structural modification and compositional enhancement. This approach offers a practical and sustainable pathway for crop residue valorisation, particularly during periods of high post-harvest residue availability.

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