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Research Article

Chemical and Sensory Characteristics of Cookies from Nixtamalized Pearl Millet (*Pennisetum glaucum*) and Tiger Nut (*Cyperus esculentus*) Flour Blends

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Abstract

Aside relatively low protein profile, Pearl millet utilization for food product development has been hampered by its contents of anti-nutritional factors, which can be reduced through nixtamalization. Development of Pearl millet-based food products may contribute to food security strategies in developing nations. Therefore, the quality of cookies from Nixtamalized Pearl Millet Flour (NPMF) and Tigernut Flour (TNF) blends was evaluated. NPMF and TNF were produced using the established procedures. Precisely, (5-25%) of TNF was blended with (75-95%) of NPMF using mixture, simplex lattice design (version 6.0.8), which was used for Cookies development. Proximate composition, phytochemical properties, mineral contents, and sensory attributes of the cookies were determined using standard methods. Data were analyzed with SPSS. The nutritional composition, antioxidant properties (TAC, Phenol, Flavonoid,) and sensory characteristics of Cookies were significantly improved by supplementation of TNF for NMF. However, contents of alkaloid, oxalate, tannin and phytate which are commonly referred to as antinutrients at higher doses, fell within tolerable limits. The cookie with 20%TNF was the most accepted with acceptability score of 7.23 on nine-point hedonic scale. Acceptable and nutritious cookies were developed from TNF and NPMF blends.

Keywords: *Supplementation; Functional foods; Snacks; Nixtamalization, Acceptablity.*

Introduction

Snacks refer to small, quick bites of food that are consumed between meals and are commonly recognized as an essential part of human diets and the total global production value of snack in year 2022 was estimated as $584.58 billion (Parker, 2023; Lawal *et al.*, 2024). Various eating habits occasioned by the demand at work has made snack consumption the order of the day; from banking, to manufacturing, designing, construction, lecturing and even farming (Olatoye and Arueya., 2021). However, consumption of snacks can have both positive and negative effects on overall health. On one hand, snacks such as cookies produced from cereal like millet can help regulate blood sugar levels, prevent overeating during main meals, and provide essential nutrients (Dayakar *et al.*, 2017; Irondi *et al.*, 2024). This is because millets contain bioactive components and essential minerals (Dayakar *et al.*, 2017) In addition, millets do not contain gluten and may therefore be the best choice for people with celiac disease. (Dayakar *et al.*, 2017)). However, its food application is still limited as its was known for its antinutrient properties. This content of antinutrient can be reduced through nixtamalization (Sefa-

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Dedeh *et al.,* 2003). Nixtamalization is a traditional process that involves cooking grain such as millet in lime water and then steeping it for a period. This increases the bioavailability of essential amino acids and improves the texture and flavour of the grain (Sefa-Dedeh *et al.,* 2003).in addition, by nixtamalizing millet, the phytic acid content and other antinutrients may decrease, and thereby making the minerals it contains more available for absorption by the body. Moreover, since some essential amino acids are limited in cereals generally, including the millet, supplementing it with tigernut in nutritious cookies production will provide a nutritious and healthy snack option. Tigernut is an underutilized crop which produces rhizomes from the base and tubers that are somewhat spherical. It is very popular because the tubers are directly consumed in its raw form (Adel *et al.,* 2015). It contains bioactive substances such as organic acids, alkaloids and phenols (Nina *et al.,* 2019). It is a low-cost food ingredient and possesses a relatively high amount of protein found to be suitable for diabetic patients or those with digestive dysfunctions and may prevent heart disease after consumption (Ogunlade *et al.,* 2015). Due to the presence of flavonoids, the tiger nut has good antioxidant properties. It also rich in plant oils such as oleic and linoleic acids and can be used as a source of natural antioxidants (Ogunlade *et al.,* 2015). Previous works involving the simultaneous utilization of Millet and African locust bean pulp was centered on the production of local dishes and beverages such as kunu (Olatoye *et al.,* 2023). There is dearth of literature on combined usage of nixtamalized millet and Tigernut flour blends, especially for a ready to eat food. It is envisaged that the application of nixtamalization in millet-based products may improve the nutrients digestibility and bioavailability of product. Hence, the aim of this study is to evaluate the quality of cookies from nixtamalized pearl millet and tiger nut flour blends.

Materials and Methods

Sources of Materials: Millet (*Pennisetum glaucum*) and Tigernut (*Cyperus esculentus*) were sourced from Mandate market in Ilorin and were transported to the Food Science and Technology Laboratory of Kwara State University, where the experiments were conducted.

Flour Preparation: Millet and Tigernut were sorted, washed and dried at 65 ºC for 14 hrs in a convection oven. The dried millet was dry-milled and passed through a 100-micron sieve. Millet flour at 5-25% Tigernut Flour (TNF) and 75-95% of Nixtamalised Pearl Millet Flour (NPMF) were properly blended. The Tigernut-Millet composite flours were stored in dry air-tight bags at ambient conditions for further analyses.

Preparation of cookies: Margarine (130 g) was mixed with sugar (100g) until the right consistency was achieved. The mixture was then mixed with whole milk powder (80 g), Salt (5g) and either NPMF or TNF-NPMF composite flour, (400 g), Table 1. The dough was thoroughly kneaded, shaped into uniform slices, and baked at 150 °C for 18 minutes. The cookies were cooled and stored in airtight bags at ambient conditions for further analyses.

Chemical Analyses: Proximate compositions of samples were determined using the method described by (AOAC, 2010). Samples were analyzed for moisture, crude protein, fibre, ash, fat. Carbohydrate derermination was by difference, (i.e. 100- addition of other components). Determination of Phytochemicals Contents: Flavonoid was determined using the method described by Koleva, *et al.,* (2002). Alkaloid content was determined by the alkaline precipitation-gravimetric method described by Harborne (1973).Total phenol was determined by the Folin-spectrophotometer method (AOAC, 2005) and Oxalate content was determined according to Day and Underwood (1986).Total tannins content was quantified using the methyl cellulose precipitable tannin (MCT) assay with slight modifications. Epicatechin was used as standard, and the results were expressed as grams of epicatechin equivalents per hundred grams of sample (g ECE/100 g).

Determination of Minerals: Potassium (K), Calcium (Ca), Sodium (Na), and Iron (Fe), contents were determined using the atomic absorption spectrophotometer method (AOAC, 2005).

Determination of Sensory Attributes: Hedonic test was conducted on cookies from nixtamilized pearl millet and tiger nut flour blends using untrained panelists, (n = 50). The panelist comprises of undergraduate students who are habitual snack consumers. Panelists were asked to adjudge the cookies based on degree of their preference for appearance, taste, flavor, crispness, hardness, and overall acceptability. They were asked to score them on a 9-point Hedonic scale, where 9 represented = like extremely, 8 = like very much, 7 = moderately like, 6 = like slightly, 5 = neither like nor dislike, 4 = dislike slightly 3 = dislike moderately, 2 = dislike very much and 1 = dislike extremely as described by Peryam and Pilgrim (1957) and Iwe, (2002).

Data obtained were analyzed using analysis of variance technique, and Duncan multiple range tests was used to separate the means. All generated data in triplicate, were subjected to Statistical Package for Social (SPSS Version 21) software. The means were separated, using Duncan's multiple range test ($p<0.05$).

Results and Discussion

Supplementation of TNF for NPMF significantly influenced Proximate composition of cookies. The moisture contents of the cookie samples ranged from 1.50% to 4.90% and decreased with addition of TNF (Table 2). This is beneficial as low moisture contents in foods is indicative of less susceptibility to microbial proliferation, extended shelf-storability, and biochemical spoilage (Ayo-Omogie, 2021). Crude fat however increases with increase in TNF addition and ranged between 16.67 and 22.67%. The observed increased in fat suggested possibility of contribution to calories and as well as being a vehicle of transportion of fat- soluble vitamins (Okwunodulu et al., 2019). Similarly, crude protein contents improved with increased addition of TNF in cookies formulation and ranged between 9.63 and 11.25 %. This could be attributed to the higher protein content of Tiger nut than millet. The protein contents of the cookie samples which increased with tigernut flour supplementation is a plus as protein can contribute to growth and repair of wornout tissues in the human diet (Baah, 2009). Ash contents ranged from 0.80 to 1.13% in the formulated cokies and improved as more TNF is being incorporated. The increase in ash contents of the cookie samples is attributed to the high proportion in the tigernut flour with range (0.89 – 1.57%) as reported by Dada *et al*., (2012). This may be an indication of high mineral content in such foods. The fibre contents, which ranged from 0.57 to 0.64% similarly increased with substitution level of TNF for NMF. Studies have shown that fibre reduces constipation and help reduce the risk of diseases associated with colon such as piles, appendicitis, and cancer (Galisteo *et al.,* 2008). However, the carbohydrate contents of the cookie samples varied from 63.11 to 68.43% and inclusion of tigernut flour at 5% and 10% improved their respective carbohydrate contents while further addition of tigernut flour resulted in reduction of carbohydrate contents of other cookie samples. The range of 63.11 to 68.43% carbohydrate in this study was comparable to 61.64-79.80% reported by Olatoye *et al.*, (2019) who evaluated the potentials of African Nutmeg as a flavourant in Cookie Production. Therefore, the cookies in this study may be a good energy source sequel to its high contents of carbohydrate (Ijeh *et al.*, 2010).

The phytochemical composition of cookies from nixtamalized pearl millet-tiger nut flour blends was significantly boosted with increase in substitution of TNF for NPMF (Table 3), with exceptions of Tannin and phytate. Total antioxidant capacity (TAC) ranged between 0.63 and 1.02mg/g. The enhancement of total antioxidant capacity in cookies samples would be of immense benefit from the viewpoint of consumers health. Flavonoid contents also ranged between 0.79 and 1.05mg/g and was in accordance with the range (0.4 – 1.3 mg/g) flavonoid contents of biscuit supplemented with sun and oven-dried sweet orange pulp flours reported by Akubor and Ihotu (2020). Total phenol ranged from (0.90-1.15mg/g) and was similar to (0.50 – 1.24 mg GAE/g extract) phenol contents of composite bread from wheat-moringa leave flour blends studied by Aishat *et al.* (2021). All these showcase the potential health benefit of the developed cookies in this study. On the other hands, the alkaloids contents ranged from 0.57 to 0.73mg/g was quite lower than 4.95 to 18.91% alkaloid contents of fibre-enriched cracker biscuit from wheat, lemon basil, and scent leaves powder and cashew kernel flour blends reported by Ujong *et al.* (2023). With respect to alkaloid content, a substantial proportion of alkaloid can be harmful to human health though, especially if it reaches the fatal level of 20 mg/100 g. Oxalate contents ranged between 0.15 and 0.17 (mg/g) and increases with substitution of TNF for NPMF. The results of oxalate too are lower than 0.82 – 5.34 mg/g reported for oxalate contents of chin-chin from maize-soybean-orange fleshed sweet potato by Ndife *et al.* (2020) and were within the recommended safe level of 3 to 5 mg/kg in foods (Ndidi et al., 2014). Oxalates are considered poisonous at high concentration, but harmless when present in small amounts as found in cookie samples in the present study (Chai and Liebman, 2004). The blend of tiger nut with proportional amount of NPMF significantly reduced the tannin contents of the products to the range of 0.15 to 0.48 mg/g which was in agreement with 0.11 – 0.26 mg/g reported by El-Gohery (2021) who at one time or the other differently made biscuits from wheat flour and differently proceesed lima bean. The phytate contents of the cookies ranged from 0.16 to 0.29 mg/g and was significantly lower compare to 10-60mg/g that could pose health problem to humans according to Farinde *et al.*, (2018). Nixtamalization was earlier reported to have reduce the contents of these antinutrients in similar products (Sefa-Dedeh *et al.,* 2003; Valderrama-Bravo *et al.,* 2017). Thus, the incidence of antinutrient related to oxalate and phytate will not be expectedly pronounced in this product, probably due to initial nixtamalization of millet.

Table 4 presents the mineral composition of cookies samples from nixtamalized pearl millet and tigernut flour blends. The iron contents of the cookie samples ranged from 1.62 to 2.39 mg/100 g. Cookie from 100% NPMF has the highest iron content (2.39 mg/100 g) while cookie from sample 75% NPMF has the least (1.62 mg/100 g). This may be attributed to the low iron content 0.65 – 0.80 mg/100 g in tiger nut flour (Oladele and Aina, 2007). On the contrary side, the calcium content of cookie from 75% NPMF was the highest (375.00 mg/100 g) and predominantly higher than the rest of the samples. The range of calciulm contents (276.67-375.00 mg/100 g) in the current study are higher than 25.98 – 42.08 mg/100 g for calcium contents of biscuit from acha-ripe plantain flour blends by Popoola *et al.*, (2020). Presence of calcium in diets usually help in bones and dental development and can also helps in enzymes and hormonal release (Beto, 2015). The mean results for the potassium contents of the cookie samples ranged from 316.67 to 406.67 mg/100 g. The result showed gradual increase in potassium contents of the cookies samples with increased level of tigernut flour addition. The highest potassium content (406.67 mg/100 g) was observed in 75% NPMF based cookie while 316.67 mg/100 g was observed in sample from 100% NPMF. Potassium plays vital role in maintenance of cellular water balance, it regulates the pH of the body and enhances protein and carbohydrate metabolism (Oribon *et al.,* 2007). conversily, the sodium contents of the cookie samples ranged from 170.00 to 200.00 mg/100g, with cookie from 100 % NPMF being the highest, while cookie from 75% NPMF possessing the least. Thus, the inclusion of tigernut flour resulted in reduction of sodium contents of the cookie from NPMF. The results are higher than the values 0.04 – 0.05 mg/100 g reported for sodium contents of biscuit from wheat-bambara and cowpea flour blends by Durojaiye *et al*., (2018). Low level of sodium is beneficial for consumers with high blood pressure to maintain electrolyte balance (Bamigboye and Adepoju, 2015).

The sensory characteristics of cookies from nixtamalized pearl millet and tigernut flour blends were significantly influenced ($p > .05$) with increased supplementation of TNF for NPMF, except in hardness and crispiness (Table 5). Appearance ranged between 5.53 and 6.67, and it decreases with increase in TNF. Sample with 25% TNF possess the least rating by the panelist in terms of appearance. Similar reports were documented by Olatoye and Arueya (2021), in a study, evaluation of chemical and sensory characteristics of extruded snacks from selected aerial yam cultivar and African breadfruit seed. This could be because of maillard reaction between glucose and protein components in the flour mixtures (Olatoye and Arueya 2021). In snack's sensory assessment, great emphasis is usually laid on appearance, since it provides basic information about the suitability of raw material used for its preparation, formulation and product quality (Abu-Salem and Abou-Arab, 2011;

Olayiwola, *et al* ., 2012). Taste preference increases with supplementation of TNF for NPMF up to (20%) and ranged from 5.20 to 6.93. On average, samples with 0% and 20% TNF respectively, scored the least and highest by the assessor's judgement. Perhaps, TNF contains more sugar than the NPMF. It thus appeared, that panelists prefer moderate addition of TNF to NPMF for acceptable cookie formulation. In terms of crispness of the cookies, no significant difference was observed as adjudged by the panelists. This attribute ranged between 5.60 and 6.57 with 0% and 20% TNF based samples being scored the lowest and highest by the consumer panelist. The flavor scores ranged between 5.67 and 6.77 and were significantly different ($P < .05$) among sample. This range however, clearly indicated good acceptance level for the product, since all scores ≥ 5 on 9-point Hedonic scale. In practice, the assessment of flavour is perceived as the combination of taste and aroma. According to Abu-Salem and Abou-Arab, (2011), the main criterion that determines the likeness or otherwise of a product is flavour. The developed cookie in this study was, therefore, highly liked by the panelist from flavor viewpoints, with sample having 20% TNF being liked the most. The hardness scores of the cookies as adjudged by the panelist ranged between 5.23 and 6.17 and were not significantly different ($P < .05$) between the samples. The measure of physical effort required to cause a material to break is termed hardness. In food materials, this property is usually dependent on available moisture within the food system and it provides an indication of its dryness (Olatoye and Arueya 2021). From the foregoing, one may suggest that there is a uniform degree of hardness in the cookie samples. Virtually all sample were generally accepted with overall acceptability scores ranging between 5.87 and 7.23. The highest overall acceptability score (7.23) of the cookie by the sensory evaluation panelists was for the 20% supplementation of TNF for NPMF. In addition, non of the evaluated parameters had scores below the minimum acceptable rating of 5 on a 9-point hedonic scale (Peryam & Pilgrim, 1957; Iwe, 2002). In order to evaluate the quality of food, detectable sensory attributes of food by human senses can be evaluated. This can also serve as references for selections of food. Consumers are usually the end judges of product acceptability, and it can be concluded from this view point that acceptable cookies can be developed from 20-25% substitution of TNF for NPMF.

Conclusion

A nutrient dense, bioactive compound rich and acceptable snack food (cookies) was developed from blends of NPMF and TNF. Supplementation of 5-25% TNF for NPMF significantly improved the Nutritional composition (protein, fat, ash and fibre), mineral contents (Ca and K), antioxidant properties (Total Antioxidant Capacity (TAC), Total Phenol and Flavonoid) and sensory characteristics (Appearance, Taste, crispiness, flavour, hardness and overall acceptability) of Cookies. However, the highest overall acceptability score (7.23) of the cookie by the sensory evaluation panelists was from the 20% supplementation of TNF for NPMF. This is pointing at the limit which should not be exceeded when formulatimg cookies from NPMF and TNF for optimum consumer acceptability.

Conflict of Interest Statement: Authors hereby declared no conflict of interest.

Authors' Contributions

Authours OKK represented Olatoye Kazeem Koledoye; SM: Sani maimuna; AMR: Akinpade Mayowa Racheal and LAI: Lawal Adetunji Ismael and AD: Adams Damilola. OKK, SM and LAI managed data collection, interpretation of data, writing of manuscript, material support, and review of manuscript and wrote the first draft of the manuscript. SM and AMR managed the literature searches. LAI, OKK and AMR managed the development of methodology, data analysis, and the development of the model. All authors read and approved the final manuscript.

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Table 1: Ingredients Formulation Of Nixtamilized Pearl Millet (NPMF) And Tiger Nut Flour (TNF) Blends For Cookies Production

Material used	SAZ	KYA	MZW	ADS	XYA	HLP
NPMF (g)	400.00	380.00	360.00	340.00	320.00	300.00
TNF (g)	0.00	20.00	40.00	60.00	80.00	100.00
Margarine (g)	130	130	130	130	130	130
Sugar (g)	100	100	100	100	100	100
Egg (g)	80	80	80	80	80	80
Salt (g)	5	5	5	5	5	5
Baking Powder(g)	4.8	4.8	4.8	4.8	4.8	4.8

Whole milk Powdered (g)	80	80	80	80	80	80

Source: (Akinola et al., 2014)

Table 2: Proximate Composition of Cookies From Nixtamalized Pearl Millet-Tigernut Flour Blends

Sample	NPF :TNF	Moisture (%)	Fat (%)	Ash (%)	Protein (%)	Crude fibre(%)	Carbohydrate (%)
SAZ	100:0	4.90±0.79^{a}	16.67±0.58^{d}	0.73±0.12^{b}	10.16±0.04^{d}	0.58±0.00^{d}	66.96±0.68^{b}
KYA	95:5	2.53±0.25^{b}	18.33±0.58^{c}	0.80±0.00^{b}	9.63±0.04^{f}	0.55±0.00^{f}	68.15±0.63^{ab}
MZW	90:10	2.00±0.00^{bc}	18.33±0.58^{c}	0.73±0.12^{b}	9.94±0.04^{e}	0.57±0.00^{e}	68.43±0.69^{a}
ADS	85:15	2.50±0.00^{b}	20.67±0.58^{b}	1.13±0.06^{a}	10.38±0.04^{c}	0.59±0.00^{c}	64.73±0.52^{c}
XYA	80:20	1.80±0.30^{c}	21.00±1.00^{b}	1.07±0.23^{a}	10.55±0.04^{b}	0.60±0.00^{b}	64.98±1.02^{c}
HLP	75:25	1.50±0.00^{c}	22.67±0.58^{a}	0.83±0.06^{b}	11.25±0.04^{a}	0.64±0.00^{a}	63.11±0.61^{d}
Mean	_	2.54	19.61	0.88	10.32	0.59	66.06
P level	_	***	***	**	*	*	***

Values are mean ± standard deviation of triplicate determinations. Data with different superscripts along the same column are significantly different at *p < .05, **p<0.01, ***p<0.001.

Table 3 Phytochemical Properties of Cookies from Nixtamalized Pearl Millet-Tigernut Flour Blends

Sample	NPF: TNF	TAC (mg/g)	Flavonoid (mg/g)	Phenol (mg/g)	Alkaloid (mg/g)	Oxalate (mg/g)	Tannin (mg/g)	Phytate (mg/g)
SAZ	100:00	0.91±0.01^{b}	0.79±0.03^{e}	1.03±0.01^{c}	0.67±0.01^{b}	0.15±0.00^{c}	0.48±0.00^{a}	0.27±0.00^{b}
KYA	95:5	0.70±0.01^{d}	0.99±0.01^{b}	1.10±0.01^{b}	0.73±0.02^{a}	0.16±0.00^{b}	0.30±0.00^{c}	0.19±0.01^{c}
MZW	90:10	0.75±0.01^{c}	0.83±0.01^{d}	0.99±0.02^{e}	0.57±0.00^{d}	0.16±0.00^{b}	0.43±0.00^{b}	0.29±0.00^{a}
ADS	85:15	0.63±0.00^{e}	0.86±0.00^{c}	1.15±0.01^{a}	0.59±0.01^{d}	0.17±0.00^{a}	0.29±0.01^{d}	0.19±0.00^{c}
XYA	80:20	1.02±0.01^{a}	1.05±0.01^{a}	1.01±0.01^{d}	0.73±0.01^{a}	0.17±0.00^{a}	0.15±0.00^{e}	0.16±0.00^{d}

HLP	75:25	0.76±0.03^{c}	0.89±0.02^{c}	0.90±0.01^{f}	0.61±0.01^{c}	0.17±0.00^{a}	0.15±0.00^{e}	0.19±0.00^{c}
Mean	------	0.79	0.91	1.07	0.68	0.16	0.30	0.23
P level	------	**	**	*	***	***	***	***

Values are mean ± standard deviation of triplicate determinations. Data with different superscripts along the same column are significantly different at *p < .05, **p<0.01, ***p<0.001

Table 4: Mineral Composition (mg/100 g) of Cookies From Nixtamalized Pearl Millet-Tigernut Flour Blends

Sample	NPMF:TNF	Iron	Calcium	Potassium	Sodium
SAZ	100:0	2.39±0.02^{a}	372.50±2.50^{a}	316.67±28.87^{d}	200.00±0.00^{a}
KYA	95:5	2.37±0.01^{a}	368.33±1.44^{b}	350.00±0.00^{c}	190.00±0.00^{b}
MZW	90:10	2.12±0.02^{b}	329.17±2.89^{c}	380.00±10.00^{abc}	190.00±0.00^{b}
ADS	85:15	2.10±0.02^{b}	327.50±2.50^{c}	365.00±25.89^{bc}	180.00±0.00^{c}
XYA	80:20	1.78±0.02^{c}	276.67±2.89^{d}	393.33±2.89^{ab}	180.00±0.00^{c}
HLP	75:25	1.62±0.01^{d}	375.00±0.00^{a}	406.67±2.89^{a}	170.00±0.00^{d}
Mean	_	2.06	341.53	368.61	185.00
P level	_	***	***	***	***

Values are mean ± standard deviation of triplicate determinations. Data with different superscripts along the same column are significantly different at ***p<0.001.Key: SAZ = 100% nixtamalized pearl millet flour.

Table 5: Sensory Characteristics Of Cookies From Nixtamalized Pearl Millet And Tigernut Flour Blends.

Sample	NPMF:TNF	Appearance	Taste	Crispiness	Flavor	Hardness	Overall Acceptability
SAZ	100:0	6.40±2.01^{ab}	5.20±1.56^{c}	5.60±1.87^{a}	5.67±1.77^{b}	5.27±2.36^{a}	5.87±1.76^{b}
KYA	95:5	5.93±1.53^{ab}	6.30±1.58^{ab}	5.67±1.83^{a}	6.03±1.94^{ab}	5.43±2.03^{a}	6.33±1.56^{ab}
MZW	90:10	6.67±1.67^{a}	6.17±1.32^{ab}	6.10±1.52^{a}	6.07±1.57^{ab}	5.60±1.81^{a}	6.43±1.55^{ab}
ADS	85:15	6.20±1.56^{ab}	6.10±2.04^{ab}	6.17±1.70^{a}	5.68±2.11^{b}	6.03±1.96^{a}	6.77±1.72^{ab}

XYA	80:20	6.53 ± 1.68^{a}	6.93 ± 1.51^{a}	6.57 ± 2.14^{a}	6.77 ± 1.94^{a}	6.17 ± 2.09^{a}	7.23 ± 1.77^{a}
HLP	75:25	5.53 ± 1.48^{b}	5.97 ± 1.27^{b}	5.93 ± 1.41^{a}	6.13 ± 1.55^{ab}	5.23 ± 1.69^{a}	6.40 ± 1.22^{ab}
Mean		6.21	6.11	6.01	6.06	5.62	6.5
P level		NS	***	NS	NS	NS	NS

Values are mean ± standard deviation of triplicate determinations. Data with different superscripts along the same column are significantly different at *p < .05, **p<0.01, ***p<0.001