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Anti Diabetics And Antioxidant Potentials of Whole Wheat Bread Substituted With Lagos Red Spinach Flour and Extract (*Celosia Argentea*)

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ABSTRACT

This study assesses the possibilities of incorporating Lagos spinach (*Celosia argentea*) into wheat bread recipes to improve its nutritional, functional, and sensory characteristics. The study meets the demands created by wheat dependency and current public health problems such as diabetes and obesity. Bread was made from wheat flour substituted with dry Lagos spinach flour, 10-50%, and its aqueous extract, 230-300ml were analyzed using standard methods for proximate composition, physicochemical and sensory properties, antioxidant capacity, and anti-diabetic potential. Results showed that with increased spinach substitution, there is an increase in protein (17.60 – 30.10 g/100g), fiber (1.69 -1.86 g/100g), ash (3.33 – 3.80 g/100g), and antioxidant levels; a reduction in carbohydrate content and glycemic index was also observed with a reduced. The bread also showed increased α -amylase and α -glucosidase inhibition, hence a potential for diabetes management. The sensory studies showed that the acceptability reduced from 6.83 ± 1.64 in 100% wheat bread to 3.60 ± 1.81 in 50% spinach substituted wheat bread due to changes in texture and color but are nutritionally superior to 100% wheat flour bread. This infers that Lagos red spinach is a sustainable ingredient for functional food production, providing the consumers with nutritionally value added bread that can confer health benefits.

Keywords: Wheat, bread, diabetes, spinach.

Introduction

Bread, known as a global staple meal is distinguished for its substantial nutritional value, comprising vital components like protein, vitamins, minerals, fiber, and carbs, while exhibiting low fat and cholesterol levels (Petchsomrit *et al.*, 2020). Bread is a quintessential medium for integrating food-grade fractions from grains, legumes, and native food sources to improve its functional and nutritional attributes (Wada and Kassahun, 2019).

The tender leaves and shoots of Lagos spinach contain high amounts of nutritious components such as

protein, calcium, phosphorus, and iron, representing a great addition to diets. Incorporation of Lagos spinach into bread making may greatly improve the nutritional content of the bread while harnessing its functional advantages.

Previous study has shown the potential of leafy vegetables to enhance the quality, antioxidant, and physicochemical characteristics of bread (Smith *et al.*, 2024). Various studies involving the use of powders from vegetables such as *Amaranthus viridis* (green amaranth), *Solanum macrocarpon* (African eggplant) and *Telfairia occidentalis* (fluted pumpkin) in bread-making have shown changes in chemical composition and qualitative attributes (Odonlade *et al.*, 2017). While antioxidant qualities such as total

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phenolic content and DPPH inhibition exhibited a drop with higher supplementation, the research indicated that such bread varieties might acquire broader appeal with greater knowledge owing to their potential health advantages. Lagos spinach has been highlighted for its high antioxidant content, particularly vitamin C and flavonoids, which are responsible for its ability to preventing deterioration, neutralize free radicals and prolonging its shelf life. Its inclusion may significantly boost bread's texture, nutritional profile and sensory quality (Smith *et al.*, 2024; Smith, 2019).

This work establishes a precedent for the incorporation of indigenous green vegetables in bread making, promoting better nutritional alternatives while tackling public health challenges such as obesity, cardiovascular illnesses and diabetes.

Materials and Methods

Material

Lagos red spinach and other baking ingredients were purchased from Shasha market in Akure Ondo state. Whole wheat flour was obtained from Golden Penny Flour Mills, Lagos, Nigeria. All chemicals and reagent used were analytical grade.

Methods

Lagos red spinach flour and extract: The fresh red spinach was trimmed, washed, oven dried at 60 °C for 7 h, pulverized and packaged ready for use while 250g of another set of fresh vegetable was trimmed, washed and boiled with 500ml water for 5minutes. It was drained and the aqueous red spinach extract obtained was used in baking the bread.

Bread production: Standard method was used and baking done at 200 °C.

The formulations for blending Red Lagos Spinach Flour with whole wheat flour at various proportions of 0% (control) through to 50% is as stated below:

RLS 1 – 100% Wheat Flour

RLS 2- 90% Wheat flour, 10 % Red Lagos Spinach flour& 250 ml extract

RLS 3- 80% Wheat flour, 10% Red Lagos Spinach

flour & 250 ml extract

RLS 4 – 70% Wheat flour, 30 % Red Lagos spinach flour & 250ml extract

RLS 5 – 60% Wheat flour, 40% Red Lagos Spinach flour & 260 ml extract

RLS 6 – 50 % Wheat flour, 50% Red Lagos Spinach flour & 300ml extract

Analysis

Proximate composition of the bread was done using the AOAC, 2012 standard method and sensory evaluation was done on a 9-point Hedonic scale with 30 semi-trained panelists. Color determination was done with a Hunter colorimeter (Konica Minolta CR 410 chromameter) according to the method reported by Lui-Ping *et al.*, (2005).

The Glycemic Index (GI) was done as described by Ijarotimi *et al.*, 2021. The α -amylase and α -glucosidase activities were assayed as described by Smith *et al.*, (2019).

Bioactive properties: ABTS Radical Scavenging Ability was determined according to the method of Re *et al.*, (1999). Total phenolic content was determined by using the Folin-Ciocalteu method (Singleton *et al.*, 1999). The ferric-reducing antioxidant power of the extracts was assayed by the method of Pulido *et al.*, (2000).

Statistical Analysis: All analysis were done in triplicate and result were subjected to ANOVA using SPSS version 2001 USA while the means were separated using Multiple Range Test.

Results and Discussion

Proximate Composition of Whole Wheat Bread Substituted with Red Lagos Spinach Flour and Extract.

Table 1 shows the proximate composition of whole wheat bread substituted with Red Lagos Spinach Flour. The moisture content in the various samples ranged between 25.46% and 36.70%, with RLS 6 having the highest moisture content value at 36.70%

Table 1: Proximate Composition (g/100g) of veggie substituted whole wheat bread with Red Lagos Spinach flour and extract

Samples	Moisture	Total Ash	Crude fat	Crude fibre	Crude protein	Carbohydrate
RLS 1	34.20 ^b ± 0.01	2.60 ^c ± 0.01	7.12 ^b ± 0.01	1.65 ^c ± 0.02	16.60 ^f ± 0.07	37.76 ^c ± 0.09
RLS 2	32.84 ^c ± 0.10	1.37 ^f ± 0.01	7.17 ^b ± 0.04	1.69 ^{bc} ± 0.01	17.60 ^e ± 0.21	38.44 ^b ± 0.27
RLS 3	25.46 ^e ± 0.16	1.44 ^e ± 0.02	8.06 ^a ± 0.05	1.71 ^b ± 0.05	21.23 ^d ± 0.03	42.08 ^a ± 0.07
RLS 4	31.26 ^d ± 0.04	3.33 ^b ± 0.01	8.09 ^a ± 0.03	1.76 ^b ± 0.02	21.85 ^c ± 0.14	33.69 ^d ± 0.16
RLS 5	32.73 ^c ± 0.10	2.24 ^d ± 0.02	8.11 ^a ± 0.02	1.82 ^a ± 0.01	30.10 ^a ± 0.07	24.97 ^e ± 0.12
RLS 6	36.70 ^a ± 0.02	3.80 ^a ± 0.07	8.13 ^a ± 0.05	1.86 ^a ± 0.03	28.20 ^b ± 0.07	22.23 ^f ± 0.02

Values are presented as mean ± SD (n=2). Values with the same superscript in the same column are not significantly different at p<0.05.

KEYS:

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RLS 6 – 50 % Wheat flour, 50% Red Lagos Spinach flour & 300ml extract.

while RLS 3 gave the lowest value with 25.46% with significant difference at p<0.05. The relative high moisture content of RLS 6 may be indicative of a softer and tenderer crumb. It may also indicate better fermentation, which enhances the flavor and nutritional profile of the bread. However, the high moisture content in the vegetable wheat bread of RLS sample may heightened the risk of microbial growth. However, substitution with Lagos red spinach significantly improved the nutritional values of the resultant bread showing increase in crude protein, total ash, crude fibre, and decrease in carbohydrate, comparable to Adepeju *et al.*, 2025 but however lower than the values obtained from bread fortified with soy flour (Olaoye *et al.*, 2006; Natal *et al.*, 2013).

Sensory Evaluation of Whole Wheat Bread Substituted with Red Lagos Spinach Flour and Extract.

The sensory analysis of the whole wheat bread substituted with Red Lagos Spinach flour and extract

are as stated in Table 2. Sample RLS 2 (5.53) was the most preferred and the least was RLS 6 (6.83). The acceptability decreases as level of substitution increases. p<0.05. The crust appearance were becoming darker as level of substitution increases. The scoring pattern were the same for the parameters. Polyphenols, strong grassy and sulphurous odor of spinach maybe due to dimethyl sulphide, methanethiol and dimethyl trisulphide had shown to have effect on the bread acceptability (Díaz-Mula *et al.*, 2017; Prasad *et al.*, 2025).

Glycemic Indices of Whole Wheat Bread Substituted with Red Lagos Spinach Flour and Extract.

The glycemic indices (GI) results of whole wheat bread substituted with Red Lagos Spinach flour and its extract is as stated in Table 2. RLS 6 had the lowest value for GI at 40.56% while RLS 1 had the highest value for GI at 60.17%. The lower the GI of food, the better for diabetic patients. The glycemic

Table 2: Sensory Attributes of veggie substituted whole wheat bread with Red Lagos Spinach flour and extract

Samples	Crust color	Aroma	Taste	Texture	Crumbiness	Overall Acceptability
RLS 1	7.23 ^a ± 1.48	6.77 ^a ± 1.55	6.53 ^a ± 1.57	6.30 ^a ± 1.34	6.23 ^a ± 1.78	6.83 ^a ± 1.64
RLS 2	5.53 ^b ± 1.72	5.30 ^{ab} ± 1.82	4.80 ^{ab} ± 1.77	5.53 ^a ± 1.68	5.57 ^a ± 1.83	5.53 ^b ± 1.71
RLS 3	4.87 ^b ± 1.94	4.97 ^{ab} ± 1.75	4.20 ^{ab} ± 1.65	4.90 ^{ab} ± 2.04	5.23 ^a ± 2.14	4.73 ^b ± 1.79
RLS 4	4.30 ^{bc} ± 1.86	4.47 ^{ab} ± 1.66	4.03 ^{ab} ± 1.40	4.83 ^{ab} ± 1.91	5.37 ^a ± 2.19	4.33 ^b ± 1.48
RLS 5	3.63 ^b ± 2.03	4.77 ^{ab} ± 2.05	3.17 ^b ± 1.51	4.30 ^{ab} ± 1.84	4.97 ^a ± 2.22	3.80 ^b ± 1.81
RLS 6	3.77 ^b ± 2.30	2.93 ^b ± 1.72	4.30 ^{ab} ± 1.98	3.53 ^b ± 0.36	5.23 ^a ± 2.06	3.60 ^b ± 2.06

Values are presented as mean ± SD (n=2). Values with the same superscript in the same column are not significantly different at p<0.05.

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RLS 6 – 50 % Wheat flour, 50% Red Lagos Spinach flour & 300ml extract

index (GI) of whole wheat bread 60.17%. Addition of Red Lagos Spinach Flour and extract was able to lower the value to 40.56 in RLS 6 (50% substitution). The reduced values for the RDS (Rapidly Digestible Starch), SDS (Slowly Digestible Starch), and RS (Resistant Starch) also shows that the starch is not easily broken down. This might be as a result of phenolic compounds in Lagos spinach slowing down starch digestion, inhibit starch hydrolysis thus reducing the glycemic response (Lawal *et al.*, 2022) hence it will not easily cause a spike in blood sugar making it ideal food for diabetic patient.

α- amylase and α- glucosidase enzyme of Whole Wheat Bread Substituted with Red Lagos Spinach Flour and Extract.

α-amylase and α-glucosidase enzymes in the small intestine break down complex carbohydrates and sugars into simpler sugars (maltose, dextrins) and glucose. α-glucosidase inhibitors block these enzymes, preventing the complete breakdown and absorption

of carbohydrates (Corkovi'c *et al.*, 2022). This leads to a more gradual increase in blood glucose levels after eating, rather than a sharp spike. Both α-amylase and α-glucosidase inhibitory activities increased in the bread samples with increase in spinach flour substitution significantly (p<0.05) thereby lowering the rate at which carbohydrate were broken down. It was 1.35 and 1.19 for α-amylase and α-glucosidase respectively RLS 6 which was 50% substitution. The bread may therefore have the capability to regulate postprandial blood glucose.

Antioxidant Capacity of Whole Wheat Bread Substituted with Red Lagos Spinach Flour and Extract.

Table 5 shows the antioxidant capacity of whole wheat bread substituted with Red Lagos Spinach flour and its extract. DPPH, ABTS and Phenolic compound had their highest values (6.89.55% 35.20mMol/g and 19.13mg/g respectively) in Sample RLS 6 which is 50% substitution. The trends indicated that as the

Table 3: Glycemic indices (%) of veggie substituted whole wheat bread with Red Lagos Spinach flour and extract.

Samples	RDS	SDS	RS	GI
RLS 1	24.53 ^b ± 0.61	19.83 ^a ± 0.49	19.83 ^a ± 0.48	60.17 ^a ± 1.49
RLS 2	27.53 ^a ± 0.68	14.11 ^b ± 0.35	14.11 ^c ± 0.35	47.04 ^b ± 1.16
RLS 3	23.16 ^{bc} ± 0.76	13.11 ^b ± 0.49	13.11 ^d ± 0.49	45.55 ^b ± 0.61
RLS 4	23.23 ^{bc} ± 0.78	18.88 ^a ± 0.78	18.88 ^a ± 0.78	44.11 ^c ± 0.01
RLS 5	23.86 ^b ± 0.38	8.51 ^c ± 1.49	18.51 ^{ab} ± 1.49	42.84 ^d ± 0.08
RLS 6	24.45 ^b ± 1.79	16.37 ^b ± 0.11	16.37 ^b ± 0.11	40.56 ^d ± 0.75

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RLS 6 – 50 % Wheat flour, 50% Red Lagos Spinach flour & 300ml extract

RDS – Rapidly Digestible Starch,

SDS – Slowly Digestible Starch

RS – Resistant Starch,

GI -Glycemic Index

Table 4 α- amylase and α- glucosidase (mg/g) indices of veggie substituted whole wheat bread with Red Lagos Spinach flour and extract

Samples	α-amylase	α-glucosidase
RLS 1	1.18 ^c ± 0.01	1.49 ^f ± 0.02
RLS 2	1.20 ^{bc} ± 0.02	1.56 ^e ± 0.03
RLS 3	1.23 ^b ± 0.01	1.69 ^d ± 0.01
RLS 4	1.24 ^b ± 0.02	1.76 ^c ± 0.01
RLS 5	1.31 ^a ± 0.02	1.87 ^b ± 0.02
RLS 6	1.35 ^a ± 0.01	1.91 ^a ± 0.01

Values are presented as mean ± SD (n=2). Values with the same superscript in the same column are not significantly different at p<0.05.

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RLS 6 – 50 % Wheat flour, 50% Red Lagos Spinach flour & 300ml extract

Table 5: Antioxidant Capacity of veggie substituted whole wheat bread with Red Lagos Spinach flour and extract

Samples	DPPH (%)	ABTS (mMol/g)	Phenolic mg/g
RLS 1	67.15 ^f ± 0.49	13.71 ^e ± 0.45	10.24 ^e ± 0.01
RLS 2	85.30 ^e ± 0.14	28.65 ^d ± 0.07	11.40 ^d ± 0.02
RLS 3	86.30 ^d ± 0.28	31.85 ^c ± 0.07	12.97 ^c ± 0.01
RLS 4	87.40 ^c ± 0.14	33.70 ^b ± 0.57	12.98 ^c ± 0.01
RLS 5	88.35 ^b ± 0.35	33.50 ^b ± 0.14	15.74 ^b ± 0.04
RLS 6	89.55 ^a ± 0.21	35.20 ^a ± 0.21	19.13 ^a ± 0.01

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DPPH – 2,2 – Diphenyl-1-picrylhydrazyl

ABTS - 2,2¹ – azino-bis (3-ethylbenzothiazoline-6-sulfonic acid)

FRAP – Ferric reducing ability

Table 6. Color analysis of veggie substituted whole wheat bread with Red Lagos Spinach flour and extract

Samples	L*	a*	b*	c*	h*
RLS 1	59.51 ^a ± 0.39	11.83 ^a ± 0.02	17.40 ^a ± 0.08	21.04 ^a ± 0.09	55.79 ^e ± 0.78
RLS 2	45.93 ^b ± 0.15	9.05 ^b ± 0.09	17.63 ^a ± 0.12	19.82 ^b ± 0.14	62.82 ^d ± 0.09
RLS 3	36.58 ^c ± 0.35	6.48 ^d ± 0.03	14.39 ^c ± 0.05	15.78 ^c ± 0.05	65.77 ^b ± 0.08
RLS 4	35.64 ^c ± 0.10	6.00 ^f ± 0.04	15.06 ^b ± 0.29	16.22 ^d ± 0.29	68.26 ^a ± 0.27
RLS 5	34.63 ^d ± 0.01	7.39 ^c ± 0.04	15.76 ^b ± 0.11	17.41 ^c ± 0.11	64.88 ^c ± 0.04
RLS 6	32.22 ^e ± 0.04	6.19 ^e ± 0.03	15.32 ^b ± 0.14	16.53 ^d ± 0.14	67.98 ^a ± 0.12

Values are presented as mean ± SD (n=2). Values with the same superscript in the same column are not significantly different at p<0.05.

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RLS 6 – 50 % Wheat flour, 50% Red Lagos Spinach flour & 300ml extract

L* – lightness, **a*** –Green to Red axis, **b*** – Blue to Yellow axis, **c***- chroma, **h***– hue angle

increase of substitution increased, the DPPH, ABTS and phenolic compound also have higher values. This pattern follows the trend reported by Junejo *et al.*, (2021). The high values of the antioxidants indicates that the bread samples have a strong antioxidant capacity that can effectively neutralize free radicals and reduce oxidative stress.

Color Analysis

The color analysis of the bread becomes darker as the substitution of whole wheat flour with red spinach flour increases. The Lightness value decreased significantly at $p < 0.05$ from 59.51 in 0% to 32.22 in 50% level of substitutions. This trend was also reported by Prasad *et al.*, (2025) in spinach enriched bread. This actually affected the acceptability of the bread produced as the acceptability followed the trend of lightness.

Conclusion

It can be concluded that the resultant bread produced from the whole wheat substituted with Lagos red spinach flour and the extract had better nutritional qualities compare to the control which was 100% whole wheat flour. It was high in antioxidant properties, α - amylase and α - glucosidase activities and has better antidiabetic potential. It may therefore be capable of preventing or mitigating degenerative diseases and managing diabetes.

References

AOAC. (2012). Association of Official Analytical Chemist. Official Methods of Analysis of the Analytical Chemist International, 19th ed. Gathersburg, MD USA.

Eyasu, W., Tileye, F., and Kassahun T., (2019). Proximate, Mineral and Antinutrient Contents of Cocoyam (*Xanthosoma sagittifolium* (L.) Schott) from Ethiopia. *International Journal of Food Science*, (1), 8965476.

Ijarotimi, O. S., Fatiregun, M. R., and Oluwajuyitan, T. D (2022). Nutritional, antioxidant and organoleptic properties

of therapeutic-complementary-food formulated from locally available food materials for severe acute malnutrition management. *Bulletin of the National Research Centre* 46 (1) 39.

Odunlade, O., Adeodu, O., Owa, J., and Obuotor, E. (2019) Iron deficiency: still a rarity in children with sickle cell anemia in Ile-Ife, Nigeria. *Hematology, Transfusion and Cell Therapy* 41 (3), [216-221](#)

Petchsomrit, A., McDermott, M.I., Chanroj, S., and Choksawangarn W (2020). Watermelon seeds and peels: fatty acid composition and cosmeceutical potential. *OCL* 27: 54

Re, R., Pellegrini, N., Proteggente, A., Pannala, A., Yang, M., and Rice-Evans, C. (1999). Antioxidant activity applying an improved ABTS radical cation decolorization assay. *Free radical biology and medicine* 26 (9-10), 1231-1237.

Singleton, V. L., Orthofer, R. Lamuela-Raventós, R. M (1999). Analysis of total phenols and other oxidation substrates and antioxidants by means of folin-ciocalteu reagent. *Methods in enzymology* (299),152-178.

Smith, J., and Lee, R. (2024). Bread enrichment using local vegetables: A focus on sensory Vegetables. *Journal of Baking Science*, 34(2), 211-225.

Smith, R. A., Andrews, K. S., Brooks, Durado, S., Fedewa, A., Manassaram-Baptiste, D., Saslow, D., and Wender, R. C. (2019). Cancer screening in the United States, : A review of current American Cancer Society guidelines and current issues in cancer screening. *CA: a cancer journal for clinicians* 69 (3), [184-210](#).