Journal of Nutrients
2022 Vol. 8, No. 1, pp. 1-8.
ISSN(e): 2410-6542
ISSN(p): 2413-8428
DOI: 10.18488/87.v8i1.3141

ANTI-OXIDATIVE AND SENSORY PROPERTIES OF RICE COOKED WITH *THAUMATOCCOCCUS DANIELLI* LEAF EXTRACTS

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Article History
Received: 20 July 2022
Revised: 30 August 2021
Accepted: 13 September 2021
Published: 28 September 2022

Keywords
Antioxidant
Extracts
Phenolic
Rice
Sensory
Thaumatococcus danielli.

ABSTRACT

The use of leaf for wrapping food is an ancient practice. Wrapping of cooked rice with *Thaumatococcus daniellii* have been reported to increase consumers' appeal and functionality. There is dearth of information on exploring the use of *T. daniellii* leaf extract as an additive during rice cooking. This study evaluated the antioxidant and sensory properties of rice cooked with the inclusion of *Thaumatococcus danielli* leaf extract. Cold extraction of leaf was done using hexane, ethyl acetate, methanol, and water. The rice samples cooked with different *Thaumatococcus danielli* leaf extracts were analyzed for antioxidant and colour properties using standard methods. Sensory evaluation of cooked rice samples using 9-point hedonic scale was carried out by thirty-member panelists. The antioxidant properties determined were total phenolic content (0.37- 0.44 mg/g), Total flavonoid content (6.6-7.9 mg/100g), 1,1-diphenyl-2-picrylhydrazyl (29.96 -39.82%), total antioxidant capacity (3.57-4.71 mg/g), ferric reducing antioxidant power (3.93-4.37 mg/100g). The instrumental lightness (L*) measurement of cooked rice ranged from 44.13 to 53.95, redness and greenness (a*) ranged from -4.91 to -3.02, yellowness – blue (b*) ranged from 13.22 to 15.94. Sensorial assessment showed that rice cooked with water extract of *T. daniellii* leaf was the most acceptable. There was significant difference ($p<0.05$) in antioxidant, colour and sensory properties of cooked rice samples. *Thaumatococcus daniellii* leaf extracts increased the antioxidant properties of cooked rice and imparted a higher sensorial acceptability.

Contribution/Originality: The study explored the effect of inclusion of *Thaumatococcus daniellii* leaf extracts on cooked rice samples. The work established the most suitable extraction solvent for *T. daniellii* leaf. It was discovered that *T. danielli* leaf extracts increased the antioxidant and sensory acceptability of cooked rice samples.

1. INTRODUCTION

Numerous traditional practices employed in food processing have been identified to impart natural nutrients and organoleptic benefits on food. An example of such practice is the use of *Thaumatococcus danielli* leaves to wrap foods such as rice, beans pudding and pap among others [1]. *Thaumatococcus danielli* (*Benn*) benth is a perennial monocotyledeon, self propagatory plant that belong to the marantacea family [2]. The leaf of *T. danielli* is used extensively in cooking and wrapping food both for domestic use and commercial enterprise. Its uses transcend the confines of the rural dwellers as evidenced by its uses for special packaging of rice by a prominent and well spread urban-based fast food restaurant [3].

According to Ukwubile, et al. [4] *T. daniellii* is of wide application in taste modification and flavor enhancement of food and drinks. This has been attributed to the presence of an intensely sweet protein known as thaumatin, which is the major component responsible for flavor modification, and development of sweetness in foods and drinks [5]. Toxicology studies established the safe consumption of the leaf extracts of *T. daniellii* [1, 6, 7]. Previous researches also reported its antimicrobial potential [2, 8].

Rice consumption as cooked grains is a common practice all over the world due to the absence of any known allergic problem [9]. The nutritional composition of rice in terms of macronutrient made it to be highly sought for daily consumption, as rice contain 80% carbohydrate, 7-8% protein, 3% fat and 3% fibre [10, 11] . However, the process of milling and polishing eliminates greater percentage of the micronutrients including its antioxidant properties [12, 13]. The presence of antioxidants in rice enhances human health [12]. Simply due to the potential of antioxidants to scavenge free radicals that causes damage to human health. This also leads to delay to some types of cell damage. Hence, the continuous research to improve on the percentage constituent of dietary antioxidant [14].

The uttilization of leaf for food packaging has been reported to increase food functionality and appeal to consumer. Previous findings by Hamid, et al. [8] and Ukwubile, et al. [4] indicated that *T. daniellii* is a rich source of antioxidant. Wrapping of cooked rice with *T. daniellii* have been reported to increase consumers' appeal [1, 15]. However, the use *T. daniellii* leaf extract has not been assessed for its impact on rice sensory and antioxidative properties. Therefore this research examined the anti-oxidative and sensory properties of rice cooked with *Thaumatococcus* danielli leaf extract.

2. MATERIALS AND METHODS

2.1. Materials

Fresh leaf of *Thaumatococcus danielli* was purchased from Oje market in Ibadan, Oyo State South west of Nigeria. The medium rice grain (royal stallion®) and salt were purchased from Sango market, Saki, Oyo state Nigeria. All chemicals used were of laboratory standard.

2.2. Preparation of Thaumatococcus Leaf Extracts

The method of Adeogun, et al. [3] was used with slight modification. The fresh leaf of *Thaumatococcus danielli* was first washed to remove adhering soil particles. The cleaned leaves were grounded using hand operated attrition mill. Two hundred gram of grounded leaves was soaked with solvent (distilled water, ethyl acetate, methanol and hexane) in different conical flasks plugged with cotton wool, and manually shaking intermittently for seventy two hours. The extract was filtered through a Whatman No. 1 filter paper. The filtrate was evaporated and the extract kept at 4 °C in refrigeration until further use.

2.3. Preparation of Cooked rice with T. daniellii Leaf Extracts

Rice (500 g) was weighed into five separate bowls, 5g of salt was weighed for each of the rice separately and 0.5 g of the leaf extracts (Hexane, methane, ethyl acetate and water extract) was weighed for each of the rice, except for the control sample without the extract. The rice was washed to remove any adhering soil particles. Water (250 ml) was boiled before the addition of cleaned rice to parboil for eight minutes. The parboiled rice was poured into a sieve, drained and rewashed to reduce the starchy substance in the rice. The parboiled rice was cooked with the addition of water (250 ml) and salt (5 g) for five minutes before the addition of *T. daniellii* extracts (0.5 g). Thereafter, the rice was allowed to cook for about seven minutes.

2.4. Determination of Color Properties of Cooked Rice Samples

The color properties was measured using Chroma meter CR 410 Konica Minolta, sensing Inc; Japan. This was done by placing the cooked rice samples on a petri-dish and the battery powered Chroma meter equipment was used to capture the sample. The triplicate data read by the Chroma meter for each sample included the L, a, b, ΔL, Δa, Δb, and ΔE.

2.5. Determination of Antioxidant Properties of Cooked Rice

2.5.1. Extraction of Cooked Rice Samples

A slight modification of the method adopted by Tugli, et al. [16] was used to obtain extracts for Total Phenolic Content (TPC), Total Flavonoid Content (TFC), Total Antioxidant Capacity (TAC), FRAP and DPPH analysis. Briefly, 1 g of cooked rice samples was mashed and extracted with 20 mL of 70% ethanol for 48 hours by intermittent agitation. The mixture was filtered using Whatman No 1 filter paper. The filtrate was kept at 4°C in a refrigerator for subsequent analysis of antioxidant properties.

2.5.2. Determination of Total Phenolic Content of Cooked Rice

The folin ciacolteu assay [17] was used to determine the total phenolic content of cooked rice samples. The assay involved the addition of folin ciocalteu reagent (2.5 mL) and 7% (w/v) of Sodium carbonate (2 mL) to 0.5 mL of the cooked rice extract. The mixture was allowed to react for 1.5 h before reading the absorbance at 765nm on spectrophotometer. Gallic acid was used as standard and the TPC was estimated from the standard curve obtained.

2.5.3. Determination of Total Flavonoid Content of Cooked Rice

The total flavonoid content of cooked rice samples was evaluated by the Saikia, et al. [18] method. Briefly, the cooked rice extract (0.25 mL) was diluted with 1.25 mL distilled water followed by the addition of $NaNO_3$ solution (75 µL). The resulting mixture was incubated for 6 min before the inclusion of 10% $AlCl_3$ (150 µL) followed by 5 min of incubation. Thereafter, 1 M Sodium hydroxide (0.5 mL) was added to the mixtures and made up to 3 mL before reading at 510 nm on the spectrophotometer. Quercetin was used to construct the calibration curve which estimated the TFC concentration in mg/100g.

2.5.4. Determination of Total Antioxidant Content of Cooked Rice

The phosphomolybdum assay reported by Dutta, et al. [19] was used to determine the total antioxidant capacity of cooked rice samples. The mixture of sulphuric acid (3.3 mL), Sodium phosphate (335 mg) and Ammonium molybdate were dissolved in 100 mL of distilled water to produce the phosphomolybdenum reagent. Boiling of the mixture 0.1 mL of rice extract and the phosphomolybdenum reagent at 95 °C for 90 minutes was done. Absorbance of the resulting mixture was read at 695 nm. Standard curve was constructed from the absorbance readings of different concentration of gallic acid. The total antioxidant capacity was estimated in mg/g from the standard curve.

2.5.5. Determination of 1,1diphenyl-2-Picrylhydrazyl (DPPH) of Cooked Rice Samples

The radical scavenging potential of cooked rice extract was determined by the DPPH radical scavenging assay reported by Turkoglu, et al. [20]. Briefly, 0.004% DPPH solution was prepared. Cooked rice extract (0.1 mL) was mixed with DPPH reagent (p.3 mL) and kept in the dark for 30 min of the resulting mixture was taken at 516nm. The DPPH reagent without the sample extract was used as control.. The percentage inhibition of the extract was calculated using Equation 1.

$$Percentage\ inhibition = \frac{Ac - Ae}{Ac} \times 100 \qquad (1)$$

Where Ac= Absorbance of control.
Ae= Absorbance of extract.

2.5.6. Determination of Ferric Reducing Antioxidant Power (FRAP)

The ferric reducing antioxidant power for cooked rice was determined by the method of Sukrasno, et al. [21]. The FRAP reagent was prepared by the mixture of Acetate buffer, 2,4,6-Tris(2-pyridyl)-s-triazine (TPTZ) and $FeCl_3.6H_2O$ in the ratio 10:1:1, respectively. Distilled water (0.7 mL) was used to dilute 0.3 mL of cooked rice extract, followed by the addition of FRAP reagent (2.85 mL). The reacting mixture was kept for 20 minutes at 50 ^{o}C before reading its absorbance at 700 nm. Ascorbic acid was used to prepare the standard curve for the estimation of antioxidant power of cooked rice samples.

2.6. Sensory Evaluation

A thirty untrained member panelist was used to evaluate the cooked rice samples with respect to appearance, mouth feel, taste, aroma, color and overall acceptability on a 9 point hedonic scale. The panelists were made to rinse their mouth and wait for five minutes before the assessment of the next sample.

2.7. Statistical Analysis

The data reported from this study was average of triplicate observation. The result was analyzed using SPSS, version 20.00. The mean and standard error of mean of the triplicate analysis was calculated. The analysis of variance was performed in determining significant differences between the mean ($P<0.05$). The mean was separated using the Duncan Multiple Range Test (DMRT).

3. RESULTS AND DISCUSSION

3.1. Colour Properties of Rice Cooked with Thaumatococcus Danielli Leaf Extract

Cooked rice colour is an important parameter that determines its level of acceptability by consumers. The colour of cooked rice samples was determined by Chroma meter (CR 410 konica Minolta sensing inc, Japan), which displayed the L*, a*, b*, ΔL, Δa, Δb and ΔE values. The L*, a* and b* values represented lightness, redness and yellowness, respectively. The inclusion of *T. daniellii* extracts caused a significant ($p<0.05$) variation in the colour properties of cooked rice samples. As presented in Table 1, the rice sample cooked with methanolic extract of *T. daniellii leaf* had the highest lightness (L=53.95) and yellowness (b=15.94). The lowest value of lightness and yellowness was from sample cooked with ethyl acetate extract. This could be due to the observed extraction of deep greenish pigments by ethyl acetate and hence reduction in lightness of the cooked rice.

Table 1. Colour properties of rice cooked with *Thaumatococcus danielli* leaf extract.

Sample	L	a	b	ΔL	Δa	Δb	ΔE
RHX	46.57 ± 0.01^b	-4.16 ± 0.01^b	13.46 ± 0.03^b	28.63 ± 0.01^b	-7.73 ± 0.01^b	11.76 ± 0.02^b	31.9 ± 0.01^b
REA	44.13 ± 0.01^a	-4.91 ± 0.01^a	13.22 ± 0.02^a	26.21 ± 0.00^a	-8.49 ± 0.01^a	11.54 ± 0.01^a	29.86 ± 0.01^a
RMT	53.95 ± 0.01^e	-3.92 ± 0.01^c	15.94 ± 0.00^e	36.02 ± 0.01^e	-7.49 ± 0.01^c	14.26 ± 0.00^e	39.46 ± 0.01^e
RWC	48.77 ± 0.01^c	-3.28 ± 0.01^d	14.55 ± 0.00^c	30.84 ± 0.01^c	-6.86 ± 0.00^d	12.87 ± 0.00^c	34.11 ± 0.01^c
RWW	50.76 ± 0.01^d	-3.02 ± 0.03^e	15.09 ± 0.03^d	32.83 ± 0.01^d	-6.59 ± 0.03^e	13.42 ± 0.04^d	36.06 ± 0.01^d

Note: Mean with different superscript on a column are significantly different ($P<0.05$). Value are mean of sample ± standard deviation of the triplicate determination
RHX = rice with hexane extract; REA = rice with ethyl acetate extract; RMT = rice with methanol extract; RWC = rice with water extract; RWW = control sample.

Generally, there was significant difference ($p<0.05$) in lightness, redness and yellow colouration among all samples of cooked rice. This showed the strong influence of the type of solvent used for the extraction of *T. daniellii*

. This could be due to the polarity and potential of solvents to extract variable colour compounds from *T. daniellii*. Previous report stated that different type of anthocyanins and pro-anthocyanidins were the phytochemicals responsible for the diverse colour of pigmented rice, and that these were widely distributed in different concentrations in the coloured rice cultivars [22].

3.2. Antioxidant Properties of Rice Cooked with Thaumatococcus Danielli Leaf Extract

In this present study, antioxidant component of rice cooked with *T. daniellii* , leaf extract evaluated were the total phenolic content and total flavonoid content while the antioxidant power was explored by the total antioxidant capacity, 1,1diphenyl-2-picrylhydrazyl (DPPH) and FRAP.

3.3. Total Phenolic Content

The potential of phenolic compounds in cell enhancement, inhibition of new blood vessel development that causes tumour growth, alongside attenuating adhesiveness and invasions of cancer cells, which help to reduce its metastatic potential have severally been reported [22, 23]. The total phenolic content of the cooked rice samples ranged from 0.37mg/g to 0.44mg/g Table 2. The least total phenolic content was obtained from control sample (Rice cooked without *T. daniellii* extract). The extracts of *T. daniellii* leaf increased the TPC of cooked rice samples. Although, the result obtained from rice cooked with ethyl acetate extract was not significantly different from the control sample at $p<0.05$. Rice samples cooked with water extract of T. daniellii leaves had the highest TPC followed by hexane and methanolic extract respectively. Contrary result was reported by Tugli, et al. [16], they recorded the lowest value of TPC for water extract of sorghum bicolor leaves.

Table 2. Antioxidant properties of rice cooked with *Thaumatococcus danielli* leaf extract.

Sample	Flavonoid mg /100g	TPC mg /g	TAC Mg/g	FRAP mg/100g	DPPH %
RHX	7.9 ± 0.15^{c}	0.42 ± 0.01^{c}	4.31 ± 0.21^{b}	4.37 ± 0.07^{c}	37.56 ± 0.40^{c}
REA	7.44 ± 0.23^{bc}	0.39 ± 0.01^{ab}	3.8 ± 0.02^{a}	4.26 ± 0.12^{c}	33.3 ± 0.41^{b}
RMT	6.9 ± 0.19^{ab}	0.41 ± 0.01^{bc}	4.71 ± 0.05^{c}	4.10 ± 0.22^{b}	35 ± 0.90^{b}
RWC	$7\pm0..42^{ab}$	0.44 ± 0.00^{c}	3.81 ± 0.01^{a}	4.13 ± 0.9^{b}	39.82 ± 0.52^{d}
RWW	6.6 ± 0.09^{a}	0.37 ± 0.00^{a}	3.57 ± 0.04^{a}	3.93 ± 0.1^{a}	29.96 ± 0.16^{a}

Note: Mean with different superscript in the column are significantly different ($P<0.05$). Value are mean of sample ± standard deviation of the triplicate determination
RHX = rice with hexane extract; REA = rice with ethyl acetate extract; RMT = rice with methanol extract; RWC = rice with water extract; RWW = control sample.

3.4. Total Flavonoid Content

Flavonoid are potent water soluble antioxidant and free radical scavenging which prevent oxidative cell damage, have strong anticancer activity and protect against the different levels of carcinogenesis. The presence of flavonoid in the intestine lowers the risk of heart diseases [24]. The flavonoid content of all rice cooked with *Thaumatococcus danielli* leaf extracts were higher than the control sample. However, the flavonoid content of the water and methanolic extracts of cooked rice samples were not significantly different. As presented on Table 2, rice cooked with hexane extract of *T. daniellii* leaf had the highest flavonoid content. Only the flavonoid content of rice cooked with hexane and ethyl acetate extracts of *T. daniellii leaf* were significantly different from the control samples. This inferred the suitability of extraction of flavonoid content of *T. daniellii* leaf by hexane and ethyl acetate. The fact that methanol and water extracts cooked rice samples yielded the lowest flavonoid content could be due to weak interaction of *T. danielli* leaf with the extracting solvents. This interaction depended on the polarity of the solvent used for extraction [25].

The result obtained in this work agreed with the work of Tugli, et al. [16] that reported comparatively lower values of flavonoid content for methanolic and water extracts of sorghum bicolor leaves.

3.5. Total Antioxidant Capacity

The total antioxidant capacity assay is based on the reduction of Mo (vi) to Mo (v) by the extract and subsequent formation of green phosphate Mo (v) complex of acid pH. The result of TAC presented in Table 2, indicated that the extracts of *T. daniellii* had significant effect on cooked rice antioxidant capacity. The TAC of cooked rice samples ranged from 3.57 to 4.71 mg/g. The highest TAC was obtained from rice cooked with methanolic extract of *T. daniellii*. The TAC of rice cooked with ethyl acetate and water extract of *T. daniellii* leaf were not significantly different at $p<0.05$. Antioxidant capacity is the primary measurement to evaluate the state and potential of oxidative stress in aging and other age related diseases [14].

3.6. DPPH Scavenging potential of Cooked Rice Samples

The cooked rice samples free radical scavenging activities against the DPPH radical is presented in Table 2. The scavenging effect of rice cooked with *T. daniellii* extracts (irrespective of solvent used) was higher than the control sample. The rice sample cooked with water extract of *T. daniellii* had the highest scavenging activity against DPPH radical. There was no significant difference ($p<0.05$) in the percentage inhibition of DPPH radicals of rice cooked with ethyl acetate and methanolic extract of *T. daniellii* . Low DPPH inhibition by rice cooked with methanolic extract agreed with the work of Hamid, et al. [8] that reported a low DPPH for methanolic extract of *T. daniellii* leaf. The highest DPPH 39.82% was found in sample rice cooked with water extract. While the lowest was recorded to be 29.98% RWW (control samples). This result agreed with the work [26] that reported a DPPH radical scavenging activity of 27.4 % for Carnaroli variety of rice.

3.7. FRAP (Ferric Reducing Antioxidant Power) of Cooked Rice

The Ferric Reducing Antioxidant Power assay is often used to evaluate the ability of antioxidant agent to denote electron. The ferric reducing ability of the cooked rice sample is a measure of its antioxidant activity. All the rice samples cooked with *T. daniellii* extracts had significantly ($p<0.05$) higher ferric reducing ability when compared to the control sample. The highest ferric reducing activity was obtained from rice sample cooked with hexane extracts of *T. daniellii*, and this was not significantly different from rice cooked with ethyl acetate extract. It was also observed that there was no significant difference between rice cooked with methanolic and water extract of *T. daniellii*. Generally the reducing properties are associated with the presence of compound which exert their action by breaking the free radical chain by denoting a hydrogen atom [21].

3.8. Sensory Properties of Rice Cooked with Thaumatococcus Danielli Leaf Extract

The sensory properties of rice cooked with *Thaumatococcus danielli* leaf extract are shown in Table 3. Attributes such as appearance, mouthfeel, aroma, taste and colour were evaluated by panelist ranged between 7.1 to 8.1, 7.1 to 8.03, 7.07 to 8.23, 7 to 8.1 and 7.4 to 8.2 respectively.

Table 3. Sensory properties of rice cooked with *Thaumatococcus danielli* leaf extract.

Sample	Appearance	Mouthfeel	Aroma	Taste	Colour	Overall aceptability
RHX	7.23 ± 0.23^{ab}	7.27 ± 0.19^{ab}	7.37 ± 0.19^{ab}	7.23 ± 0.22^{a}	7.67 ± 0.22^{ab}	7.9 ± 0.16^{ab}
REA	7.1 ± 0.16^{a}	7.1 ± 0.15^{a}	7.07 ± 0.20^{a}	7 ± 0.18^{a}	7.4 ± 0.21^{a}	7.57 ± 0.14^{a}
RMT	7.77 ± 0.16^{bc}	7.63 ± 0.16^{bc}	7.7 ± 0.19^{bc}	7.87 ± 0.18^{b}	8.13 ± 0.14^{b}	8.24 ± 0.11^{bc}
RWC	8 ± 0.21^{c}	7.77 ± 0.20^{bc}	7.9 ± 0.18^{bc}	8.03 ± 0.18^{b}	8.2 ± 0.17^{b}	8.47 ± 0.12^{c}
RWW	8.1 ± 0.19^{c}	8.03 ± 0.18^{c}	8.23 ± 0.17^{c}	8.1 ± 0.19^{b}	8.2 ± 0.16^{b}	8.38 ± 0.14^{c}

Note: Mean with different superscript in the column are significantly different ($P<0.05$). Value are mean of sample $\pm$ standard deviation of the triplicate determination
RHX = rice with hexane extract; REA = rice with ethyl acetate extract; RMT = rice with methanol extract; RWC = rice with water extract; RWW = control sample.

The sensory ratings for the control sample were not significantly different from rice cooked with methanolic and water extracts of *T. daniellii*. Rice sample cooked with water extract of *T. daniellii* had the highest rating with respect to overall acceptability. There was no significant difference in the sensory rating of rice cooked with hexane and ethyl acetate extracts of *T. daniellii*. The low appearance rating observed in sample REA (rice with ethyl acetate extract) could be as a result of leaching of some colorant from the leaf extract into the rice during cooking altering the colour.

4. CONCLUSION

Cooking rice with the extracts of *T. daniellii* significantly increased the antioxidant properties of cooked rice. However, rice cooked with methanolic and water extracts of *T. danielli* had significantly higher antioxidant properties when compared to the control sample. The incorporation of *T. daniellii* during rice cooking significantly caused variation in the instrumentation assessment of the colour of cooked rice. Sensory panelist adjudged rice cooked with water extracts of *T. daniellii* most acceptable. The addition of *T. danielli* leaf extract to rice during cooking has a potential for higher sensory acceptability and reduction in the development of disease caused by oxidative stress.

Funding: This study received no specific financial support.
Competing Interests: The authors declare that they have no competing interests.
Authors' Contributions: All authors contributed equally to the conception and design of the study.

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