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Antioxidant and Anti-inflammatory Activities of Methanolic Pulp Residue Extract of African Star Apple (*Chrysophyllum albidum*)

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Abstract: This present study evaluated the *in vitro* antioxidant and anti-inflammatory activities of methanolic pulp extract of *C. albidum* fruits to determine its use in treatments of oxidative and inflammatory-related conditions. Fresh fruits of *C. albidum* were sliced and squeezed to expel the juice. Then the pulp residues were dried in an oven for twenty-four hours at 40°C and pulverized. The powdered sample (76.5 g) was extracted in 80% (v/v) methanol for 72 h and filtered. Filtrate was evaporated *in vacuo* to obtain the methanolic extract. *In vitro* evaluations of the antioxidant (via 1,1-diphenyl-2-picryl hydroxyl (DPPH), hydroxyl radical scavenging, lipid peroxidation, total antioxidant capacity (TAC), ferric reducing antioxidant power (FRAP) assays) and anti-inflammatory activities (via anti-proteinase, anti-denaturant, anti-haemolytic and membrane stabilizing assays) were carried out based on standard methods. Also, the total flavonoids and phenolics contents were determined. From the results, *C. albidum* fruit pulp had DPPH and hydroxyl radical scavenging activities of 50.97 ± 1.18 and 23.38 ± 2.59% and inhibited lipid peroxidation to 60.17 ± 1.24%. The TAC was 272.94 ± 11.15 mg/g AAE while the IC_{50} for FRAP was 0.34 ± 0.01 µg/ml. The percentage inhibitions of proteinase, denaturation, and hemolytic activities were 65.32 ± 4.57, 40.00 ± 0.87, and 15.79 ± 1.34 %, respectively, while the percentage membrane stability was 67.55 ± 6.80 %. The total flavonoid and phenolic contents were 35.31 ± 0.74 mg/g QE and 59.64 ± 0.52 mg/g TAE. In conclusion, the pulp residue of *C. albidium* fruits possessed strong antioxidant and anti-inflammatory properties, which could be linked to the presence of flavonoids and phenols. Therefore, consuming the fruit pulp residue may enhance the health benefits of *C. albidium*.

Keywords: *Chrysophyllum albidum*; antioxidant; anti-inflammatory; anti-denaturant; proteinase.

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1. Introduction

Fruits are dietary sources of fiber, antioxidants, vitamins, minerals, etc. [1]. They help in decreasing the risks of cardiovascular, cancer, and diabetic diseases. Some of the vitamins and minerals provided by fruit sources include vitamins A, C, E, and B, while minerals include phosphorus, magnesium, and zinc. In addition to reducing the blood levels of homocysteine, a major player in coronary disease, folic acid also promotes cellular growth and reproduction [2].

The *C. albidum* (family Sapotacea) is found in the tropics like Nigeria, Benin Republic, Togo, Ghana, etc. The fruit is seasonal and becomes available from December to April. It is

called Agbalumo (Yoruba), Udara (Igbo) and Ehya (Igala) [3]. The fruit pulp is widely consumed because of its several ethnomedicinal benefits, while the seed is discarded [4]. After five days of harvesting, the fruits begin to deteriorate at room temperature. To reduce losses, the fruits have been investigated for suitability in making drinks and jams [5].

The *C. albidum* fruits contain higher amounts of vitamin C than oranges, cashew, and guava [6]. It also contains other vitamins, iron, food flavor, fat, carbohydrates, and minerals such as sodium, magnesium, and potassium [7]. The seeds are good sources of oil with several benefits [8]. The roots, barks, fruit pulp, and seeds of *C. albidum* are natural remedies for sprains, bruises, and wounds. The high saponin content of *C. albidum* leaves and roots y their used in controlling human cardiovascular diseases and treating cholesterolemia [9].

The *C. albidum* bark is used for treating fever, malaria, fibroid, etc. *C. albidum* bark leaves are used for treating stomach aches, diarrhea, skin cancer, etc. [10]. The high phenolic contents make it ideal as an antiseptic, antifungal antibiotic [11]. Therefore, this work aims to investigate the antioxidant and anti-inflammatory potential of *C. albidum* to employ the plant in managing oxidative and inflammatory disorders.

2. Materials and Methods

2.1. Fruit sample collection.

C. albidum fruits were obtained at Akoda Ede, Osun State, Nigeria. This was identified at Ife Herbarium, Department of Botany, Obafemi Awolowo University, Ile-Ife, Nigeria.

2.2. Fruit sample preparation.

The fruit samples were sliced and juice squeezed. Pulp residue was oven dried at 40^{o}C for 24h and pulverized. Then 76.5g were extracted in 80% methanol for 72h, filtered, and evaporated *in vacuo* to obtain the methanolic extract.

2.3. Collection and preparation of bovine erythrocyte (RBC).

Whole bovine blood from Araromi abattoir, at Ile-Ife, was collected into an anti-coagulant vial and rocked to mix. The blood sample was centrifuged at 3000 rpm for 15 min to separate the blood plasma. The supernatant was carefully removed, and the packed erythrocytes were washed by re-suspending in an equal volume of n-saline and centrifuged. The procedure was repeated until a clear supernatant was obtained. Then, 2% (v/v) erythrocyte suspension was prepared with n-saline.

2.4. Quantitative phytochemical screening for phenols and flavonoids.

2.4.1. Determination of total phenolic content of *C. albidum* fruit pulp extract.

The total phenolic content of *C. albidum* fruit pulp was described by Platzer *et al*. 2021 [12]. The standard curve was prepared using varying volumes of tannic acid (0, 200, 400, 600, 800, 1000 µl) made to 1000 µl using distilled water. After that, 1500 µl each of 7.5% sodium carbonate and Folin Ciocalteu reagent were added and allowed to stand in the dark for 90 min. The absorbance was measured at 725 nm against the blank and plotted against individual concentrations to generate a standard curve. After that, phenolic content was extrapolated and expressed as TAE mg/g extract.

2.4.2. Determination of total flavonoid content of *C. albidum* fruit pulp extract.

Total flavonoid was carried out using quercetin as standard [13]. Typically, varying volumes of quercetin (0.0, 0.2, 0.4, 0.6, 0.8, 1.0 ml) were measured and made up to 2 ml using clean distilled water. Then, 300 µl of 5% sodium nitrate, 10% aluminum chloride, and 2000 µl of 4% sodium hydroxide were added and allowed to stand at room temperature for ten minutes. The absorbance was read at 500 nanometers against blank. The procedure was followed for methanolic extract. The same process was followed for the pulp extract of *C. albidum*. The concentration of flavonoids in the extract was extrapolated as described above.

2.5. Antioxidant assays.

2.5.1. DPPH radical scavenging.

This was determined following the standard method [14]. Different volumes (0.0, 0.2, 0.4, 0.6, 0.8, 1.0 ml) of the extract prepared in methanol were pipetted separately and adjusted to 1.0 ml with methanol. This was followed by adding 1000 µl DPPH (0.3 mM). After incubation in the dark for thirty minutes, absorbance was measured at 517 nanometres against blank. The same process was followed for L-ascorbic acid as standard.

$$\% \text{ activity} = \frac{A-B}{A} \text{ x } 100$$

where A = Control absorbance; B = Test sample absorbance

2.5.2. Total Antioxidant Capacity (TAC).

The method was based on standard procedure [15]. Various volumes (0.0, 20, 40, 60, 80, 100 µl) of L-ascorbic acid (150 µg/ml) were measured and adjusted to 300 µl using distilled water. This was followed by adding 3000 µl of TAC reagent (0.6 M sulphuric acid, 28 mM sodium phosphate, and 4 mM ammonium molybdate) and incubating at 95°C for ninety minutes. After that, absorbance was measured at 695 nanometres against a blank. The same process was repeated using 300 µl of the test sample (1 mg/ml).

2.5.3. Ferric Reducing Antioxidant Power (FRAP).

The procedure was described by Manzocco *et al*., 2020 [16]. Various volumes (37.5, 75, 150, 225, 300 µl) of the test sample (1 mg/ml) were adjusted to 1000 µl with distilled water. Following the additions of 2500 µl phosphate buffer pH 6.6 (0.2 M) and 1% potassium ferricyanate, the mixture was incubated at 50°C for twenty minutes. After that, 2500 µl of 10% TCA were centrifuged at 3000 rpm for ten minutes. Then, 2500 µl of the supernatant, distilled water, and 0.1% (w/v) $FeCl_3.6H_2O$ were added, and absorbance was measured at 700 nanometres against phosphate buffer blank. The same procedure was followed for the L-ascorbic acid standard.

2.5.4. Hydrogen peroxide radical scavenging.

The procedure was based as described by Reddy *et al*. 2021 [17]. Different volumes (0.2, 0.4, 0.6, 0.8, 1.0 milliliter) of test samples were made up to 1.0 ml using distilled water. After 500 µl phosphate buffer (20 mM, pH 7.4) and 400 µl H_2O_2 (2 mM) were added and allowed to stand for five minutes at room temperature, 5% dichromate acetic acid (2.0 ml) was

added, and absorbance was measured at 570 nanometers. The procedure was repeated for the ascorbic acid standard (1 mg/ml).

$$\% \text{ inhibition of hydrogen peroxide} = \frac{Absorbance\ of\ test - Absorbance\ of\ standard}{Absorbance\ of\ test}\ x\ 100$$

2.5.5. Lipid peroxidation assay.

This was based on standard procedure [18]. Varying volumes (0.0, 0.2, 0.4, 0.6, 0.8, 1.0 ml) of the test sample were adjusted to 1000 µl with distilled water. This was followed by the addition of KCl (0.15 M, 1.0 ml), egg yolk (0.5 ml), and 0.2 mM $FeCl_3$ (100 µl). After incubation at 37ºC for thirty minutes, ice-cold HCl (2.0 ml) was added and incubated at 80ºC for 1 h. Thereafter, it was centrifuged at 3000 rpm, and absorbance was measured at 532 nm. The procedure was carried out for the ascorbic acid standard.

$$\text{Percentage inhibition} = (A\text{-}B)/A \times 100$$

Where A = Test sample absorbance; B = control absorbance

2.6. Anti-inflammatory assays.

2.6.1. Anti-proteinase assay.

This was carried out following the method of Bonsou *et al*., 2022 [19]. Varying volumes (0.0, 0.1, 0.2, 0.3, 0.4, 0.5 ml) of Ibuprofen (500 µg/ml) were made up to 500 µl with distilled water. Then, 500 µl of trypsin (0.06 mg) prepared in 0.5 ml of tris buffer (20 mM, pH 7.4) was added and allowed to stand at room temperature for five minutes, after which 500 µl of 0.8% (w/v) casein was added. After allowing it to stand at room temperature for twenty minutes, 1000 µl of 70% (v/v) perchloric acid was added to terminate the reaction process. After centrifuging for fifteen at 3000 rpm, 500 µl supernatant was added to a mixture of 500 µl alkaline copper reagent and 10% (v/v) Folin-Ciocalteau's reagent (1.0 ml). After incubating at 55ºC for ten minutes, absorbance was measured at 650 nm. The same procedure was repeated for the methanolic extract.

$$\% \text{ inhibition of proteinase} = \frac{Absorbance\ of\ control - Absorbance\ of\ test}{Absorbance\ of\ test}\ x\ 100$$

2.6.2. Anti-denaturant assay.

The procedure was based on the method described by Islam *et al*., 2021 [20]. Varying volumes of the test sample (0.0, 0.2, 0.4, 0.6, 0.8, 1.0 ml) and Ibuprofen (1 mg/ml) were adjusted to 1000 µl with distilled water. Then, 500 µl of bovine serum albumin was added, pre-incubated at 37ºC for 20 minutes, and heated at 57ºC for 30 minutes. This was cooled, then 2500 µl phosphate buffer (0.5 M, pH 6.3) was added, heated at 55ºC for ten minutes, and cooled. After that, the absorbance was measured at 660 nm. The same procedure was repeated for the methanol extract.

$$\% \text{ of Albumin denaturation} = \frac{Quantity\ of\ protein\ left}{Total\ protein}$$

2.6.3. Bovine red blood cell membrane stabilizing assay.

The procedure was described by Morakinyo *et al*., 2020 [21]. Varying volumes (0.0, 0.15, 0.30, 0.45, 0.60, 0.75, and 0.90 ml) of extract were made up to 1.0 ml using isosaline. After that, 500 µl hyposaline, 500 µl phosphate buffer (pH 7.4, 0.15M), and 500 µl of 2% RBC were added and heated at fifty-six degree Celsius for thirty minutes and then centrifuged at 2500 rpm for 5 min. Absorbance was read at 560 nm. The same procedure was followed for Ibuprofen, the standard drug.

2.6.4. Anti-haemolytic assay.

The assay used Ibuprofen as a standard drug [22-25]. Varying volumes (0.0, 0.2, 0.4, 0.6, 0.8, 1.0 ml) of the test sample were made up to 1 ml with distilled water. After adding 500 µl of RBC (2%) and 500 µl of phosphate buffer (0.15 M), the mixture was allowed to stand for five minutes at room temperature. After that, 250 µl $FeSO_4$ (2.5 mM) was added and allowed to stand for thirty minutes at room temperature. Following centrifugation at 2500 rpm for 5 min, the absorbance of the supernatant was measured at 540 nanometers.

% Inhibition of anti-haemolytic =

$$\% \text{ activity} = \frac{A-B}{A} \times 100$$

Where A = Control absorbance; B = Test sample absorbance

3. Results and Discussion

3.1. Phenolic and flavonoid contents of C. albidum fruit pulp extract.

Results showed that *C. albidum* pulp had higher phenolic content (59.64 ± 0.52 mg/g extract TAE) than flavonoids (35.31 ± 0.74 mg/g extract QE) (Table 1).

Table 1. Total phenolic and flavonoid contents of methanolic extract of *C. albidum* pulp residue.

Extract	Total Phenolics (mg/g extract TAE)	Total Flavonoids (mg/g extract QE)
***C. albidum* pulp**	59.64 ± 0.52	35.31 ± 0.74

3.2. Antioxidant activity of methanolic extract of C. albidum pulp residue.

The antioxidant profiles of *C. albidium* pulp extract are presented in Table 2. The percentage lipid peroxidation activity of *C. albidium* pulp (60.17 ± 1.24%) was significantly lower than ascorbic acid (86.87 ± 3.13%). Similarly, the DPPH radical scavenging activity of the extract (50.97 ± 1.18%) was significantly lower than the ascorbic acid standard (73.39 ± 1.49%). The H_2O_2 radical scavenging activity of the pulp extract (23.38 ± 2.59%) was significantly lower than the ascorbic acid (76.94 ± 0.41%). Meanwhile, *C. albidium* pulp had higher FRAP activity (IC_{50} 0.34 ± 0.01 mg/ml) than the ascorbic acid standard (IC_{50} 2.52 ± 0.05 mg/ml). The total antioxidant capacity (TAC) of *C. albidium* pulp was 272.94 ± 11.15 mg/g AAE.

Table 2. Antioxidant activity of crude extract of *C. albidium* pulp residue.

Samples	% Inhibition of Lipid Peroxidation	% inhibition of DPPH	FRAP IC_{50} (mg/ml)	H_2O_2 Radical (%)	TAC (mg/g AAE)
C. albidium	60.17 ± 1.24	50.97 ± 1.18	0.34 ± 0.01	23.38 ± 2.59	272.94 ± 11.15
Ascorbic acid	86.87 ± 3.13	73.39 ± 1.49	2.52 ± 0.05	76.94 ± 0.41	NA

NA: not applicable

3.3. Anti-inflammatory activity of C. albidium pulp extract.

Anti-inflammatory results of *C. albidium* pulp extract were presented in Figures 1 a-d. The *C. albidium* pulp extract had a lower percentage of anti-proteinase activity of 65.32% ± 4.57% compared with 83.70 ± 5.12% of Ibuprofen standard (Figure 1a) Conversely, both *C. albidium* extract (40.00 ± 0.87%) and Ibuprofen drug (40.00 ± 0.64%) maintained equal levels of percentage anti-denaturant activity (Figure 1b). Ibuprofen had the highest percentage of RBC membrane stabilizing activity of 84.39 ± 1.62%, while *C. albidium* extract had 67.55 ± 6.80% (Figure 1c). The effects of both drugs were found to be the biphasic mode of protection. Ibuprofen drug (16.50 ± 0.88%) and *C. albidium* (17.64 ± 1.02%) exhibited weak levels of anti-hemolytic activity (Figure 1d).

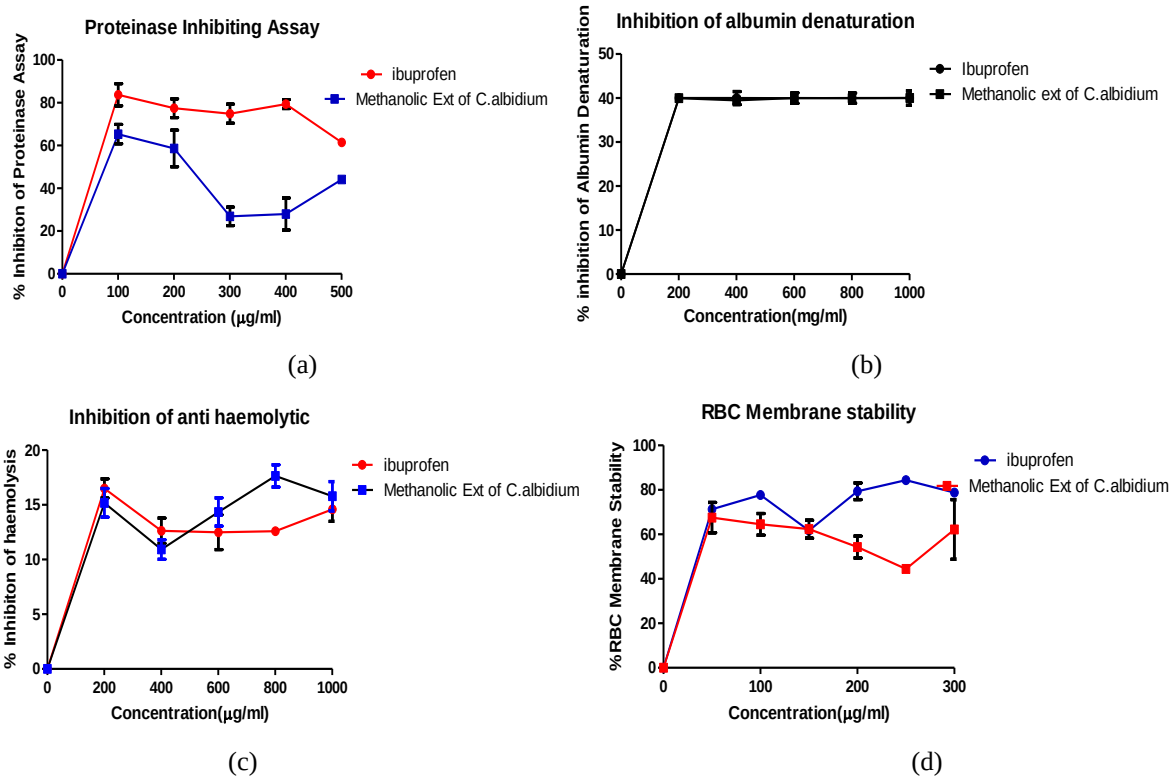

Figure 1a- d. Anti-inflammatory profile of *C. albidium* pulp residue.

When inflammation occurs, the lysosomal enzymes are released due to the damaged membrane and several typical alterations [26-30]. Protection or stabilization of membrane integrity prevents the release of lysosomal constituents, which may damage some tissue proteins, slowing down the inflammation process [31]. Many anti-inflammatory drugs have been shown to exhibit anti-inflammatory activity by stabilizing the lysosomal membrane [32,33]. This present study has shown that Ibuprofen elicited a higher percentage of membrane stabilizing activity than *C. albidium* pulp. Conversely, digestive enzymes such as proteinases have been shown to play important roles in inflammatory processes such as arthritis. Studies have shown that neutrophils harbor many serine proteinases in their lysosomal granules, which mediate tissue damage during inflammatory processes. Also, several proteinase inhibitors have been developed or isolated from plant-based products which guide against tissue protein degradation [34]. *C. albidium* pulp was found to possess anti-proteinase activity significantly lower than Ibuprofen. There was no significant difference between the anti-denaturant and anti-hemolytic activities of pulp and Ibuprofen. Several flavonoids and polyphenols have been shown to possess robust antioxidant and anti-inflammatory properties [34]. Therefore, it could

be surmised that the antioxidant and anti-inflammatory activities observed in this present study may have been due to the presence of these phytochemicals in the pulp extract of *C. albidium*.

4. Conclusions

In conclusion, the pulp extract of *C. albidum* fruits as an antioxidant and anti-inflammatory agent was investigated using *in vitro* approach. The pulp residue was observed to possess strong antioxidant and anti-inflammatory properties, which could be linked to the presence of flavonoids and phenols. Thus, feeding on the fruit pulp residue may improve the health profits of *C. albidium*. Also, the findings from this study may open the door for further studies in academics and industry.

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Conflicts of Interest

The authors declare no conflict of interest.

References

1. Xu, K.; Cui, X.; Wang, B.; Tang, Q.; Cai J.; Shen, X. Healthy adult vegetarians have better renal function than matched omnivores: a cross-sectional study in China. *BMC Nephrology* **2020**, *21*, https://doi.org/10.1186/s12882-020-01918-2.
2. Wassie, T.H.; Ewnetu, M.; Akililu A.; Demissie, D.B. Proportion of Caesarean Section and its Associated Factors among Women who Gave Birth in Debre Tabor General Hospital, North West Ethiopia. *EC Gynaecology* **2020**, *9*, 1-10.
3. Dzarma, G.W.; Agu, C.M.; Nwosu-Obieogu, K.; Onyedikachi, L.N.; Kalu, O.A.; Ekezie, E.O.; Adiele M.C.; Chukwulebile, A.S. Parametric study of oil extraction from African Star Apple (Chrysophyllum albidum) seeds. *Cleaner Chemical Engineering* **2022**, *2*, https://doi.org/10.1016/j.clce.2022.100018.
4. Akpe, A.R.; Okwu, G.I.; Abu, D.; Femi, I.J. Mycoflora and nutritional composition of Chrysophyllum albidum fruits/ seed oil. *International Journal of Sciences & Applied Research*. **2022**, *7*, 23-37.
5. Dimkpa, S.O.N.; Chukunda, F.A.; Onyige, H.U. Effects of Culture Filtrates of Pathogenic Fungi on Seed Germination and Seedling Indices of Chrysophyllum albidum (G.Don). *Journal of Biology, Agriculture and Healthcare* **2022**, *12*, 22-29.
6. Oluyori, A.P.; Dada, A.O.; Ogunnupebi, T.A.; Owolabi, A.O.; Adelani-Akande, T.A.; Inyinbor, A.A. Antimicrobial activity of *Chrysophyllum albidum* seed extract and its effect on the physicochemical properties of cherry tomato fruits during postharvest storage. *Journal of food Processing and preservation* **2022**, *46*, http://dx.doi.org/10.1111/jfpp.17062.
7. Ureigho, U.N.; Ohwo, A.K.; Odjegba, E.O.; Okunomo K. The effect of shifting cultivation practice on sakponba forest reserve, Edo State, Nigeria. *International Journal of Biosciences* **2021**, *13*, 1-9.
8. Usman, K.; Abba H.; Iyun, O.R.A. Preparation and Characterization of African Star Apple Seed Shell (Chrysophyllum Africanum) For The Removal of Acid Red 9. *Communication in Physical Sciences* **2022**, *8*, 23-32.
9. Adekanmi D.G.; Olowofoyeku, A.E. African Star Apple: Potentials and Application of Some Indigenous Species in Nigeria. *J. Appl. Sci. Environ. Manage.* **2020**, *24*, 1307-1314, https://doi.org/10.4314/jasem.v24i8.1.
10. Osebhahiemen, I.; Obuseh, A.F.; Omoregie· S.E. Comparative Study on Effect of *Chrysophyllum albidum* and *Irvingia gabonensis* Leaves Extracts on High-Fat Diet-Induced Obesity in Wistar Rats. *Food and Nutrition Sciences*, **2022**, *13*, 259-283, https://doi.org/10.4236/fns.2022.133022.
11. Ogunleye, F.A.; Fapohunda, O.; and Nwangwu, S. A Review on Medicinal Uses and Pharmacological Activities of African Star Apple (Chrysophyllum albidum). *Acta Scientific Cancer Biology* **2020**, *1*.

12. Platzer, M.; Kiese, S.; Herfellner, T.; Schweiggert-Weisz, U.; Eisner, P. How Does the Phenol Structure Influence the Results of the Folin-Ciocalteu Assay? *Antioxidants* **2021**, *10*, https://doi.org/10.3390/antiox10050811.
13. Farhadi, R.; Afkari-Sayyah, A.H.; Jamshidi, B.; Gorji A.M. Prediction of internal compositions change in potato during storage using visible/near-infrared (Vis/NIR) spectroscopy. *Int. J. Food Eng.* **2020**, https://doi.org/10.1515/ijfe-2019-0110.
14. Munteanu, I.G.; Apetrei, C. Assessment of the Antioxidant Activity of Catechin in Nutraceuticals: Comparison between a Newly Developed Electrochemical Method and Spectrophotometric Methods. *Int. J. Mol. Sci.* **2022**, *23*, https://doi.org/10.3390/ijms23158110.
15. Abdou, R.M.; El-Maadawy, W.H.; Hassan, M.; El-Dine, R.S.; Aboushousha, T.; El-Tanbouly, N.D.; El-Sayed, A.M. Nephroprotective activity of Aframomum melegueta seeds extract against diclofenac-induced acute kidney injury: A mechanistic study. *Journal of Ethnopharmacology* **2021**, *273*, https://doi.org/10.1016/j.jep.2021.113939.
16. Manzocco, L.; Romano, G.; Calligaris, S.; Nicoli, M.C. Modeling the Effect of the Oxidation Status of the Ingredient Oil on Stability and Shelf Life of Low Moisture Bakery Products: The Case Study of Crackers. *Foods* **2020**, *9*, https://doi.org/10.3390/foods9060749.
17. Mohan Reddy, Y.; Jeevan Kumar, S. P.; Saritha, K. V.; Gopal, P.; Madhusudana Reddy, T.; Simal-Gandara, J. Phytochemical Profiling of Methanolic Fruit Extract of *Gardenia latifolia* Ait. by LC-MS/MS Analysis and Evaluation of Its Antioxidant and Antimicrobial Activity. *Plants (Basel)* **2021**, *10*, https://doi.org/10.3390/plants10030545.
18. Adegbola, M.V.; Anyim, G.; Ntwasa, M.; Ayeleso, A.O.; Oyedepo, T.A. Potential Effect of Syzygium aromaticum (Cloves) Extract on Serum Antioxidant Status and Lipid Profiles in Wistar Rats with Artesunate Toxicity. *Appl. Sci.* **2022**, *12*, https://doi.org/10.3390/app12168216.
19. Bonsou, I.N.; Mbaveng, A.T.; Nguenang, G.S.; Chi, G.F.; Kuete, V.; Efferth, T. Cytotoxicity, acute and sub-chronic toxicities of the fruit extract of Tetrapleura tetraptera (Schumm. & Thonn.) Taub. (Fabaceae). *BMC complementary medicine and therapies* **2022**, *22*, https://doi.org/10.1186/s12906-022-03659-1.
20. Islam, M.D.; Islam, A.; Tasnin, N.; Akter S.F.; Uddin, M.S. Exploration of Antioxidant and Anticancer Activity of Stephania japonica Leaves Extract. *Journal of Pharmaceutical Research International* **2021**, *33*, 256-272, https://doi.org/10.9734/jpri/2021/v33i28A31529.
21. Adetoun, M.; Akinpelu, B.; Bolaji, A.; Adelowo, J.; Babarinde, S.; Oyedapo, O. Wound Healing Activities of Latex of Grassleaf Spurge (Euphobia graminea Jacq.). *American Journal of Biomedical Research* **2020**, *8*, 63-71, https://doi.org/10.12691/ajbr-8-3-2.
22. Chen, H.; Zhang, X.; Liu, L.; Cai, M.; Guo Z.; Qiu, L. Application of red clover isoflavone extract as an adjuvant in mice. *Experimental And Therapeutic Medicine* **2020**, *19*, 1175-1182, https://doi.org/10.3892%2Fetm.2019.8315.
23. Ogori, A.F, Abraham, G.T.; Eke, M.O, Abu, J.O.; Adeniyi, A.S.; Famuwagun A.A. The effect of balanities aeqyptiaca defatted protein meal and protein concentrate supplemented diet on biochemical and molecular stability of diabetic wister albino rat. *Biomedicine & Pharmacotherapy* **2022**, *153*, https://doi.org/10.1016/j.biopha.2022.113510.
24. Azeez, S.O.; Olowu, A.N.; Godwin, A.; Apata, J.T.; Akinpelu, B.A. Evaluation of Cytotoxic Effects of Methanolic Extract and Fractions of Mirabilis jalapa (L.) Leaf. *Journal of Applied Life Sciences International* **2021**, *24*, 25-35, http://dx.doi.org/10.9734/JALSI/2021/v24i630242.
25. Ajayi, O.B.; Oyetayo, F.L.; Akomolafe, S.F. Starch composition, glycemic indices, antioxidant properties and carbohydrate hydrolyzing enzymes activities of African star apple fruit parts. *BMC Complement Med Ther* **2020**, *20*, https://doi.org/10.1186/s12906-020-03053-9.
26. Humbare, R.B.; Sarkar, J.; Kulkarni, A.A.; Juwale, M.G.; Deshmukh, S.H.; Amalnerkar, D.; Chaskar, M.; Albertini, M.C.; Rocchi, M.B.L.; Kamble, S.C.; Ramakrishna, S. Phytochemical Characterization, Antioxidant and Anti-Proliferative Properties of Rubia cordifolia L. Extracts Prepared with Improved Extraction Conditions. *Antioxidants* **2022**, *11*, https://doi.org/10.3390/antiox11051006.
27. Tao, H.; Li, L.; He, Y.; Zhang, X.; Zhao, Y.; Wang, Q.; Hong, G. Flavonoids in vegetables: improvement of dietary flavonoids by metabolic engineering to promote health. *Critical Reviews in Food Science and Nutrition* **2022**, 1-15, https://doi.org/10.1080/10408398.2022.2131726.
28. Qiao, X.; Kashiouris, M.G.; L'Heureux, M.; Fisher, B.J.; Leichtle, S.W.; Truwit, J.D.; Nanchal, R.; Hite, R.D.; Morris, P.E.; Martin, G.S.; Sevransky, J.; Fowler, A.A. Biological Effects of Intravenous Vitamin C on Neutrophil Extracellular Traps and the Endothelial Glycocalyx in Patients with Sepsis-Induced ARDS. *Nutrients* **2022**, *14*, https://doi.org/10.3390/nu14204415.
29. Vítek, L.; Jirásková, A.; Malíková, I.; Dostálová, G.; Eremiášová, L.; Danzig, V.; Linhart, A.; Haluzík, M. Serum Bilirubin and Markers of Oxidative Stress and Inflammation in a Healthy Population and in Patients with Various Forms of Atherosclerosis. *Antioxidants* **2022**, *11*, https://doi.org/10.3390/antiox11112118.
30. Jiao, X.; Balaei, M.R.; Abu-El-Rub, E.; Casoni, F.; Modarres, H.P.; Dhingra, S.; Kong, J.; Consalez, G.; Marzban, H. Reduced Granule Cell Proliferation and Molecular Dysregulation in the Cerebellum of Lysosomal Acid Phosphatase 2 (ACP2) Mutant Mice. *Int. J. Mol. Sci.* **2021**, *22*, https://doi.org/10.3390/ijms22062994.

31. Wedamulla, N.E.; Fan, M.; Choi, Y-J.; Kim, E-K. Citrus peel as a renewable bioresource: Transforming waste to food additives. *Journal of Functional Foods* **2022**, *95*, https://doi.org/10.1016/j.jff.2022.105163.
32. Salve, P.; Vinchurkar, A.; Raut, R.; Chondekar, R.; Lakkakula, J.; Roy, A.; Hossain, M.J.; Alghamdi, S.; Almehmadi, M.; Abdulaziz, O.; Allahyani, M.; Dablool, A.S.; Sarker, M.M.; Nur Azlina, M.F. An Evaluation of Antimicrobial, Anticancer, Anti-Inflammatory and Antioxidant Activities of Silver Nanoparticles Synthesized from Leaf Extract of Madhuca longifolia Utilizing Quantitative and Qualitative Methods. *Molecules* **2022**, *27*, https://doi.org/10.3390/molecules27196404.
33. Ingawale, D.K.; Mandlik, S.K. New insights into the novel anti-inflammatory mode of action of glucocorticoids. *Immunopharmacology and Immunotoxicology* **2022**, *42*, 59-73, https://doi.org/10.1080/08923973.2020.1728765.
34. Mba, J.R.; Zouheira, D.; Dairou, H.; Yadang, F.S.A.; Gael, N.N.; Ayong, L.; Kuiate, J-R.; Agbor, G.A. *In Vitro* Antioxidant, Anti-Inflammatory, and Digestive Enzymes Inhibition Activities of Hydro-Ethanolic Leaf and Bark Extracts of *Psychotria densinervia* (K. Krause) Verdc. *Adv Pharmacol Pharm Sci*. **2022**, https://doi.org/10.1155/2022/8459943.