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Short Communication

Glucogallin and Conjugated Linoleic Acids Isolated from *Ricinodendron heudelotii* (Bail.) Seeds

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ABSTRACT

In a bid to explore and enhance the use of natural products for medicinal purposes, this study analyzed the crude extract of *Ricinodendron heudelotii* seeds and isolated four biologically active compounds from ethyl acetate fraction of *R. heudelotii* seeds extract. The study utilized GC-MS in the analysis of the crude extract. Various spectroscopic and spectrometric methods in addition to reported data were employed in characterizing isolates. Twenty-two compounds were detected in the crude extract via GC-MS. Palmitic acid had the highest abundance of 23.9%, followed by α-linolenic acid (14.72%). The ethyl acetate fraction by means of chromatographic techniques yielded four compounds namely glucogallin **(1),** (6Z,9Z,11Z)-6,9,11-octadecatrienoic acid **(2)**, (6Z,9E,11E)-6,9,11-octadecatrienoic acid **(3)** and (6Z,9Z,13Z)-6,9,13-octadecatrienoic acid **(4)**. All four isolated compounds have biological and pharmaceutical properties including anti-inflammatory and anti-diabetic. This study, therefore, proposes linolenic acid and gallic acid derivatives as chemophenetic markers of *R. heudelotii* and suggests the medicinal use of *R. heudelotii* seeds for anti-inflammatory and anti-diabetic disorders.

***Keywords*:** *Euphorbiceae, Ricinodendron heudelotii*, Linolenic acid, Glucogallin, Gallic acid, Anti-inflammation activity, Anti-diabetic activity.

Introduction

Inflammation is a complex biological response that occurs in the body tissues as a protective reaction to pathogens, damaged cells, irritants, and stimuli.[1-2] It involves the participation of immune cells, blood vessels, and molecular mediators.[3] While inflammation normally resolves on its own in certain biochemical disorders, it can become persistent, leading to chronic inflammatory diseases.[1]

Diabetes mellitus is a group of metabolic disorders characterized by prolonged elevation of blood glucose levels, accompanied by symptoms such as increased urination, excessive thirst, and excessive hunger.[4] Long-term high blood sugar levels have been linked to dysfunction in organs such as the retina, kidneys, nerves, heart, and blood vessels.[5] Research in both humans and animal models has shown that inflammation plays a role in the development of type 2 diabetes, leading to insulin insensitivity and subsequent inflammation.[6-9] Chronic activation of pro-inflammatory pathways in insulin target cells has been suggested to contribute to obesity, insulin resistance, and related metabolic disorders, including type 2 diabetes.[9]

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The clinical relationship between inflammation, atherosclerotic cardiovascular diseases, and diabetes has been established, highlighting the need for research into therapeutic approaches that target these diseases simultaneously.[10]

The utilization of plants in complementary and alternative medicine, as well as in drug discovery, has received significant attention due to the presence of bioactive constituents that contribute to the therapeutic properties of plants.[11]

Ricinodendron heudelotii (Bail.) Pierre ex Heckel, commonly known as "njangsa," is the only known species of the *Ricinodendron* genus in the Euphorbiaceae family. It serves as a source of food, medicine, and various commodities for local populations in West Africa. The leaves of this plant have been traditionally used to treat conditions such as dysentery, female sterility, edemas, and stomach pains. The sap extracted from the plant is applied to the eyes in the treatment of filarial and ophthalmic conditions. The root bark is powdered and mixed with pepper and salt to treat constipation.[11]

Ricinodendron heudelotii has been of great benefits as food and medicinally. Although some biologically active compounds have been isolated from other parts of the plant such as the leaves and roots of the plant *Ricinodendron heudelotii*, there has not been adequately published compounds isolated from the seed extracts of plant, hence the need to explore the seeds for better utilization. This study examined the crude extract of *Ricinodendron heudelotii* seeds, isolated and characterized four biologically active compounds from the extract, and assessed their anti-inflammatory and anti-diabetic properties.

Materials and Methods

Collection and Identification of Ricinodendron heudelotii seeds
Samples of *Ricinodendron heudelotii* seeds were obtained in February, 2022 from a Mile one market in Rivers State, Nigeria. The plant sample was identified and validated by Dr. M. G. Ajuru of the Department of Plant Science and Biotechnology, Rivers State University, Nigeria and assigned a voucher number, FHI 110573.

Extraction of plant material
Ricinodendron heudelotii dried seeds (1620g) were selected to remove impurities; after which they were ground to obtain 1500g of the powdered sample. The Powdered sample (1500g) was extracted by Maceration with 80% ethanol (2500 ml) for a period of 72 hours accompanying occasional shaking and stirring. The extract was filtered with Whatman No. 1 filter paper.
The filtrate was concentrated to dryness using a rotary evaporator (BUCH rotavapor R-210) to allow evaporation of the solvent under pressure of 175mbar at 50^{o}C. The crude extract obtained 48.67 g (3.24% yield) was subjected to liquid-liquid partition using n-hexane, ethyl acetate and butanol. The extract was then stored at 4^{o}C in a refrigerator until further use.

Fractionation
The crude extract (3.0 g) was also subjected for GC-MS analysis to evaluate the chemical composition of the plant. The crude *n*-hexane fraction was a stable oil, while the ethyl acetate fraction (11.7 g) which was the only viscous fraction was subjected to repeated open column chromatography over silica gel (80 g), and eluted with a binary system of *n*-hexane/ethyl acetate (100:0-0:100, v: v) gradient. Fractions of 10 mL were collected, evaporate and monitored using TLC. Fractions were grouped and indexed as EA1-50. Fraction EA8-10 were grouped and subjected to an open column chromatography with normal phase silica gel as stationary phase and eluted with a system of *n*-hexane/ethyl acetate (100:0-0:100, v:v) gradient to afford compound **2** (4.8 g) and the mixture of compounds **3** and **4** (2.4 mg). Fraction EA33-35 were grouped (4.6 g) and subjected to HPLC to afford compound **1** (1.9 mg).

Experiment procedures
Triple Quadrupole Acquisition method was used for the characterization of *Ricinodendron heudelotii* crude extract using Agilent technologies 7000 GC-MS triple quad (MS-7000, GC-7890A). The ZEBRONZB-5HT Column (30 m× 320 µm × 0.25 µm) at 400 ℃, In: Front SS inlet He and Out: vacuum was used. The oven was equilibrated for 5 minutes, at 60 ℃, run time was 56.5 minutes (8 ℃/min to 240 ℃ for 20 minutes and 15 ℃/minute to 300 ℃ for 5 minutes). Volume of injected sample was 1.5 µl. Computer matching of mass spectra was performed using the NIST Mass Spectrometry Data Center and WILEY7.0 library and the retention times of known species injected into the chromatographic column were used for identification of peaks.
Fractionation and purification were achieved using open column chromatography (70 cm × 4 cm) with normal phase silica gel (Merck 70–230 Mesh), normal and reverse-phase HPLC and sephadex LH-20. Purity of compounds was confirmed on pre-coated normal phase TLC, while detection of isolates was done using UV light (254 and 364 nm), ceric sulfate and iodine (as spray).
NMR analyses were performed on Bruker AMS-400 and AMX-500 while masses were recorded on low resolution EIMS type JEOL MS Route JMS 600H mass spectrometer.

Results and Discussion

Result obtained from GC-MS analysis (Table 1) revealed the presence of twenty-two compounds detected from ethanol crude extract of *Ricinodendron heudelotii* seeds. Palmitic acid had the highest abundance of 23.9%, followed by α-linolenic acid (14.72%). Other compounds are contained in Table 1.

Structural elucidation and chemophenetic significance of the isolated compounds
From the ethyl acetate fraction of *Ricinodendron heudeloti*, seed extract, four secondary metabolites were isolated by the means of repeated chromatographic techniques and characterized using spectroscopic and spectrometric methods. All the isolated compounds were deduced as known compounds after the interpretation and the comparison of their NMR and MS data to previous reported literature; then identified as glucogallin (**1**),[12] (6Z,9Z,11Z)-6,9,11-octadecatrienoic acid (**2**), (6Z,9E,11E)-6,9,11-octadecatrienoic acid (**3**) and (6Z, 9Z, 13Z)-6,9,13-octadecatrienoic acid (**4**). [13-15] The presence of these compounds in *Ricinodendron heudelotii* and as constituent of *Ricinodendron heudelotii* seed oil has been reported[16] using the GC-MS technique of analysis. Also, GC-MS result of the crude extract (Table 1) shows the presence of octadecatrienoic acid (α-linolenic acid) and its isomers.

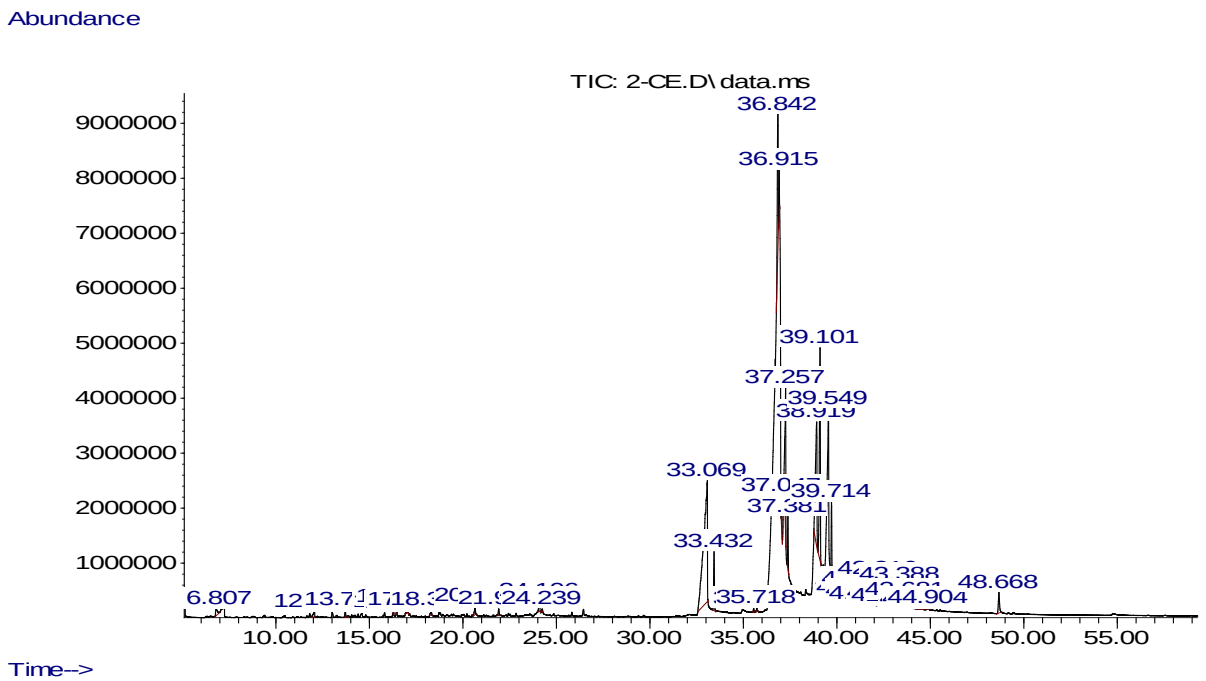

Figure 1: Chromatogram of Crude extract of *Ricinodendron heudelotii* seeds

Table 1: Chemical composition of Crude extract of *Ricinodendron heudelotii* seeds

Number	Compound	Retention time (Minute)	Concentration (%)
1	1-[(Trimethylsilyl)oxypropan-2-ol	6.82	0.65
2	1,2-Diazaspiro(2.5)octane	13.71	0.30
3	4,4,6-Trimethyl-cyclohexa-2-en-1-ol	16.46	0.40
4	Bicyclo(3.3.1)nonane-2,6-dione	16.93	0.09
5	2-*tert*-Butyl-5-propyl-[1,3]dioxolan-4-one	18.30	0.40
6	3,6-Dimethyl-octa-2-one	20.64	0.20
7	Lauric acid	21.91	0.30
8	Monomethyl azelate	24.11	0.60
9	Methyl octadec-9-ynoate	24.23	0.30
10	Palmitic acid	33.06	23.90
11	Ethyl palmitate	33.43	2.12
12	Linoleic acid	36.84	6.21
13	n-Propyl linoleate	37.05	10.82
14	Stearic acid	37.25	10.25
15	Methyl-15-ethylheptadecanoate	37.38	1.62
16	α-Linolenic acid	38.91	14.72
17	Ethyl linolenate	39.09	10.79
18	Methyl-8,11,14-heptadecatrienoate	39.55	12.62
19	1,3-Dipalmitin trimethylsilyl ether	41.37	0.51
20	Diisooctyl phthalate	42.22	0.85
21	γ-Tocopherol	48.67	1.20
22	Ethyl linoleate	35.55	1.15

Classes of secondary metabolites are often specific and restricted to taxonomically related organisms. Classification of plants based on specific class of secondary metabolite and their biosynthetic pathway is on the bases of chemophenetic study.[17-19] It has been noticed that the species of plants found in the same genus as well as family usually synthesized similar classes or derivatives of almost the same compounds due to the presence of similar enzymes.[20-21]

Spectroscopic data of the Isolated compounds 1-4.

Glucogallin (1): Amorphous white powder; Molecular formula: $C_{13}H_{16}O_{10}$; 1H NMR (400 MHz, CD_3OD) δ 6.78 (d, J = 2.4 Hz, 2H), 4.58 (d, J = 5.2 Hz, 1H), 4.20 – 4.11 (m, 1H), 4.10 (dd, J = 13.5-2.4 Hz, 1H), 4.03 (s, 1H), 3.97 (s, 1H), 3.81 – 3.74 (m, 1H), 3.72 – 3.61 (m, 1H); LR-EI-MS, M^+ *m/z* 332.1 (calcd. for $C_{13}H_{16}O_{10}$, *m/z* 332.1).

(6Z,9Z,11Z)-6,9,11-octadecatrienoic acid (2): Amorphous white powder; Molecular formula: $C_{18}H_{30}O_2$; ^{1}H NMR (500 MHz, CD_3OD) δ 5.33 (td, J = 10.9, 4.7 Hz, 6H), 2.77 (t, J = 6.5 Hz, 2H), 2.27 (t, J = 7.4 Hz, 2H), 2.05 (td, J = 8.2, 4.0 Hz, 2H), 1.64 – 1.54 (m, 6H), 1.42 – 1.26 (m, 9H), 0.90 (t, J = 6.9 Hz, 3H); ^{13}C NMR (125 MHz, CD_3OD) δ 177.69, 135.00, 132.37, 130.09, 129.12, 129.06, 127.11, 34.99, 32.68, 30.31, 28.67, 28.18, 28.14, 28.12, 26.56, 23.74, 23.29, 14.28; LR-EI-MS, M^+ *m/z* 278.1 (calcd. for $C_{18}H_{30}O_2$, *m/z* 278.2).

(6Z,9E,11E)-6,9,11-octadecatrienoic acid (3): Amorphous white powder; Molecular formula: $C_{18}H_{30}O_2$; ^{1}H NMR (500 MHz, CD_3OD) δ 5.40 – 5.27 (m, 6H), 2.77 (t, J = 6.5 Hz, 2H), 2.26 (t, J = 7.4 Hz, 2H), 2.09 – 2.00 (m, 2H), 1.64 – 1.54 (m, 2H), 1.41 – 1.30 (m, 4H), 1.29 (brs, 9H), 0.89 (t, J = 7.1 Hz, 3H); ^{13}C NMR (125 MHz, CD_3OD) δ 177.73, 135.79, 135.00, 132.37, 130.94, 129.06, 127.11, 34.99, 33.53, 32.68, 30.85, 30.71, 30.13, 29.14, 28.67, 26.56, 23.74, 14.44; LR-EI-MS, M^+ *m/z* 278.1 (calcd. for $C_{18}H_{30}O_2$, *m/z* 278.2).

(6Z,9Z,13Z)-6,9,13-octadecatrienoic acid (4): Amorphous white powder; Molecular formula: $C_{18}H_{30}O_2$; ^{1}H NMR (500 MHz, CD_3OD) δ 5.40 – 5.27 (m, 6H), 2.77 (t, J = 6.5 Hz, 2H), 2.26 (t, J = 7.4 Hz, 2H), 2.09 – 2.00 (m, 2H), 1.64 – 1.54 (m, 2H), 1.41 – 1.30 (m, 4H), 1.29 (brs, 9H), 0.89 (t, J = 7.1 Hz, 3H); ^{13}C NMR (125 MHz, CD_3OD) δ 177.73, 134.97, 132.37, 130.94, 129.12, 127.11, 33.79, 32.68, 30.13, 28.67, 28.18, 28.14, 28.12, 26.56, 26.09, 23.74, 23.29, 14.28; LR-EI-MS, M^+ *m/z* 278.1 (calcd. for $C_{18}H_{30}O_2$, *m/z* 278.2).

Biological activities of isolated compounds

Compound 1: Glucogallin

β-Glucogallin, a compound found in plants, was obtained from the ethyl acetate portion of the extract obtained from *Ricinodendron heudelotii* seeds. It is also commonly known as 1-O-galloyl-*β*-D-glucopyranose. This compound is a polyphenolic ester derived from plants and plays a crucial role in the production of hydrolysable tannins. Some of its reported biological activities include its ability to scavenge free radicals, which makes it potentially beneficial in the treatment of diseases such as diabetes and its associated complications like glaucoma, retinopathy, inflammation, hepatic damage, and skin damage caused by UV light.[22] It has also been reported to possess nutraceutical properties. Various derivatives of *β*-glucogallin, including 1,4,6-tri-O-galloylglucose, 3,4,6-tri-O-galloylglucose, 1,2,6-tri-O-galloylglucose, and 4,6-di-O-galloylglucose, have been identified in the leaves of *Ricinodendron heudelotii*. Pupalla *et al.*[23] conducted a study that demonstrated the therapeutic potential of glucogallin in the treatment of diabetic complications such as cataracts. The antiglycation activity of *β*-glucogallin derived from *Asparagus racemosus* has been suggested, indicating its potential pharmacological benefits in managing metabolic disorders associated with advanced glycation end products. Considering the reported therapeutic properties of *β*-glucogallin, its presence in *Ricinodendron heudelotii* seeds may contribute to the plant's anti-inflammatory abilities.

Compound 2: (6Z,9Z,13Z)-6, 9, 13-octadecatrienoic acid.

γ-Linolenic acid, also known as GLA, was identified by Odinga *et al*[16] in the essential oil extracted from *Ricinodendron heudelotii* seeds. GLA is an intermediate compound in the production of arachidonic acid and has been detected in certain seed oils.[25] When γ-linolenic acid is enzymatically converted into arachidonic acid, it can generate pro-inflammatory substances like series 2 prostaglandins and series 4 leukotrienes.[26] Moreover, GLA exhibits anti-inflammatory properties by acting as a precursor to eicosanoids, which are known for their anti-inflammatory effects. It can also suppress inflammation mediators such as interleukin 1β (IL-1β), interleukin 6 (IL-6), and cytokine-tumor necrosis factor α (TNF-α).[27] As an omega-6 fatty acid, γ-linolenic acid plays a crucial role in the proper functioning of various tissues in the human body. It serves as a precursor for anti-inflammatory eicosanoids, including series 1 prostaglandins and 15-hydroxyeicosatrienoic acid.[26] Notably, extracts derived from *Ricinodendron heudelotii* seeds have been found to inhibit reactive oxygen species (ROS).[28] This activity can be attributed to the presence of γ-linolenic acid in the seed extract and oil, which possesses anti-inflammatory properties. Patients with atopic dermatitis have been observed to have deficiencies in γ-linolenic acid and other linolenic acid isomers in their plasma.[29] Based on the aforementioned findings, it is plausible to propose that γ-linolenic acid, isolated from *Ricinodendron heudelotii* extract, may serve as the active anti-inflammatory component of the seeds, as reported.[28]

Compound 3 and 4: Conjugated Linoleic Acids

Conjugated linolenic acid (CLA) refers to various positional and geometric isomers of octadecatrienoic acid.[30] CLA includes compound 3 [(6Z,9E,11E)-6,9,11-octadecatrienoic acid] and compound 4 [(6Z,9Z,13Z)-6,9,13-octadecatrienoic acid].[31] CLA is a mixture of these isomers found in seeds, and there is evidence suggesting its numerous health benefits, such as its anti-carcinogenic, lipid metabolism regulatory, anti-inflammatory, anti-diabetic, anti-obesity, and antioxidant activities. These benefits are primarily based on in vitro studies of CLA isomers derived from natural sources, particularly edible seeds.[32-33] A study reported that jacaric acid and four of its geometric isomers of octadecatrienoic acid selectively induced apoptosis in both hormone-dependent and independent human prostate cancer cells, while not affecting the viability of normal human prostate epithelial cells.[34] Compounds 2, 3, and 4 have been identified as γ-linolenic acid and isomers of octadecatrienoic acid, respectively. Gómez-Cortés *et al*[35] investigated the heat-induced cis-trans isomerization of ethylenic bonds in octadecatrienoic acids and found that the predominant isomer formed is trans-5, cis-9, trans-12 18:3 octadecatrienoic acid. Additionally, the heat-induced geometrical isomerization of pinolenic acid differs from that of α- and γ-linolenic acids in at least two aspects. The aforementioned literature further supports the anti-inflammatory and anti-diabetic activities of *Ricinodendron heudelotii* seeds, as reported.[28]

Conclusion

Four antidiabetic and anti-inflammatory compounds have been successfully isolated from *Ricinodendron heudelotii* seeds and characterized as glucogallin (a derivative of gallic acid) and conjugated linolenic acids (γ-linolenic acid, (6Z,9E,11E)-6,9,11-octadecatrienoic acid and (6Z,9Z,13Z)-6,9,13-octadecatrienoic acid) using various chromatographic, spectrometric and spectroscopic methods. The structures of the isolates were found to be in agreement with previous studies. This study also proposed linolenic acid and gallic acid derivatives as chemophenetic markers of *Ricinodendron heudelotii*.

Conflict of Interest

The authors declare no conflict of interest.

Authors' Declaration

The authors hereby declare that the work presented in this article is original and that any liability for claims relating to the content of this article will be borne by them.

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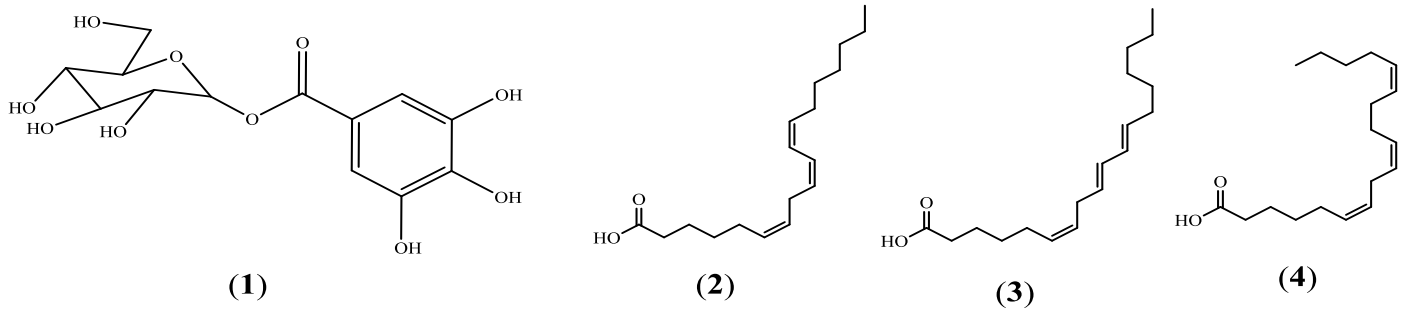

Figure 2: Structures of Isolated compounds 1-4

References

1. Ferrero-Miliani L, Nielsen OH, Andersen PS, Girardin SE. Chronic inflammation: importance of NOD2 and NALP3 in interleukin-1beta generation. Clin Exp Immunol. 2007;147(2):227-235.
2. Chen L, Deng H, Cui H, Fang J, Zuo Z, Deng J, Li Y, Wang X, Zhao L. Inflammatory responses and inflammation-associated diseases in organs. Oncotarget, 2017; *9*(6), 7204–7218.
3. Signore A. About inflammation and infection. EJNMMI Res. 2013;3(1):8. doi: 10.1186/2191-219X-3-8. PMID: 23374699; PMCID: PMC3564704.
4. American Diabetes Association. Diagnosis and classification of diabetes mellitus [published correction appears in Diabetes Care. 2010;33(4): e57]. Diabetes Care. 2010;33 Suppl 1(Suppl 1): S62-S69.

5. Inzucchi SE. Diagnosis of diabetes. N Engl J Med. 2013; 368:193.
6. Alzamil H. "Elevated Serum TNF-α Is Related to Obesity in Type 2 Diabetes Mellitus and Is Associated with Glycemic Control and Insulin Resistance", J. Obes. , 2020, Article ID 5076858, 1-5.
7. Rehman K. Akash MSH. Mechanisms of inflammatory responses and development of insulin resistance: how are they interlinked? J Biomed Sci. 2016; **23**, 87.
8. Odinga T, Nwaokezi CO. Effect of *Ricinodendron heudelotii* seed extract on the oxidative stress biomarkers of Diabetic albino rats. J. Pharm. Res, 2020; 11-1905.
9. Marques-Vidal P, Schmid R, Bochud M, Bastardot F, Von Känel R, Paccaud F, Glaus J, Preisig M, Waeber G, Vollenweider P. Adipocytokines, hepatic and inflammatory biomarkers and incidence of type 2 diabetes. the CoLaus study. PLoS One. 2012;7(12): e51768.
10. Tsalamandris S, Antonopoulos AS, Oikonomou E, Papamikroulis GA, Vogiatzi G, Papaioannou S, Deftereos S, Tousoulis D. The Role of Inflammation in Diabetes: Current Concepts and Future Perspectives. Eur. cardiol., 2019; 14(1), 50–59.
11. Odinga T, Worlu-Wodu QE, Deekae S. Bioprospective screening of *Ricinodendron heudelotii* seeds. J. Anal. Pharm. Res, 2016; 3(10); 00084-92
12. Yakubu OF, Adebayo AH, Dokunmu TM, Zhang YJ, Iweala E. Cytotoxic Effects of Compounds Isolated from *Ricinodendron heudelotii*. Molecules, 2019; 24(1), 145.
13. Salsinha AS, Pimentel LL, Fontes AL, Gomes AM, Rodríguez-Alcalá LM. Microbial Production of Conjugated Linoleic Acid and Conjugated Linolenic Acid Relies on a Multienzymatic System. Microbiol Mol Biol Rev. 2018;82(4): e00019-18.
14. Ngo Njembe MT, Dormal E, Gardin C, Mignolet E, Debier C, Larondelle Y. Effect of the dietary combination of flaxseed and *Ricinodendron heudelotii* or *Punica granatum* seed oil on the fatty acid profile of eggs. Food Chem. 2021; 344:128668.
15. Ezekwe MO, Samuel AB, Ramon J. Nutritive composition of omega-3 fatty acids-rich *Ricinodendron heudelotii* and its potential for nutrition. Inter. J. nutr. Met., 2014; 6(6), 56-62.
16. Odinga T, Yousuf S, Choudhary MI, Ndukwe GI, Obinna PC, Otobo MB, Nwokogba CC. (2023). Bioactive Components, Anti-Dengue and Insecticidal Potencies of Ricinodendron heudelotii (Baill.) Seed Oil. Int. J. Med. Plant. Nat. Prod. 2023; 9(1); 6-13.
17. Zidorn C. Plant chemophenetics - A new term for plant chemosystematics/plant chemotaxonomy in the macro-molecular era. Phytochemistry. 2019; 163:147-148. doi: 10.1016/j.phytochem. 2019.02.013. Epub, PMID: 30846237.
18. Teoh ES. Secondary Metabolites of Plants. Medicinal Orchids of Asia. 2015; 5:59–73.
19. Borenstein E, Kupiec M, Feldman MW, Ruppin E. Large-scale reconstruction and phylogenetic analysis of metabolic environments. Proc Natl Acad Sci U S A. 2008;105(38):14482-14487.
20. Akesa TM. Phytotaxonomy and phytochemicals of Eight species of the Family Moraceae in Benue State, Nigeria. Int. J. Sci. Engr. Res., 2016; 7, 588.
21. Ntie-Kang F, Lifongo L. L, Mbaze LM, Ekwelle N, Owono Owono LC., Megnassan E, Judson PN, Sippl W, Efange SM. Cameroonian medicinal plants: a bioactivity versus ethnobotanical survey and chemotaxonomic classification. BMC Complement Altern Med. 2013; 13:147.
22. Khan AN, Singh R, Bhattacharya A, Chakravarti R, Roy S, Ravichandiran V, Ghosh DA. Short Review on Glucogallin and its Pharmacological Activities. Mini Rev Med Chem. 2022;22(22):2820-2830. doi: 10.2174/1389557522666220513150907. PMID: 35570562.
23. Puppala M, Ponder J, Suryanarayana P, Reddy GB, Petrash JM, LaBarbera DV. The Isolation and Characterization of β-Glucogallin as a Novel Aldose Reductase Inhibitor from Emblica officinalis. PLoS ONE, 2012; 7(4): e31399.
24. Ahmad S, Pandey AR, Singh SP, Singh S, Sashidhara KV, Tamrakar AK. Antiglycation activity of *β*-glucogallin from *Asparagus racemosus*. Nat Prod Res. 2022;36(24):6329-6335.
25. William W. Christie, Xianlin Han, Chapter 1 - Lipids: their structures and occurrence, Editor(s): William W. Christie, Xianlin Han, In Oily Press Lipid Library Series, Lipid Analysis (Fourth Edition), Woodhead Publishing, 2012, 3-19, ISBN 9780955251245.
26. Białek M, Rutkowska J. The importance of γ-linolenic acid in the prevention and treatment. Adv. Hyg. Exp. Med. 2015; 69:892–904.
27. Timoszuk M, Bielawska K, Skrzydlewska E. Evening Primrose (*Oenothera biennis*) Biological Activity Dependent on Chemical Composition. Antioxidants *(Basel)*. 2018;7(8):108-119
28. Odinga T, Yousuf S, Muhammad CI, Ndukwe IG, Otobo MB, Lemii BC, Enebeli SK, Edward UF. Evaluation of the *in-vitro* biological activities of the seed extract of *Ricinodendron heudelotii*. Trends in Med. Res., 2024, 19(2): 1-12.
29. Senapati S, Sabyasachi B, Gangopadhyay DN. Evening primrose oil is effective in atopic dermatitis: A randomized placebo-controlled trial. Indian J. Dermatol. Venereol. Leprol. 2008; 74:447–452.
30. Białek A, Teryks M, Tokarz A. Sprzężone trieny kwasu linolenowego (conjugated linolenic acid–CLnA, super CLA)–źródła i działanie biologiczne [Conjugated linolenic acids (CLnA, super CLA)--natural sources and biological activity]. Postepy higieny i medycyny doswiadczalnej (Online), 2014; 68, 1238–1250.
31. Yuan GF, Chen XE, Li D. Conjugated linolenic acids and their bioactivities: a review. Food func., 2014; 5(7), 1360–1368.
32. Fontes AL, Pimentel LL, Simões CD, Gomes A, Rodríguez-Alcalá LM. Evidences and perspectives in the utilization of CLNA isomers as bioactive compounds in foods. Critical rev. food sci. nutria., 2017; 57(12), 2611–2622.
33. Hennessy AA, Ross RP, Devery R, Stanton C. The health promoting properties of the conjugated isomers of α-linolenic acid. Lipids, 2011; 46(2), 105–119.
34. Gasmi J, Thomas Sanderson J. Jacaric acid and its octadecatrienoic acid geoisomers induce apoptosis selectively in cancerous human prostate cells: a mechanistic and 3-D structure-activity study. Phytomed.: int. j. phytother. phytopharm, 2013; 20(8-9), 734–742.
35. Gómez-Cortés P, Tyburczy C, Brenna JT, Juárez M, de la Fuente MA. Characterization of cis-9 trans-11 trans-15 C18:3 in milk fat by GC and covalent adduct chemical ionization tandem MS. J Lipid Res. 2009;50(12):2412-2420.