Journal of Chemical Research Advances
ISSN: 2582-7995

Research Article
Open access

# Phytochemical screening and antioxidants activities of four different solvent extracts of *Justicia secunda* leaf extracts

**Bako B[1], [1]Ushie OA, [1]Malu SP and [2]Kendeson AC**
[1]Department of Chemical Science, Federal University Wukari, Nigeria
[2]Department of Chemical Sciences, Federal University of Kashere, Gombe State, Nigeria

**Corresponding author:** ushie@fuwukari.edu.ng

Received on: 15/02/2023 Accepted on: 25/04/2023 Published on: 01/05/2023

## ABSTRACT

**Aim:** This study was aimed to evaluate phytochemical and antioxidant capabilities of the crude extracts of *J. secunda* leaves from four different solvents in increasing polarities, with the idea of isolating some of the metabolites.

**Method and Materials:** *J. secunda* leaves collected from their natural habitat in Wukari Local Government Area of Taraba State, Nigeria, was macerated for 96 hours in hexane, ethyl acetate, acetone, and methanol in increasing polarity; the crude extracts were concentrated at 40 $^{o}$C and used for the analyses. The percentage yields of the crude extracts were as follows: hexane 2.44 %, ethyl acetate 1.24 %, acetone 3.34 %, and methanol 5.33 %.

**Results:** The phytochemical screening of the leaves revealed that it contained secondary metabolites, with tannins and phenols present in all the extracts, terpenoids and phlobatannins were present in ethyl acetate acetone and methanol extracts only, steroids was present on hexane extract only while, anthraquinones was found in only methanol extract. The antioxidant activity using DPPH assay method was concentration dependant and showed that the methanol extract exhibit the highest antioxidant activity within the concentration rang of (0.0313 – 0.5 mg/mL) than the other extracts when compared with that of the standard drug (vitamin C). Among the extracts, methanol had the highest antioxidant potential with an increase in the percentage inhibition of 16.22 – 61.62 % as the sample concentration increased from 0.0313 – 0.5 mg/mL and $IC_{50}$ value of 35.49 mg/mL; acetone had percentage inhibition of 23.24 – 55.14 % (41.73 mg/mL); ethyl acetate had percentage inhibition of 22.16 – 47.57 % (53.31 mg/mL). The least was hexane with an increase in the percentage inhibition of 16.22 – 45.41 % and $IC_{50}$ value of 56.81 mg/mL when compared with that of the standard (vitamin C) having a percentage inhibition increase of 28.11 – 76.22 % and $IC_{50}$ value of 22.76 mg/mL.

**Conclusion:** It was concluded that the crude extracts possess antioxidant activities which may be as a result of the phytochemicals present in the plant giving credence to the use of the plant by most people implying that the plant can be harnessed for it antioxidant potential in developing antioxidant drugs.

**Keywords:** Anti-oxidant, extract, *Justicia secunda,* phytochemical.

**How to cite this article**: Bako B, Ushie OA, Malu SP and Kendeson AC (2023). Phytochemical screening and antioxidants activities of four different solvent extracts of *Justicia secunda* leaf extracts. J. Chem. Res. Adv., 04(01): 15-20.

## Introduction

Plants have been a great source of medicine to many nations due to the presence of metabolites found in them and its numerous advantages of low side effects and availability as compared to the synthetic drugs. The African nations have been abundantly blessed with these plants though highly underutilized for their medicinal properties. Such include *Justicia secunda*, used for ornamental purposes and grown around domestic homes, gardens and offices to enhance the beauty of the area (Ajuru et al., 2022).

*J. secunda* Vahl of the family Acanthaceae is also known as ''Blood root" (Barbados) and ''Sanguinaria" (Venezuela) as reported by Onoja et al. (2017) is commonly found in South-Eastern Nigeria, Congo and South Cote-d'Ivoire and the leaf decoction is consumed for the management of anemia, used for healing wounds, abdominal pains. It is reported to possess phytochemicals, which are attributed to its medicinal properties such as anti-sickling (Carrington et al., 2012), haematinic (Yamoah et al., 2020), antimicrobial (Manda et al., 2011) and anti-hypertensive (Mpiana et al., 2010), antioxidant (Osioma and Hamilton-Amachree, 2017), anti-inflammatory (Anyasor et al., 2019), and antinociceptive (Onoja et al., 2017) activities. Also, the leaves are reported to increase the level of follicle stimulating hormones (FSH) by

the pituitary gland or cells of the hypothalamus producing gonadotrophin releasing hormones (Okpara et al., 2022). This research aimed to evaluate phytochemical and antioxidant capabilities of the crude extracts of *J. secunda* leaves from four different solvents in increasing polarities, with the idea of isolating some of the metabolites.

## Materials and Methods

*Sample Collection and Preparation*

*J. secunda* leaves collected from their natural habitat in Wukari Local Government Area of Taraba State, Nigeria, were identified at the Department of Forestry and Wild Life Sciences, Federal University, Wukari. The fresh leaves were washed with distilled water, cut into smaller pieces then air-dried for three weeks; these were then crushed into powder form using a mortar & pestle and stored in sterile container for further use.

The extraction process was by maceration using four solvents of hexane, ethyl acetate, acetone, and methanol in increasing polarity (Ushie et al., 2022); 100 g of the sample was macerated in 250 mL hexane for 96 hours with frequent agitation. The crude extract was filtered and concentrated using rotary evaporator at 40 $^{o}$C, this process was repeated on the residue for the other solvents (ethyl acetate, acetone, and methanol) and the percentage yields were calculated and noted.

*Phytochemical Screening*

Standard procedures as described by Ushie et al., (2019) to identify the following metabolites: alkaloids, saponins, flavonoids, tannins, terpenoids, cardiac glycoside, steroids, anthraquinones, phenols and phlobatannins.

*Antioxidant Activity*

The antioxidant activity of the crude extracts was determined using DPPH (1,1-diphenyl-2-picryl hydrazyl) free radical scavenging assay with ascorbic acid (vitamin C) as the standard using the procedures described by Kendeson et al., (2021) and Ushie et al., (2019) with some slight modifications in the concentrations of the extract/standard used. 0.1 mM of DPPH in ethanol was prepared and 1.5 mL of this solution was added to 1.5 mL of extract solution in ethanol at different concentrations (0.5, 0.25, 0.125, 0.0625, 0.03125 mg/mL) and mixed thoroughly; these were allowed to stand for 30 minutes in a dark cupboard and the absorbance was measured at 517 nm using a spectrophotometer (V-730 UV-Vis Spectrophotometer, Jasco, USA). The DPPH scavenging activity was calculated as follows:

Percentage (%) inhibition = $A_0 - A_1 / A_0 \times 100$;

Where $A_0$ = the Absorbance of control and $A_1$ = the Absorbance of standard/sample. The (%) inhibition values were plotted against the concentrations of standard/sample and the half maximal inhibitory concentration ($IC_{50}$) values calculated using the linear regression equation (Ushie et al., 2023).

## Results and Discussion

The percentage yield, phytochemical screening and antioxidant activity of the crude extracts of *J. secunda* were recorded. The percentage yield of the crude extracts shown (Table 1 & Fig 1) indicated that all the solvents had varying quantity of plant's components with the highest yield in methanol solvent and ethyl acetate having the lowest, this had to do with the polarity of the solvents. The low yield in ethyl acetate could be as a result of the affinity of the components present in the samples for more polar solvents (Ushie et al., 2023).

The results of the phytochemical analysis (Table 2) revealed that the leaves of *J. secunda* contain secondary metabolites - alkaloids, saponins, flavonoids, tannins, terpenoids, cardiac glycoside, steroids, anthraquinones, phenols and phlobatannins in the extracts in varying degrees with tannins and phenols present in all the extracts, while, anthraquinones was found in only methanol extract. This could be due to the polarity of the solvent. The presence of the metabolites confirms the medicinal attributes of the plant. The result agrees with findings in literature which reported the presence of secondary metabolites in the leaf of this pant (Osioma and Hamilton-Amachree, 2017; Ogunbamowo et al., 2020; Yamoah et al., 2020; Onochie et al., 2020; Ajuru et al., 2022). The presence of these phytochemicals may be responsible for the ethno-medicinal uses of the leaves of this plant.

Antioxidant activity of *J. secunda* carried out using DPPH assay revealed the antioxidant capability of the extracts (Table 3 & 4) in a concentration dependant manner (Fig 2 & 3). It was observed that among the extracts, methanol had the highest antioxidant potential with an increase in the percentage inhibition of 16.22 – 61.62 % as the sample concentration increased from 0.0313 – 0.5 mg/mL and $IC_{50}$ value of 35.49 mg/mL.

Table 1: Nature and % yield of *J. secunda* leaf crude extracts

| Solvent | Nature/colour of the extract | Yield of extracts |
|---|---|---|
| Hexane | Sticky/Black | 2.44 |
| Ethyl acetate | Sticky/Dark Brown | 1.24 |
| Acetone | Solid/Dark Green | 3.34 |
| Methanol | Solid/Green | 5.33 |

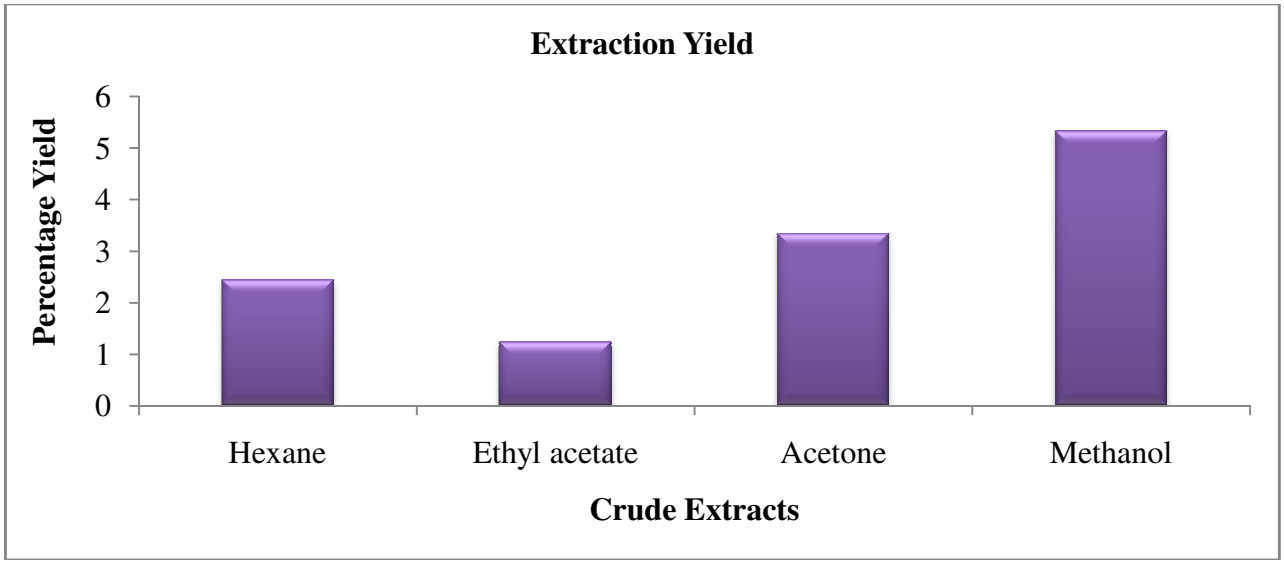


Fig 1: Chart of Percentage (%) Yield of *J. secunda* leaf Crude Extracts

Table 2: Results of Phytochemical Analysis

| S/N | Phytochemical | Crude Extracts | | | |
|---|---|---|---|---|---|
| | | Hexane | Ethyl acetate | Acetone | Methanol |
| 1 | Alkaloids (a) Mayer | + | ++ | - | + |
| | (b) Wagner | + | - | + | - |
| | (c) Dragedroff's | + | + | + | - |
| | (d) Hager | + | + | + | - |
| 2 | Flavonoids | - | + | - | + |
| 3 | Anthraquinones | - | - | - | + |
| 4 | Steroids | + | - | - | - |
| 5 | Glycosides (a) Keller Killani | + | + | - | - |
| | (b) Salkowski | - | + | - | - |
| 6 | Tannins | ++ | + | + | ++ |
| 7 | Terpenoids | - | + | + | + |
| 8 | Saponins (Froth test) | - | - | + | + |
| | (Foam test) | - | + | + | - |
| | (Emulsion test) | - | - | - | + |
| 9 | Phenols | ++ | + | + | + |
| 10 | Phlobatannins | - | + | + | + |

Key: - implies absence of constituents; + implies slight presence of constituents;
++ implies moderate presence of constituents.

Table 3: Absorbance of *J. secunda* leaf extracts and Standard

| Concentration | Absorbance | | | | |
|---|---|---|---|---|---|
| (mg/mL) | Methanol | Acetone | Ethyl acetate | Hexane | Vitamin C |
| 0.0313 | 0.155 | 0.142 | 0.144 | 0.155 | 0.133 |
| 0.0625 | 0.149 | 0.133 | 0.134 | 0.146 | 0.124 |
| 0.125 | 0.127 | 0.125 | 0.128 | 0.141 | 0.108 |
| 0.25 | 0.104 | 0.113 | 0.117 | 0.126 | 0.085 |
| 0.5 | 0.071 | 0.083 | 0.097 | 0.101 | 0.044 |

Blank = 0.185

Table 4: Inhibition (%) for *J. secunda* leaf extracts and Standard

| Concentration (mg/mL) | % Inhibition | | | | |
|---|---|---|---|---|---|
| | Methanol | Acetone | Ethyl acetate | Hexane | Vitamin C |
| 0.0313 | 16.22 | 23.24 | 22.16 | 16.22 | 28.11 |
| 0.0625 | 19.46 | 28.11 | 27.72 | 21.08 | 32.97 |
| 0.125 | 31.35 | 32.43 | 30.81 | 23.78 | 41.62 |
| 0.25 | 43.78 | 38.92 | 36.76 | 31.89 | 54.05 |
| 0.5 | 61.62 | 55.14 | 47.57 | 45.41 | 76.22 |
| $IC_{50}$ | **35.49** | **41.73** | **53.31** | **56.81** | **22.76** |

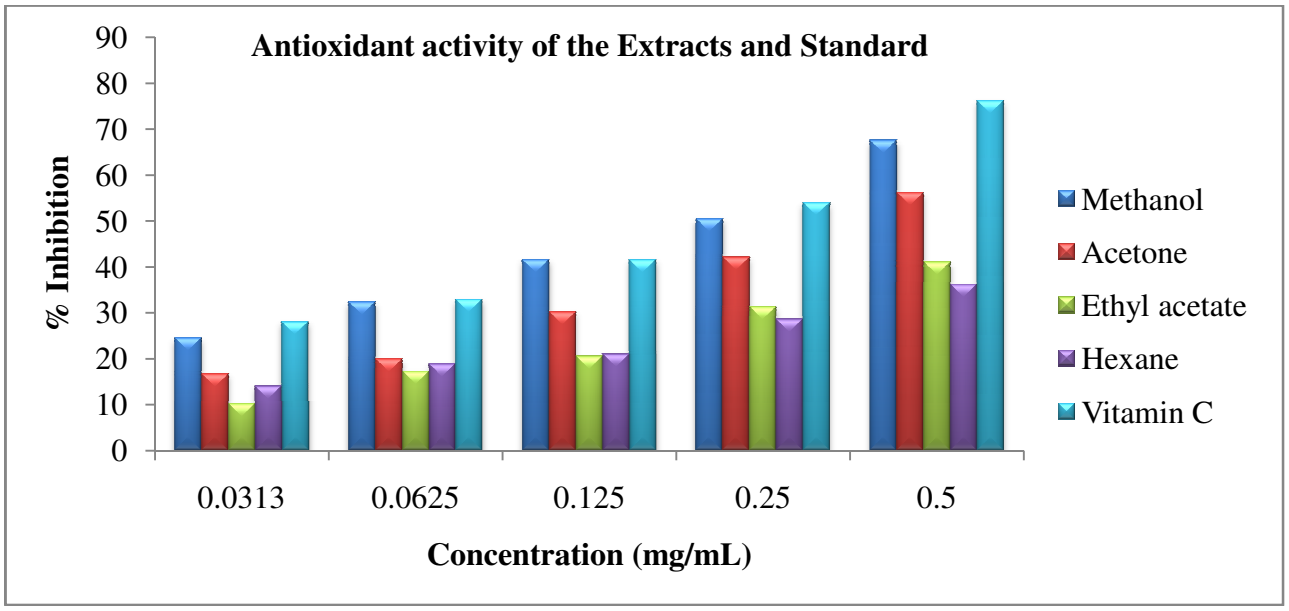


Fig 2: Chart of percentage inhibition of Standard and Crude Extracts of *J. secunda* Leaf

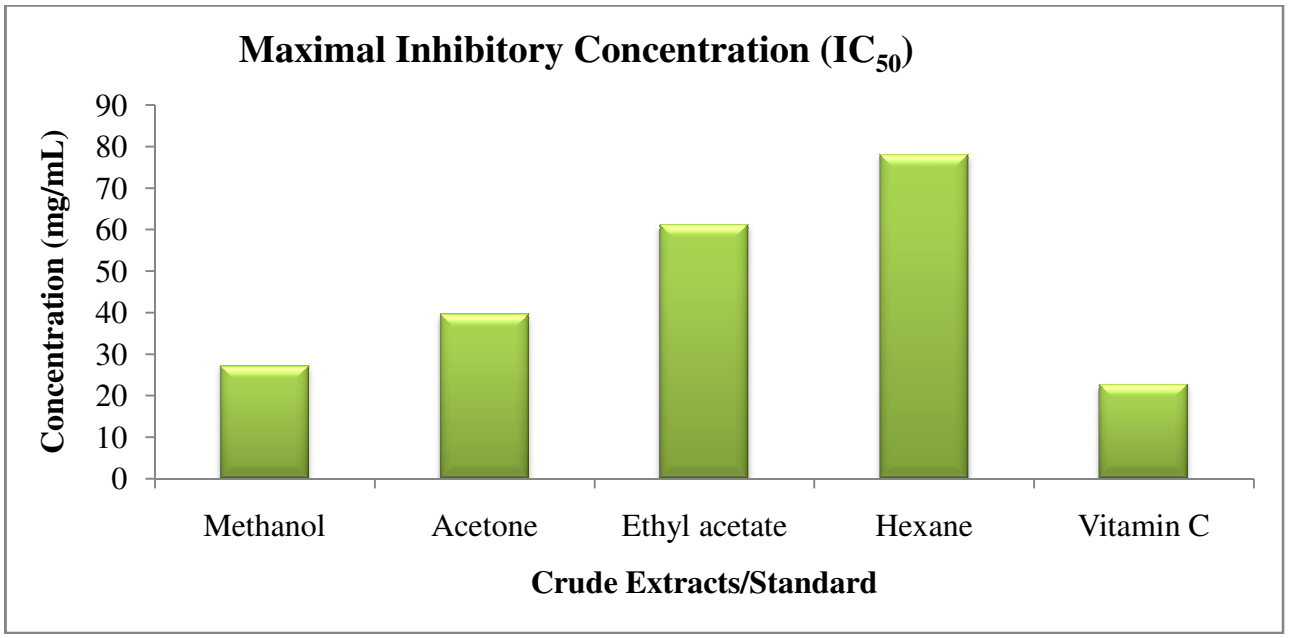


Fig 3: Chart of Maximal Inhibitory Concentration ($IC_{50}$) of Standard and Crude Extracts of *J. secunda* Leaf

The least was hexane with an increase in the percentage inhibition of 16.22 – 45.41 % and $IC_{50}$ value of 56.81 mg/mL when compared with that of the standard (vitamin C) having a percentage inhibition increase of 28.11 – 76.22 % and $IC_{50}$ value of 22.76 mg/mL. This result has shown that leaves of *J. secunda* possess antioxidant activity and supports previous works (Onoja et al., 2016; Osioma and Hamilton-Amachree, 2017). This plant can be harnessed for it antioxidant potential in developing antioxidant drugs.

**Conclusion**

It was concluded that phytochemicals present in the leaves of *J. secunda* and the antioxidant activities which are known to be as result of the presence of these phytochemicals, giving credence to the use of the plant by most.

**Reference**


Ajuru MG, Kpekot AK, Robinson EG and Amutadi CM (2022). Proximate and Phytochemical Analysis of the Leaves of *Justicia carnea* Lindi. and *Justicia secunda* Vahl and its Taxonomic Implications. Journal of Biomedicine and Biosensors, *2*(1): 1-12.

Onoja OS, Ezeja IM, Omeh NY, and Onwukwe CB (2017). Antioxidant, Anti-Inflammatory and Antinociceptive Activities of Methanolic Extract of *Justicia secunda* Vahl leaf. Alexandria Journal of Medicine, *53*: 207–213.

Carrington S, Cohall DH, Gossell-Williams M and Lindo JF (2012). The Antimicrobial Screening of A Barbadian Medicinal Plant With Indications for use in the Treatment of Diabetic Wound Infections. West Ind Med J, *61*(9): 861–4.

Yamoah A, Adosraku RK, Amenu JD, Baah MK and Abaye DA (2020). Evaluation of the Haematinic Activities of Extracts of *Justicia secunda* Vahl Leaves in Red Blood Cells of Laboratory Rats. Journal of Biosciences and Medicines, 8, 48-57. https://doi.org/10.4236/jbm.2020.83006

Manda P, Abrogoua DP, Bahi C, Dano DS, Gnahoui G and Kablan BJ (2011). Evaluation of the antihypertensive activity of total aqueous extract of *Justicia secunda* Valh (Acanthaceae). Afric J Pharm Pharmacol, *5*(16): 1838–45.

Mpiana PT, Bokota MT, Ndjele MBL, Mudogo V, Tshibangu KN and Ngbolua KN (2010). Antisickling activity of three species of Justicia from Kisangani (DR Congo): J. tenella, J. gendarussa and J. insularis. Int J Biol Chem Sci, *4*(6): 1953–61.

Osioma E and Hamilton-Amachree A (2017). Comparative Study on the Phytochemical and *In vitro* Antioxidant Properties of Methanolic Leaf Extract of *Justicia secunda* Vahl. Nigerian Journal of Science and Environment, *15*(1): 111- 118.

Anyasor NG, Okanlawon AA and Ogunbiyi B (2019). Evaluation of anti-inflammatory activity of Justicia secunda Vahl leaf extract using in vitro and in vivo inflammation models. Clinical Phytoscience, 5:49. https://doi.org/10.1186/s40816-019-0137-8

Okpara EP, Victor PD, Reuben E, Ajie PC and Fiala LT (2022). Effect of ethanolic extract of *Justicia secunda* (blood root) leaves on reproductive organs and hormones in female Wistar rats. Int J Res Med Sci, 10:1884-8.

Ushie OA, Azuaga TI, Aikhoje EF, Neji PA, Abeng FE and Adashu JM (2019). Antioxidant activities of Hexane, Chloroform, Acetone and Methanol Extract of *Swietenia macrophylla*. International Journal of Clinical Chemistry and Laboratory Medicine, (IJCCM), **5**(4), 6-10. https://dx.doi.org/10.20431/2455-7153.0504002.

Kendeson AC, Kagoro ML and Adelakun AE (2021). Phytochemical and Pharmacological evaluation of Nigerian *Kalanchoe pinnata* (Lam.) Stem-bark. J. Chem. Soc. Nigeria, 46(4): 0751-0756.

Ushie OA, Kendeson AC, Longbab BD and Galadima A (2023). Antioxidants Activities of Four Different Solvent Extracts of *Corchorus tridens* Leaf using DPPH free Radical Scavenging Assay. J. Chem. Res. Adv., 04(01): 10-14.

Ogunbamowo OP, Olaniyi BM, Awotedu LO and Lawal OI (2020). Assessment of the Foliar Micromorphology, Phytochemical and Mineral Composition of *Justicia secunda* Valh Leaves. Anales de Biología, 42: 183-192. http://dx.doi.org/10.6018/analesbio.42.20

Onochie UA, Adaobi Helen, Oli NA, Ezeigwe CO, Nwaka CA, Okani OC, Okam CP,

Ihekwereme PC and Okoyeh NJ (2020). The Pharmacobiochemical Effects of Ethanol Extract of *Justicia secunda* Vahl Leaves in Rattus Norvegicus. Journal of Experimental Pharmacology, 12: 423–437. http://doi.org/10.2147/JEP.S267443.

******