

**Phytochemical, antioxidant, cytotoxicity and antimicrobial investigations of
Tephrosia platycarpa GUILL.&PERR.**

ABSTRACT

Tephrosia genus belongs to the family Fabaceae and many plants in this genus have been reported to possess antimicrobial, anti-inflammatory, pesticidal and insecticidal properties, however; there is paucity of information on the chemical constituents and biological activities of *Tephrosia platycarpa*. Thus, this study was aimed at investigating the phytochemical composition and biological properties of *Tephrosia platycarpa*. Fresh samples of *Tephrosia platycarpa* were collected at Ile'gbon village, Ibadan, Southwest Nigeria and authenticated at the Herbarium Unit of the Forestry Research Institute of Nigeria with the Voucher number - FHI 112653. The plants were separated, air-dried and pulverized. Roots, stems and leaves were extracted with n-hexane, ethyl acetate and methanol successively by maceration, and the percentage yields calculated. The extracts were screened for the presence of phytochemicals using standard methods. Antioxidant activity of the extracts was determined using Hydrogen Peroxide scavenging assay with Vitamin A, Ascorbic acid and Butylated Hydroxyl Anisole as controls. The cytotoxicity of the extracts was determined through brine shrimp lethality assay at six concentrations, GraphPad Statistical package was used to obtain the Lethal Concentration at 50%. Antimicrobial susceptibility, minimum inhibitory concentration and minimum microbicidal concentration assays were carried out on the extracts using agar well diffusion and broth dilution methods. The microorganisms used are *Escherichia coli*, *Pseudomonas aeruginosa*, *Staphylococcus aureus*, *Bacillus subtilis*, *Candida albicans* and *Trichophyton rubrum*). Phytochemical screening of the crude extracts of *Tephrosia platycarpa* showed that the plant is rich in secondary metabolites including alkaloids, flavonoids, tannins, cardiac glycosides and steroids. The antioxidant assay using Hydrogen Peroxide method showed that extracts of the stems and leaves have strong activities against free radicals. Cytotoxicity assay revealed that the extracts of the stem are moderately toxic while roots extracts are highly toxic. The extracts displayed broad spectrum antimicrobial activity against the organisms tested; stem ethyl acetate extract being the most potent.

Keywords- Antimicrobial, antioxidant, cytotoxicity, phytochemicals, *Tephrosia platycarpa*

INTRODUCTION

Generally, phytochemicals can be described as plant chemicals that protect plant cells from environmental hazards such as pollution, stress, drought, UV exposure and pathogenic organisms (Hasler and Blunberg, 1999). They are naturally occurring chemical compounds found in plants which provide health benefits for humans in addition to macronutrients and micronutrients (King and Young, 1999). They also contribute to the plant's colour, aroma and flavour.

These compounds are known as secondary plant metabolites and have biological properties such as antioxidant activity, antimicrobial effect, modulation of detoxification enzymes, stimulation of the immune system, decrease of platelet aggregation and modulation of hormone metabolism and anticancer property (Dapkevicius *et al.*, 2009).

Tephrosia genus belongs to the family Fabaceae (Leguminosae) and subfamily Papilionaceae, which are made up of more than 350 species. The plants in this genus are chiefly distributed in the tropical, subtropical, and arid regions of the world (Al-Ghamdi, 2013). The plants are erect herbs, or soft woody shrubs. The bioactivity associated with the plants have been studied extensively; indicating the phytoconstituents present in the *Tephrosia* genus manifested various biological activities such as anti-diabetic, anti-ulcer, antidiarrheal, wound healing, anti-inflammatory, insecticidal, antiviral, anti-protozoal, anti-fungal, anti-plasmodial and many other activities (Chen *et al.*, 2014).

Tephrosia platycarpa is a leguminous and herbaceous annual plant with found in waste places and road sides. It furnishes grazing for cattle, sheep and horses. The seed is oil bearing, yielding cooking oil used in the Kordofan of Sudan.

MATERIALS AND METHODS

Plant Collection and Preparation

Tephrosia platycarpa were collected at Ile'gbon village in Lagelu local government, Southwest Nigeria. Samples of the plant was deposited at the Herbarium unit, Department of Taxonomy, Forestry Research Institute of Nigeria (FRIN) where it was authenticated and given Voucher No.: FHI. 112653. The plant was separated into the different parts -leaves stems and roots- air-dried for three weeks and pulverised.

Solvent Extraction- Extraction of each of the parts was done by maceration successively with hexane, ethyl acetate and methanol in increasing order of polarity. The extraction lasted for 72 hours in each solvent. The extracts collected were concentrated using rotary evaporator at 40°C. The extracts were weighed and percentage yield calculated.

Phytochemical screening to test for plant secondary metabolites was done according to the method described and modified by Harborne and Baxter, 1999.

Alkaloids: A few drops of Dragendorff's reagent were added to 1mL of the extract in a test tube. Appearance of orange colour indicates the presence of alkaloids.

Flavonoids: To the plant extract, 5 ml of 20% NaOH and HCl was added. A yellow solution indicated the presence of flavonoids.

Tannins: The extract was treated with 15% ferric chloride solution. The resultant colour was observed. A blue colour indicated the presence of hydrolysable tannins. A second confirmatory test was carried out using freshly prepared 10 mL potassium hydroxide. A dirty precipitate indicated the presence of tannins.

Saponins: The plant extract was shaken with water in a test tube. Frothing which persisted on warming was a positive indication of saponins.

Triterpenoids and Steroids: The extract was dissolved in 3 mL of methanol and then 0.2 mL each of chloroform, glacial acetic acid and concentrated sulphuric acid were then added. The solution was then observed for colour change. The appearance of a greenish blue or purple pink colour indicated the presence of sterols or triterpenes, respectively.

Glycosides: To a portion of the plant extract, 2 mL of glacial acetic acid and 1 drop of ferric chloride solution were added. Then 1mL of concentrated sulphuric acid was added. A violet brownish ring below the interface followed by the formation of a greenish ring in the acetic acid layer indicated the presence of glycosides.

Antioxidant Assay of *Tephrosia platycarpa* extracts

Hydrogen peroxide scavenging assay- The modified method of Hazra *et al*, 2008 was used for the hydrogen peroxide scavenging assays of the crude extracts. The stock solution was prepared by adding 0.6mL of 4mM hydrogen peroxide to 250 mL of 0.1M phosphate buffer (pH 7.4). Three millilitres of the solution were added to 1 mL of the extracts and standards; and incubated for 15 minutes at room temperature. The absorbance was read using UV/Vis Spectrophotometer at 285 nm and the percentage inhibition of the hydrogen peroxide was calculated as;

$$\text{H}_2\text{O}_2 \text{ scavenging activity (\%)} = \frac{\text{Abs}_{\text{control}} - \text{Abs}_{\text{sample}}}{\text{Abs}_{\text{control}}} \times 100$$

Where Abs control is the absorbance of H₂O₂ radicals, Abs sample is the absorbance of H₂O₂ radical + extract or standard.

Cytotoxicity Assay (Brine Shrimp Lethality Assay) of *Tephrosia platycarpa* extracts

The brine shrimp eggs were hatched in a bowl with two compartments, one side illuminated and the other un-illuminated for 72 hours. The extracts were prepared in six concentrations. In preparing the stock solution of the crude extracts, 6 mg of each of the extracts was dissolved in 1 mL 1% DMSO and made up to 6 mL with water to give 1000 µg/mL concentration. Three millilitre was drawn into another vial and made up with water to 6mL to give 500 µg/mL concentration. The same process of double fold dilution was done to give 250, 125, 62.5 and 31.3 µg/mL concentrations respectively. From each of the prepared concentrations of 3mL, 1mL is drawn into two different vials to have each concentration in triplicates of 1 mL each.

A blank solution was also prepared using just two drops of 1% DMSO and 5 mL of sea water. Ten active brine shrimps (nauplii) were added to all the solutions using a Pasteur drop pipette, and made up to 5 mL using sea water. The experiment was set up for 24 hours after which the number of active and dead nauplii in each concentration was counted with the use of a magnifying glass and transparent dish under light.

The mortality endpoint of this bioassay is defined as the absence of controlled forward motion during 30 sec of observation. The lethal concentration at 50% was determined by Clarkson's toxicity index using GraphPad Statistical package.

$$\text{Percentage Mortality} = \frac{\text{Total nauplii} - \text{Active nauplii}}{\text{Total nauplii}} \times 100$$

Antimicrobial Assay

The antimicrobial susceptibility and minimum inhibitory concentration of the crude extracts obtained from *Tephrosia platycarpa* were carried out using the agar well diffusion and broth dilution method respectively. Two gram-positive bacteria (*Staphylococcus aureus* and *Bacillus subtilis*), two gram-negative bacteria (*Escherichia coli* and *Pseudomonas aeruginosa*) and two fungi (*Candida albicans* and *Trichophyton rubrum*) were used for the assay. They were clinical isolates obtained from the Department of Pharmaceutical Microbiology, Faculty of Pharmacy, University of Ibadan, Nigeria. Gentamycin, ketoconazole and methanol were used as reference standards for positive and negative controls respectively. Double fold dilution of the extracts was

prepared to obtain five concentrations of the samples (100 - 6.25 mg/mL) using methanol as the solvent.

Susceptibility test was carried out using the agar well diffusion method. The organisms were sub-cultured overnight at 37°C in sterile nutrient broth and diluted in distilled water, 0.2 mL of the diluted organism was taken into the sterile nutrient agar and poured into sterile petri dishes and left to solidify for about 45-60 minutes. Uniform wells were made according to the number of the graded concentrations of the samples using sterile cork borer of 8 mm diameter. In each of the wells, the samples were added and allowed to stay on the bench for 2 hours to allow extracts to diffuse properly into the agar. The plates were incubated uprightly in the incubator for 24 hours for bacteria and 48 hours for fungi susceptibility test. The diameters (zones of inhibition) were then measured in mm after the incubation period using a graduated ruler.

Minimum Inhibitory Concentration of the extracts was determined using broth dilution method with Muller-Hinton broth used as diluents. The prepared concentration of the extracts was diluted with broth (2 mL of extract concentration was added to 18 mL of broth) to give; 10, 5, 2.5, 1.25 and 0.625 mg/mL respectively. The organisms were inoculated into each tube containing the broth and the extract; and poured into sterile petri dish, left on the bench for 1 hour. The inoculated petri dishes were incubated at 37°C for 48 hours. At the end of the incubation period, the dishes were examined for the presence or absence of growth (turbidity), the lowest concentration in the series of each of the extracts without visible sign of growth was considered to be the minimum inhibitory concentration.

Minimum microbicidal concentration was also determined by re-culturing of plates without visible growth and incubated for 48 hours after which the plates are examined, and the lowest in the series without any visible growth was also considered to be the minimum microbicidal concentration.

RESULTS AND DISCUSSION

The yield and appearance of the extracts of *Tephrosia platycarpa* are presented in Table 1. The methanol extracts have the highest yield as expected.

Table 1: Yield and appearance of *Tephrosia platycarpa* extracts

<i>Tephrosia platycarpa</i> parts	Weight of parts (g)	Yield of extract (g)	% Yield of extract	Colour of extract
TPRH	1440	7.58	0.53	Grey
TPRE	„	18.1	1.26	Reddish brown
TPRM	„	25.0	1.74	Dark-Brown
TPSH	4000	23.6	0.59	Light green
TPSE	„	37.7	0.94	Orange
TPSM	„	85.4	2.14	Dark- Brown
TPLH	430	6.5	1.5	Light-green
TPLE	„	10.0	2.3	Green
TPLM	“	39.0	9.1	Green

Keys:

TPRH- *Tephrosia platycarpa* root hexane extract, TPRE- Root ethyl acetate extract, TPRM- Root methanol extract, TPSH- stem hexane extract, TPSE- Stem ethyl acetate extract, TPSM- Stem methanol extract, TPLH- Leaves hexane extract, TPLE- Leaves ethyl acetate extract, TPLM- Leaves methanol extract

Phytochemical Screening of *Tephrosia platycarpa* extracts

Phytochemical screening of the nine crude extracts of *Tephrosia platycarpa* revealed the presence of alkaloids, flavonoids and steroids in all the extracts. Tannins were present in all the extracts except hexane of both roots and stem; and ethyl acetate of the roots. Saponins were only found in the three extracts of the leaves while glycosides were found in all but ethyl acetate extract of the stem.

The presence of alkaloids and flavonoids in all the extracts is similar to the result obtained from aqueous and chloroform extracts of *Tephrosia platycarpa* by Ambugus *et al.* (2018). Alkaloids have anti-inflammatory and analgesic functions, helps alleviate pains, develop resistance against diseases and endurance against stress (Gupta, 1994). Tannins are phenolic compounds and their derivatives are also considered as primary anti-oxidants or free radical scavengers important for protecting against cellular oxidative damages including lipid peroxidation. The tannins and flavonoids are known to have curative activities against several pathogens and therefore could be used for the treatment of various illnesses. Plant steroids are essential for normal growth and development; and also serve as natural fertiliser for plant development in agriculture (Patel and

Savjani, 2015) Flavonoids are also known to possess anti-inflammatory, antimicrobial and anti-cancer activities.

Table 2: Phytochemical screening of *Tephrosia platycarpa* extracts

Phytochemicals	TPRH	TPRE	TPRM	TPSH	TPSE	TPSM	TPLH	TPLE	TPLM
Alkaloids	+	+	+	+	+	+	+	+	+
Flavonoids	+	+	+	+	+	+	+	+	+
Tannins	-	-	+	-	+	+	+	+	+
Saponins	-	-	-	-	-	-	+	+	+
Steroids	-	+	+	+	+	+	+	+	-
Glycosides	+	+	+	+	-	+	+	+	+

Keys: + = Present, - = Absent

TPRH- *Tephrosia platycarpa* Root hexane extract, TPRE- Root ethyl acetate extract, TPRM- Root methanol extract, TPSH- stem hexane extract, TPSE- Stem ethyl acetate extract, TPSM- Stem methanol extract, TPLH- Leaves hexane extract, TPLE- Leaves ethyl acetate extract, TPLM- Leaves methanol extract

Antioxidant assay of *Tephrosia platycarpa* extracts

The results showing the scavenging activities of *Tephrosia platycarpa* extracts are given in Table 3. The ability of the extracts to scavenge free radicals of Hydrogen Peroxide was investigated using known standards – Vitamin A, Ascorbic acid and Butylated Hydroxyl Anisole-, the percentage inhibition decreased with decrease in concentrations. The ethyl acetate and methanol extracts of the leaves have the highest activities while hexane extract of the roots has the lowest activity when compared with the standards used. The result suggested that *Tephrosia platycarpa* has strong free radical scavengers and can be seen as a potential source of natural antioxidants for medicinal and commercial purposes.

Table 3: Percentage Inhibition of Hydrogen peroxide radicals by *Tephrosia platycarpa* extracts

Conc.(mg/mL)	TPRH	TPRE	TPRM	TPSH	TPSE	TPSM	TPLH	TPLE	TPLM	VitA	AscA	BHA
1.0	68.1	71.0	70.8	70.4	71.2	71.0	70.6	71.2	71.2	74.3	76.6	66.9
0.5	49.9	69.3	70.8	62.8	71.0	69.6	68.8	71.2	71.2	67.9	76.5	51.1
0.25	42.8	66.3	68.6	59.9	66.9	60.4	62.9	70.7	70.7	20.7	74.6	31.8

Keys:

n=3

TPRH- *Tephrosia platycarpa* Root hexane extract, TPRE- Root ethyl acetate extract, TPRM- Root methanol extract, TPSH- stem hexane extract, TPSE- Stem ethyl acetate extract, TPSM- Stem methanol extract, TPLH- Leaf hexane extract, TPLE- Leaf ethyl acetate extract, TPLM- Leaf methanol extract, BHA- Butylated Hydroxyl Anisole
AscA- Ascorbic acid

Cytotoxicity (Brine Shrimp Lethality) assay of *Tephrosia platycarpa* extracts

The cytotoxicity of the extracts of *Tephrosia platycarpa* was determined by brine shrimp lethality assay. The percentage mortality of crude extracts on brine shrimps were presented in figures 1, 2 and 3. There was gradual decrease in the percentage death of the nauplii with decrease in concentration i.e. the mortality rate is concentration dependent.

Clarkson's toxicity criterion for the toxicity assessment of plant extracts classifies extracts in the following order: extracts with LC₅₀ above 1000 µg/mL are non-toxic, LC₅₀ of 500 - 1000 µg/mL are of low toxicity, extracts with LC₅₀ of 100 - 500 µg/mL are moderately toxic, while extracts with LC₅₀ of 0 - 100 µg/mL are highly toxic (Clarkson *et al.*, 2004). The lethality concentrations (LC₅₀) of the hexane, ethyl acetate and methanol extracts of the roots were 83.1, 88.4 and 97.1 µg/mL respectively. These values are relatively close when compared to the positive control-cyclophosphamide (63.82 µg/mL) and this suggests that the roots extracts are highly toxic. The LC₅₀ for the stem extracts 363.6, 176.8 and 184.6 µg/mL, respectively, while that of the leaves are 328.3, 202.3 and 210.0 µg/mL implying that the stem and leaves extracts are moderately toxic. Natural products that are toxic to brine shrimps (*Artemia salina*) are said to be medicinally active. Based on the results obtained, the cytotoxicity of the extracts could be attributed to the presence of flavonoids, steroids and alkaloids detected in the extracts. There are no previous reports on the cytotoxicity of *Tephrosia platycarpa* plant in the literature.

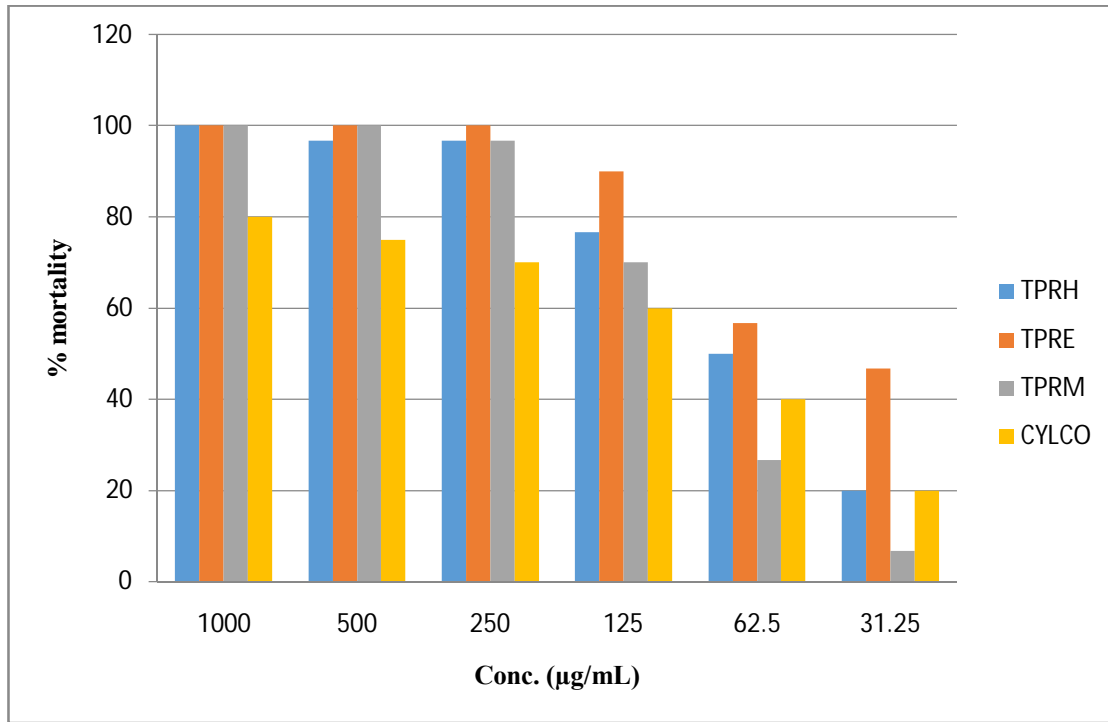


Figure 1: Percentage Mortality of *T. platycarpa* Root Extracts on Brine Shrimps

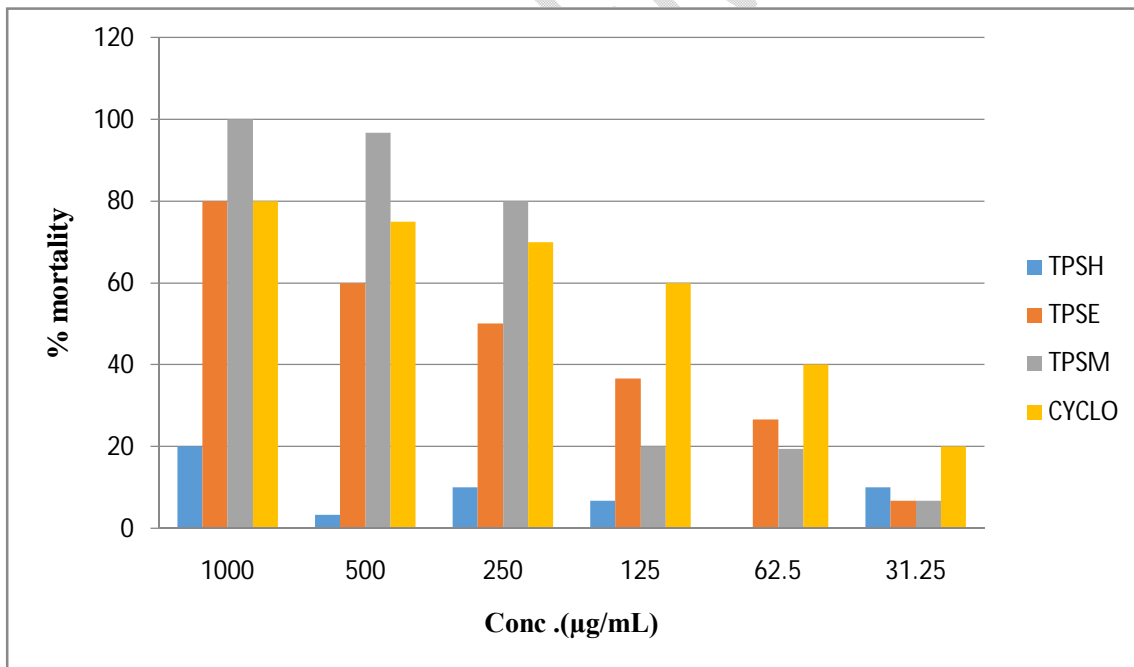


Figure 2: Percentage Mortality of *T. Platycarpa* Stem Extracts on Brine Shrimps

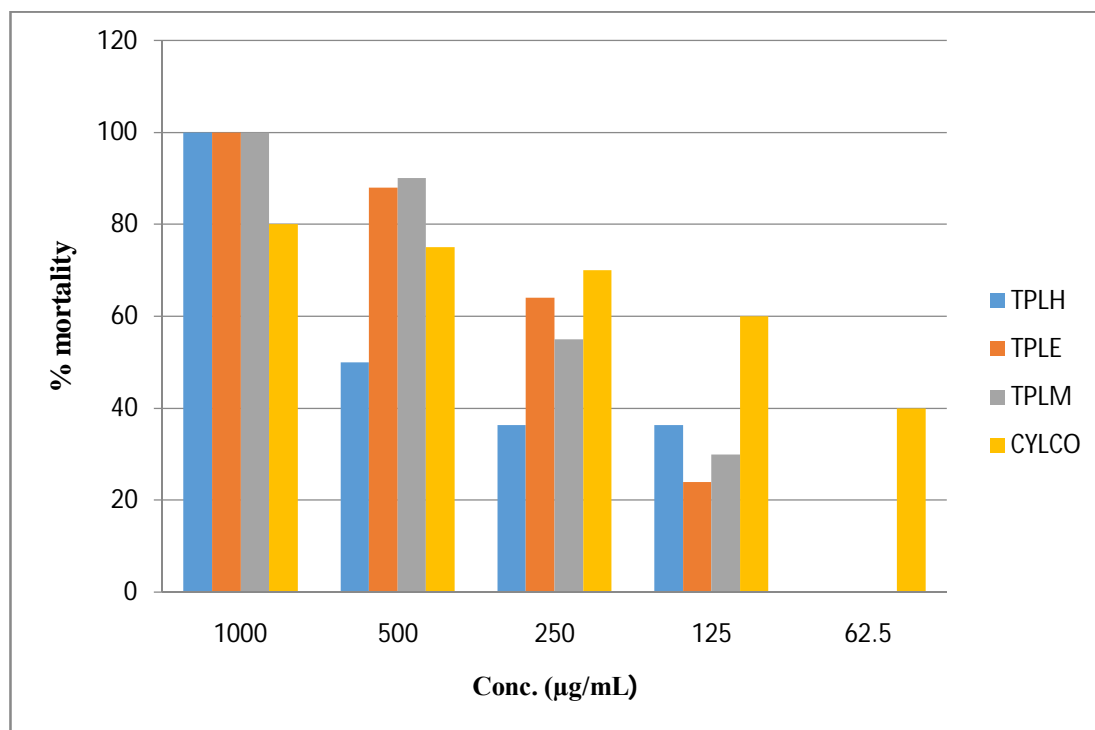


Figure 3: Percentage Mortality of *T. platycarpa* Leaves Extracts on Brine Shrimps

Table 4: Cytotoxicity of *Tephrosia platycarpa* extracts (Lethal Concentration at 50% (LC₅₀) using Pearson Tests)

Extracts	LC ₅₀ (µg/mL)
TPRH	83.10±0.81
TPRE	88.39±0.29
TPRM	97.10±0.41
TPSH	363.6±0.30
TPSE	176.8±0.30
TPSM	184.6±0.50
TPLH	328.3±0.46
TPLE	202.3±0.57
TPLM	210.0±0.64
CYLCO	63.82±0.02

Pearson Test, $P \leq 0.05$

Keys; TPRH- Root hexane extract, TPRE- Root ethylacetate extract, TPRM- Root methanol extract, TPSH- Stem hexane extract, TPSE- Stem ethyl acetate extract, TPSM- Stem methanol extract, TPSO- Essential oil of stem, TPLH- Leaf hexane extract, TPLE- Leaf ethylacetate extract, TPLM- Leaf methanol extract, CYLCO- Cyclophosphamide

Antimicrobial Activity of *Tephrosia platycarpa* extracts

Four bacteria and two fungi isolates were used for the antimicrobial assay. Susceptibility of these microorganisms to the extracts was recorded as diameter of zones of inhibition in millimetre while the MIC and MMC were determined to be the minimum concentration at which the extract showed activity on the microbes (Tables 5-9).

In the roots extracts (Table 5); all the tested organisms are susceptible to the extracts at both concentrations of 50 and 100mg/mL except *Staphylococcus aureus* which is not at 50 mg/mL. In the stems extracts (Table 6); it was observed that all the tested organisms were susceptible and while *Candida albicans* has the highest susceptibility at both concentrations of the ethyl acetate extract. It suggests that TPSE has a high antifungal activity. There was lower susceptibility of the organisms to the leaves extracts (Table 7) when compared to both the roots and stem extracts, however, *Bacillus subtilis* and *Trichophyton rubrum* are susceptible to the leaves extracts.

As it was observed in the susceptibility assay, the extracts of the roots and stems were able to inhibit microbial growth (Table 8) with their ethyl acetate having the highest activities at the minimum concentrations of 1.3 mg/mL against all the organisms except *Pseudomonas aeruginosa* which is at 2.5 mg/mL. The extracts of the leaves have low activities at the tested concentrations. The resulted observed in the minimum inhibitory concentrations are similar to that of the minimum microbicidal concentrations (Table 9).

All the tested extracts exhibited significant antimicrobial activity against the test bacterial and fungal isolates at different concentrations. Ethyl acetate extract of *Tephrosia platycarpa* stem shows the highest activity against all the tested isolates. The activities of the extracts are comparable to the positive control; gentamicin for bacteria and ketoconazole for the fungi.

The *Tephrosia platycarpa* extracts inhibited the growth of *Staphylococcus aureus*, *Bacillus subtilis*, *Pseudomonas aeruginosa*, *Escherichia coli*, *Candida albicans* and *Trichophyton rubrum* at the diluted concentrations ranging from 10 mg/mL to 1.25 mg/mL, the isolates are resistant at lower concentrations. Ethyl acetate extract of both the stem and root have higher inhibitory capabilities than the other extracts.

Table 5: Antimicrobial Susceptibility Test of *Tephrosia platycarpa* Root Extracts**[Diameter of Zones of inhibition (mm)]**

Test organism	TPRH		TPRE		TPRM 100		+vecont. Gent.10 μ g/mL	Keto. 1%	-ve cont. MeOH
	100 mg/mL	50 mg/mL	100 mg/mL	50 mg/mL	mg/mL	50 mg/mL			
<i>S. aureus</i>	18	16	12	08	18	14	12	ND	-
<i>B. Subtilis</i>	16	14	22	14	14	12	12	ND	-
<i>P. aeruginosa</i>	12	10	16	14	14	10	10	ND	-
<i>E. coli</i>	16	14	20	18	16	14	20	ND	-
<i>C. albicans</i>	20	12	20	18	20	16	ND	20	-
<i>T. rubrum</i>	12	10	14	10	16	14	ND	16	-

Keys:TPRH- Root hexane extract, TPRE- Root ethylacetate extract, TPRM- Root methanol extract, Positive control- Gentamicin 10 μ g/ml and 1% ketoconazole, Negative control- distilled methanol, *S. aureus* - *Staphylococcus aureus*, *B. Subtilis* - *Bacillus subtilis*, *P. aeruginosa* - *Pseudomonas aeruginosa*, *E. coli* - *Escherichia coli*, *C. albicans* - *Candida albicans*, *T. rubrum* - *Trichophyton rubrum*, ND- Not Determined

Table 6: Antimicrobial Susceptibility Test of *Tephrosia platycarpa* Stem**[Diameter of Zones of inhibition (mm)]**

Test organism	TPSH		TPSE		TPSM 100		+vecont. Gent.10 μ g/mL	Keto. 1%	-ve cont. MeOH
	100 mg/mL	50 mg/mL	100 mg/mL	50 mg/mL	mg/mL	50 mg/mL			
<i>S. aureus</i>	12	10	14	12	16	14	12	ND	-
<i>B. Subtilis</i>	16	14	22	14	14	12	12	ND	-
<i>P. aeruginosa</i>	12	10	16	14	14	10	10	ND	-
<i>E. coli</i>	14	10	20	18	12	20	20	ND	-
<i>C. albicans</i>	24	20	36	32	22	18	ND	20	-
<i>T. rubrum</i>	16	12	22	18	18	14	ND	16	-

Keys:TPSH- Stem hexane extract, TPSE- Stem ethyl acetate extract, TPSM- Stem methanol extract, Positive control- Gentamicin 10 μ g/ml and 1% ketoconazole, Negative control- distilled methanol, *S. aureus* *Staphylococcus aureus*, *B. Subtilis* - *Bacillus subtilis*, *P. aeruginosa* - *Pseudomonas aeruginosa*, *E. coli* - *Escherichia coli*, *C. albicans* - *Candida albicans*, *T. rubrum* - *Trichophyton rubrum*, ND- Not Determined

Table 7: Antimicrobial Susceptibility Test of *Tephrosia platycarpa* Leaves**[Diameter of Zones of inhibition (mm)]**

Test organism	TPLH		TPLE		TPLM 100		+vecont. Gent.10 μ g/mL	Keto. 1%	-ve cont. MeOH
	100 mg/mL	50 mg/mL	100 mg/mL	50 mg/mL	mg/mL	50 mg/mL			
<i>S. aureus</i>	08	08	16	12	12	10	14	ND	-
<i>B. Subtilis</i>	14	10	12	10	16	14	22	ND	-
<i>P. aeruginosa</i>	12	10	08	08	14	10	20	ND	-
<i>E. coli</i>	08	08	08	08	08	08	18	ND	-
<i>C. albicans</i>	12	10	14	12	14	10	ND	16	-
<i>T. rubrum</i>	08	08	16	12	12	08	ND	14	-

Keys:TPLH- Root hexane extract, TPLE- Root ethylacetate extract, TPLM- Root methanol extract, Positive control- Gentamicin 10 μ g/ml and 1% ketoconazole, Negative control- distilled methanol, *S. aureus* - *Staphylococcus*

aureus, *B. Subtilis* - *Bacillus subtilis*, *P. aeruginosa* - *Pseudomonas aeruginosa*, *E. coli* - *Escherichia coli*, *C. albicans* - *Candida albicans*, *T. rubrum* - *Trichophyton rubrum*,
 ND – Not Determined

Table 8: Minimum Inhibitory Concentration of *Tephrosia platycarpa*

Extracts	<i>S. aureus</i>	<i>B. subtilis</i>	<i>P.aeruginosa</i>	<i>E. coli</i>	<i>C. albicans</i>	<i>T.rubrum</i>
TPRH	5.0	5.0	10.0	5.0	2.5	5.0
TPRE	1.3	1.3	1.3	1.3	1.3	2.5
TPRM	2.5	1.3	1.3	1.3	2.5	2.5
TPSH	5.0	0.6	5.0	0.6	0.6	0.6
TPSE	1.3	1.3	2.5	1.3	1.3	1.3
TPSM	2.5	2.5	5.0	0.6	5.0	5.0
TPLH	NA	5.0	5.0	NA	5.0	NA
TPLE	2.5	1.3	NA	NA	2.5	5.0
TPLM	NA	NA	5.0	NA	5.0	10.0

Concentrations (mg/mL)

NA- No activity

Keys:TPSH- Stem hexane extract, TPSE- Stem ethyl acetate extract, TPSM- Stem methanol extract, TPRH- Root hexane extract, TPRE- Root ethyl acetate extract, TPRM- Root methanol extract, TPLH- Root hexane extract, TPLE- Root ethylacetate extract, TPLM- Root methanol extract

Table 9 Minimum Microbicidal Concentration of *Tephrosia platycarpa*

Extracts	<i>S. aureus</i>	<i>B. subtilis</i>	<i>P.aeruginosa</i>	<i>E. coli</i>	<i>C. albicans</i>	<i>T. rubrum</i>
TPRH	5.0	5.0	10.0	10.0	2.5	5.0
TPRE	1.3	1.3	2.5	1.3	2.5	2.5
TPRM	2.5	2.5	1.3	1.3	5.0	2.5
TPSH	5.0	2.5	5.0	0.6	1.3	1.3
TPSE	1.3	2.5	5.0	2.5	1.3	2.5
TPSM	2.5	2.5	10.0	2.5	5.0	10.0
TPLH	NA	10.0	NA	NA	5.0	NA
TPLE	5.0	5.0	NA	NA	5.0	10.0
TPLM	NA	NA	10.0	NA	10.0	NA

Concentrations (mg/mL)

NA- No activity

Keys;TPSH- Stem hexane extract, TPSE- Stem ethyl acetate extract, TPSM- Stem methanol extract, TPRH- Root hexane extract, TPRE- Root ethyl acetate extract, TPRM- Root methanol extract, TPLH- Root hexane extract, TPLE- Root ethylacetate extract, TPLM- Root methanol extract

CONCLUSION

The phytochemical screening of *Tephrosia platycarpa* extracts revealed the presence of alkaloids, tannins, terpenoids, flavonoids, steroids and cardiac glycosides. The cytotoxicity results of the extracts showed that they are highly toxic to brine shrimps' larvae from the LC₅₀ values obtained from the Clarkson's toxicity index. The crude extracts also displayed broad spectrum of antimicrobial activity.

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