**Antiacterial Effects of Combined Fractions Of *Jatropha curcas* And *Ricinus communis* Oils Extract Against Selected Bacteria**

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**Abstract**: Antiacterial effects of combined fractions of *Jatropha curcas* and *Ricinus communis* oils extracts were determined against the bacterial strains using agar well diffusion technique. The minimum inhibitory concentration (MIC) was investigated by broth dilution technique. Chloramphenicol and Ampicillin were used as standard controls against the Gram negative and Gram positive test organisms respectively. The two oil extracts were effective against all the test organisms (Clinical and Laboratory Standards, Zone > 18 = Active) but significantly lower (p<0.05) than the control antibiotics (26.0±1.00-34.0±0.00 mm). The MIC and the MBC of combined fractions of *Jatropha curcas* oil extract for fraction 1 & 2 and fraction 2 & 3 against all the test organisms were 6.25 % which were not significantly different from the control (P> 0.05). The combinations of fractions of *J. curcas* were more effective than that of *R. communis* against the test organisms. The combination of fractions 1 & 2 for *J. curcas* had significantly higher (P<0.05) effect against *Staphylococcus aureus* (32.0±0.20 mm), *Bacillus subtilis* (28.0±0.00 mm) and *Escherichia coli* (26.0±1.50 mm) than for *Pseudomonas aeruginosa* (21.0±0.50 mm) at 100 mg/ml. The high antibacterial activities shown by the fractions of *Jatropha curcas* and *Ricinus communis* oils against *Escherichia coli, Pseudomonas aeruginosa, Bacillus subtilis* and *Staphylococcus aureus* is an indications that the plants are potential source of standard antibacterial principles that would be cost effective. Therefore, an acceptable and effective dosage of the fractions of *Jatropha curcas* and *Ricinus communis* oils extracts can be prepared for the control and eradication of these pathogens.

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**Keywords:** *Jatropha curcas, Ricinus communis* oils, Combined Fractions, MIC, MBC

**1.0 Introduction**

*Jatropha curcas* and castor plants have been reported to have a lot of health benefits because of their wide range of medicinal uses (Edeoga *et al*., 2014). The name *Jatropha curcas* meaning Doctor’s nutrient, was related to its numerous medicinal uses. The medicinal uses of this species range from external, internal and even teeth (Agbogidi and Ekeke, 2011). Different parts of the plant including the leaves, fruits, latex and bark have been reported to contain glycosides, tannins, phytosterol, flavonoids and steroidal sapogenins that exhibits wide range of medicinal properties (Agbogidi and Eruotor, 2012). Flavonoids are phenolic compounds that are involved in plant-plant interaction (allelopathy, inhibition of germination and growth) while glycosides are synthesized for amino acids. The oil from *J. curcas* seeds is helpful with the management of rashes and parasitic skin diseases (Edeoga *et al*., 2014). From the report of Priminick (2010), when the oil is mixed with benzyl benzoate, it becomes effective against microbial infections such as scabies and dermatitis. The oil from the seed can also be applied to soothe rheumatic pain. *Jatropha* kernel oil together with about 36% linoleic acid is a possible interest for skin care industry (Priminick, 2010). The development and spread of resistance to the commonly used antibiotics by microbial cells has increased efforts in the search for new antibiotics for the treatment of microbial infections and diseases (Priminick, 2010).

*Ricinus communis* are convenient in arthralgia and urodynia while the seeds are handy in ingestion and for making a medicinal cream to treat several diseases. The seeds oil is an exceptionally viable laxative for all diseases (McGuire, 2015). Commercially the oil is used as a lubricating agent formation, food preservatives etc. (Bhagat and Kulkarni, 2010). Therefore, this study was aimed to determine the antibacterial effect of **combined fractions of *Jatropha curcas* and *Ricinus communis* oils extract against selected bacteria** against *Escherechia coli*, *Pseudomonas aeruginosa Bacillus subtilis* and *Staphylococcus aureus*.

**2.0 Materials and Method**

**2.1 Study Area**

This study was carried out at the Microbiology laboratory of the University of Abuja, Gwagwalada Federal Capital Territory, Abuja, Nigeria.

**2.2 Preparation and sterilization of media**

The media used in this study are: Nutrient agar (Himedia M001-500G), MacConkey agar (Himedia M001-500G), Eosin methylene blue agar (Titan), Mannitol salt agar (Himedia), Mueller Hinton agar (Himedia) and peptone water (Himedia) and they were prepared according to the manufacturers’ instructions.

**2.3 Sample collection and identification**

Healthy and mature *Jatropha curcas* and *Ricinus communis* seeds were collected from Gwagwalada FCT-Abuja and taken to the University of Abuja Herbarium for identification. The *Jatropha curcas* and *Ricinus communis* seeds collected were sorted, de-hulled, cleaned and dried (at room temperature) to constant weights and the oils in the kernels were extracted mechanically.

**2.4 Extraction and sterilization of *Jatrophacurcas* and *Ricinuscommunis* oils**

Extraction of oils from the kernels of *Jatropha curcas* and *Ricinus communis* was done according to the method used by Muzenda *et al.* (2012) that involved hot pressing using a hydraulic press. The clean dry kernels were crushed and then placed in the hydraulic press and pressed until they became cake to extract the oils. The resultant solid and colloidal matters were removed by sedimentation and filtered using a filter press. Finally, the oils were sterilized using membrane filtration according to Muzenda *et al.* (2012) and stored in sterile bijoux bottles at 4oC.

**2.5 Test organisms**

The test organisms (*Escherichia coli* (LMG 21766)*, Pseudomonas aeruginosa* (ATCC 9027), *Bacillus subtilis* (ATCC 6633) and *Staphylococcus aureus* (ATCC 6538) were obtained from the Diagnostic Division, NVRI, Vom, Jos. They were resuscitated by streak inoculation on Nutrient agar, incubated at 37 0C for 24 hrs and later on their various selective media and tested for purity by microscopy following Gram staining. They were then preserved on fresh nutrient agar slants in a refrigerator at 4 oC.

**2.6 Identification of test organisms**

The test organisms were identified on the basis of microscopy following Gram Staining and the characteristics used included growth patterns, colonial characteristics, color, shape, arrangement and entire surface of pure isolates which were observed by visual examinations.

Isolates from Nutrient Agar and Eeosin Methylene Blue agar (EMB) with green metallic sheen were subjected to IMViC series of tests. This provided additional evidence for the identification of *Escerichia coli*. It consists of Indole Production, Methyl red test, Voges Proskauer test and the citrate utilization test while *Staphylococcus aureus* was isolated on manitol salt agar (MSA) and then subjected to catalase and coagulase tests while spore staining was carried out to further confirm *Bacillus subtilis*.

**2.7 Standardization of the test organisms**

The test organisms were standardized using standard curves. An inoculum of the slant culture of each test organism, *Escherichia coli* (LMG 21766)*, Pseudomonas aeruginosa* (ATCC 9027), *Bacillus subtilis* (ATCC 6633) and *Staphylococcus aureus* (ATCC 6538) was subcultured unto freshly prepared nutrient agar plates and incubated for 18hrs at 37 0C.Ten - fold serial dilutions of each suspension were made from a discrete colony of each. A loop full of each test organism was separately incorporated in 10 ml of sterile distilled water as the stock culture. Ten - fold serial dilutions of the stock culture were made using sterile water as diluent. Then 1.0 ml of the stock was aseptically pipetted into a sterile test tube containing 9.0 ml of sterile water. The contents were mixed thoroughly. Other ten-fold dilutions of solution were similarly made up to 10-6. One (1) millilitre was taken and discarded from the last tube. Spectrophotometer was standardized using distilled sterile water. From the dilution tubes, samples were taken from every dilution into cuvettes to measure their optical densities and each dilution was plated by the spread plate technique for viable count. Finally, a graph of colony forming unit per ml (cfu/ml) against optical density was plotted to obtain standard curve of the test organisms.

**2.8 Fractionation of oil extracts**

The oils were fractionated using thin Layer chromatography and column chromatography and the fractions were tested for antibacterial activity individually and in combinations.

**2.8.1 Thin Layer chromatography of *Ricinus communis* Oil and *Jatropha curcas* Oil Extracts**

The thin layer chromatography (TLC) was carried out based on the method described by Philip (2013). The TLC profile of the oil extracts was obtained on pre-coated TLC plate using hexane/ethyl acetate and petroleum ether mixture in the ratio of 6:3:1 for *Jatropha curcas* oil and methanol and petroleum ether in the ratio of 4:1 for *Ricinus communis* oil in a beaker and covered to keep the concentration. The TLC plates of sizes 5.0 cm in width and 10cm in length were used. A line was drawn across the width of the plates at 1.0cm mark from the bottom of the plate as a starting mark using a ruler and a pencil. A capillary tube was then used to take the oil extracts on the centre of the line drawn on the TLC plate. The plates were taken into a chromatography chamber and observed till the solvent rose to the 6.1cm mark on the TLC plate which was the solvent front. The TLC plates were carefully removed from the chromatography chamber and observed under a UV lamp at 254/366nm to see the separated components and each of the components were mark with a pencil. Also, the plates were sprayed with a mixture of 90% ethanol and 10% sulphuric acid followed by heating on a hot plate to get the colour and permanent spot on the TLC plate. The resolution front (RF) value was calculated for each of the fractions using the formula:

Rf = Distance travelled by solute ……Equ. (1)

Distance travelled by solvent. (Philip, 2013).

**2.8.2 Column chromatography of *Ricinus communis* oil and *Jatropha curcas* oil extracts**

The flash column was washed, dried and clamped vertically unto a retort stand. A piece of cotton wool was introduced into the clean dry column followed by 30g absorbent silica gel. The column was tapped gently to give a uniform packing. Some 15g of oil extract was weighed and poured into the column followed by the addition of solvent. Solvent elution was started with 100% hexane followed by hexane/ethyl acetate (80:20 %, 60:40 %, 40:60 %, 20:80 % v/v, 100% ethyl acetate and 100% methanol respectively) for both *J. curcas* and *R. communis* oils. The fractions were collected in 50ml beakers and evaporated. The fractions were further purified using thin layer chromatography (TLC) using 100% chloroform for *J.curcas oil* and Hexane-ethyl acetate-methanol (4:4:1) for *R. communis* oil as the solvent system and 10% sulphuric acid (H2SO4) as spray reagent for both oil extracts. This was done according to Philip (2013). The fractions were tested for antibacterial activity individually and in combinations.

**2.9 Statistical analysis**

Data obtained in this study were analyzed statistically using the Statistical package for social sciences (SPSS) for windows version involving parametric test such as ANOVA at P<0.05.

**3.0 Results**

The results are presented as follows:

**3.1 Antibacterial effect of combined fractions of *Jatropha curcas* and *Ricinus communis* oil extracts**

Figure 1 and 2 showed the effect of combinations of column fractions of *Jatropha curcas* and *Ricinus communis* oil against the test organisms. It can be seen that combination of fraction 1&2 of *J. curcas* had significantly higher (P< 0.05) effect against *Staphylococcus aureus* (32±0.20 mm), *Bacillus subtilis* (28.0±0.00 mm) and *Escherichia coli* (26.0±1.50 mm) than *Pseudomonas aeruginosa* (21.0±0.50 mm) at 100 % (Figure 1). From the same Figure 1, combinations of fractions 2&3 and fraction 1&5 were not significantly different at concentration of 100% (P> 0.05) but had significant effect against all test organisms which are *Staphylococcus aureus* (29±1.10 mm and 29±2.00 mm respectively), *Bacillus subtilis* (27±0.00mm and 26±1.00 mm), *Escherichia coli* (24±1.10 mm and 22±0.00 mm respectively) and *Pseudomonas aeruginosa* (19±2.00 mm and 18±0.00 mm respectively).The antibacterial effect of *Jatropha curcas* oil extracts increased with concentrations whereas the antibacterial effects of the controls were not significantly different from the effects of combine fractions1&2 (P> 0.05).

From Figure 2, it can be seen that fraction 1&2 of *R. communis* oil extracts also had significantly higher (P< 0.05) effect against *Staphylococcus aureus* (28.0±0.11 mm) and *Bacillus subtilis* (26.0±0.00 mm) than *Escherichia coli* (22.0±2.00 mm) and *Pseudomonas aeruginosa* (19.5±0.50 mm) at 100 %whereas the antibacterial effect of the control was significantly higher than that of all test organisms. From the same Figure 2, fractions 1&5 show significantly higher effects against *Staphylococcus aureus* (25.0±0.00 mm) and *Bacillus subtilis* (23.0±0.50 mm) than *Escherichia coli* (19.0±0.20 mm) and *Pseudomonas aeruginosa* (17.0±0.00 mm) at P <0.05. The antibacterial effect of the combined fractions of *Jatropha curcas* and *Ricinus communis* oil increased with concentrations. The antibacterial effect of *Jatropha curcas* and *Ricinus communis* oil extracts against the test organisms were significantly different (P< 0.05).

**Figure 1: Antibacterial effect of combined fractions of *Jatropha curcas* oil against test organisms.**

Keys: CH= Chloramphenicol, AMP= Ampicillin, Ps= *Pseudomonas aeruginosa*, Bs= *Bacillus subtilis*,

Sa= *Staphylococcus aureus*, Ec= *Escherichia coli*.

**Figure 2: Antibacterial effects of combined fractions of *R. cummunis* oil against test organisms.**

Keys: CH= Chloramphenicol, AMP= Ampicillin, Ps= *Pseudomonas aeruginosa*, Bs= *Bacillus subtilis*,

Sa= *Staphylococcus aureus*, Ec= *Escherichia coli*.

**3.2 Minimum Inhibitory Concentration of Combined Fractions of *Ricinus communis* Oil Extracts**

The minimum inhibitory concentration of combined fractions of *Ricinus communis* oil extract fractions against *Staphylococcus aureus* (ATCC 6538), *Bacillus subtilis* (ATCC 6633), *Escherichia coli* (LMG 21766) and *Pseudomonas aeruginosa* (ATCC 9027) are as presented on Table 1. The minimum inhibitory concentration of combined fractions of *Ricinus communis* oil for fraction 1&2 against all test organisms was 6.25 %whereas only *Pseudomonas aeruginosa* had MIC of 12.50 %for fraction 2&3 combinations while *Staphylococcus aureus, Bacillus subtilis* and *Escherichia coli* were 6.25 %each which were significantly higher than the control (P< 0.05). Minimum inhibitory concentrations of combined fractions 3&4 of *R. communis* oil against *Pseudomonas aeruginosa* and *Escherichia coli* were 12.50 %each while *Staphylococcus aureus* and *Bacillus subtilis* were 6.25 % each. Fraction 4&5 and 1&5 had minimum inhibitory concentrations of 25% against *P. aeruginosa* while *Staphylococcus aureus B. subtilis* and *E.coli* were 12.50 %which were significantly higher than the control (P< 0.05).

**Table 1: Minimum inhibitory concentrations of combined fractions of *Ricinus communis* oil extracts against test organisms**

|  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- |
| Fractions | Test Organisms | Concentrations in % | | | | | |
| 100 | 50 | 25 | 12.50 | 6.25 | 3.13 |
| Fraction 1&2 | Ps | - | - | - | - | -\* | + |
| Bs | - | - | - | - | -\* | + |
| Sa | - | - | - | - | -\* | + |
| Ec | - | - | - | - | -\* | + |
|  |  |  |  |  |  |  |  |
| Fraction 2&3 | Ps | - | - | - | -\* | + | + |
| Bs | - | - | - | - | -\* | + |
| Sa | - | - | - | - | -\* | + |
| Ec | - | - | - | - | -\* | + |
|  |  |  |  |  |  |  |  |
| Fraction 3&4 | Ps | - | - | - | -\* | + | + |
| Bs | - | - | - | - | -\* | + |
| Sa | - | - | - | - | -\* | + |
| Ec | - | - | - | -\* | + | + |
|  |  |  |  |  |  |  |  |
| Fraction 4&5  Fraction 1&5  Control  CH  AMP  AMP  CH | Ps | - | - | -\* | + | + | + |
| Bs | - | - | - | -\* | + | + |
| Sa | - | - | - | -\* | + | + |
| Ec  Ps  Bs  Sa  Ec  Ps  Bs  Sa  Ec | -  -  -  -  -  -  -  -  - | -  -  -  -  -  -  -  -  - | -  -\*  -  -  -  -  -  -  - | -\*  +  -\*  -\*  -\*  -  -  -  - | +  +  +  +  +  -  -  -  - | +  +  +  +  +  +  +  +  + |

Keys: + = Growth, - = No growth, Fraction 1= 100 % Hexane, Fraction 2 = 80: 20 % Hexane/Ethyl acetate, Fraction 3 = 60: 40 % Hexane/Ethyl acetate, Fraction 4 = 20: 80 % Hexane/Ethyl acetate, Fraction 5= 100% Methanol, Ps= *Pseudomonas aeruginosa*, Bs= *Bacillus subtilis*, Sa= *Staphylococcus aureus*, Ec= *Escherichia coli,* -\* = MIC.

**3.3 Minimum Bactericidal Concentration of Combine Fractions of *Ricinus communis* Oil Extracts**

Table 2 shows the minimum bactericidal concentrations of combined fractions of *Ricinus communis* oil extract against *Staphylococcus aureus* (ATCC 6538), *Bacillus subtilis* (ATCC 6633), *Escherichia coli* (LMG 21766) and *Pseudomonas aeruginosa* (ATCC 9027). The minimum bactericidal concentration of combined fractions of *R. communis* oil extract for fraction 1&2 against *Staphylococcus aureus* and *Bacillus subtilis* were 6.25 %which were not significantly different from that of the control (P> 0.05) whereas that of *Pseudomonas aeruginosa* and *Escherichia coli* were 12.50 %. Only *Pseudomonas aeruginosa* had MBC of 12.50% while *Staphylococcus aureus Bacillus subtilis* and *Escherichia coli* were 6.25 % for fraction 2&3. Fraction 3&4 had minimum bactericidal concentrations of 12.50 % against all the test organisms. Only *Staphylococcus aureus* had MBC of 12.50 %whereas *Bacillus subtilis, Pseudomonas aeruginosa* (ATCC 9027) and *Escherichia coli* (LMG 21766) were 25%each for fraction 4&5 which were significantly higher than other combinations. Fraction 1&5 minimum bactericidal concentrations of *Ricinus communis* oil extract against *Staphylococcus aureus*, *Bacillus subtilis* and *Escherichia coli* were 12.50 %each while *Pseudomonas aeruginosa* was 25 %which was significantly higher than that of the control (P< 0.05).

**Table 2: Minimum bactericidal concentrations of combined fractions of *Ricinus communis* oil extract against test organisms**

|  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- |
| Fractions | Test Organisms | Concentrations in % | | | | | |
| 100 | 50 | 25 | 12.50 | 6.25 | 3.13 |
| Fraction 1&2 | Ps | - | - | - | -\* | + | + |
| Bs | - | - | - | - | -\* | + |
| Sa | - | - | - | - | -\* | + |
| Ec | - | - | - | -\* | + | + |
|  |  |  |  |  |  |  |  |
| Fraction 2&3 | Ps | - | - | - | -\* | + | + |
| Bs | - | - | - | - | -\* | + |
| Sa | - | - | - | - | -\* | + |
| Ec | - | - | - | - | -\* | + |
|  |  |  |  |  |  |  |  |
| Fraction 3&4 | Ps | - | - | - | -\* | + | + |
| Bs | - | - | - | -\* | + | + |
| Sa | - | - | - | -\* | + | + |
| Ec | - | - | - | -\* | + | + |
|  |  |  |  |  |  |  |  |
| Fraction 4&5  Fraction 1&5  Control  CH  AMP  AMP  CH | Ps | - | - | -\* | + | + | + |
| Bs | - | - | -\* | + | + | + |
| Sa | - | - | - | -\* | + | + |
| Ec  Ps  Bs  Sa  Ec  Ps  Bs  Sa  Ec | -  -  -  -  -  -  -  -  - | -  -  -  -  -  -  -  -  - | -\*  -\*  -  -  -  -  -  -  - | +  +  -\*  -\*  -\*  -  -  -  - | +  +  +  +  +  -  -  -  - | +  +  +  +  +  +  +  +  + |

Keys: + = Growth, - = No growth, Fraction 1= 100 % Hexane, Fraction 2 = 80: 20 % Hexane/Ethyl acetate, Fraction 3 = 60: 40 % Hexane/Ethyl acetate, Fraction 4 = 20: 80 % Hexane/Ethyl acetate, Fraction 5= 100% Methanol, Ps= *Pseudomonas aeruginosa*, Bs= *Bacillus subtilis*, Sa= *Staphylococcus aureus*, Ec= *Escherichia coli,* -\* = MBC.

**4.4 Minimum Inhibitory Concentration of Combined Fraction of *Jatropha curcas* Oil Extracts**

Table 3 shows the minimum inhibitory concentrations of combined fraction of *Jatropha curcas* oil extract fractions against *Staphylococcus aureus* (ATCC 6538), *Bacillus subtilis* (ATCC 6633), *Escherichia coli* (LMG 21766) and *Pseudomonas aeruginosa* (ATCC 9027). The minimum inhibitory concentration of combined fractions of *Jatropha curcas* oil extract for fraction 1&2 and fraction 2&3 against all the test organisms were 6.25 %which were not significantly different from the control (P> 0.05) whereas only *Pseudomonas aeruginosa* was 12.50 %for fractions 3&4 combination which was significantly higher than the control (P< 0.05), while *Staphylococcus aureus, Bacillus subtilis* and *Escherichia coli* were 6.25 %each which were not significantly different from the control (P< 0.05). From the same Table 3, fraction 4&5 had MIC of 12.50 %against all the test organisms. For fraction 1&5, the MIC for *Staphylococcus aureus* and *Bacillus subtilis* were 6.25 %which were not significantly different from the control (P> 0.05), while *Pseudomonas aeruginosa* and *Escherichia coli* were 12.50% each.

**Table 3: Minimum inhibitory concentrations of combined fractions of *Jatropha curcas* oil extractsagainst test organisms**

|  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- |
| Fractions | Test Organisms | Concentrations in % | | | | | |
|  | | | | | |
| 100 | 50 | 25 | 12.50 | 6.25 | 3.13 |
| Fraction 1&2 | Ps | - | - | - | - | -\* | + |
| Bs | - | - | - | - | -\* | + |
| Sa | - | - | - | - | -\* | + |
| Ec | - | - | - | - | -\* | + |
|  |  |  |  |  |  |  |  |
| Fraction 2&3 | Ps | - | - | - | - | -\* | + |
| Bs | - | - | - | - | -\* | + |
| Sa | - | - | - | - | -\* | + |
| Ec | - | - | - | - | -\* | + |
|  |  |  |  |  |  |  |  |
| Fraction 3&4 | Ps | - | - | - | -\* | + | + |
| Bs | - | - | - | - | -\* | + |
| Sa | - | - | - | - | -\* | + |
| Ec | - | - | - | - | -\* | + |
|  |  |  |  |  |  |  |  |
| Fraction 4&5  Fraction 1&5  Control  CH  AMP  AMP  CH | Ps | - | - | - | -\* | + | + |
| Bs | - | - | - | -\* | + | + |
| Sa | - | - | - | -\* | + | + |
| Ec  Ps  Bs  Sa  Ec  Ps  Bs  Sa  Ec | -  -  -  -  -  -  -  -  - | -  -  -  -  -  -  -  -  - | -  -  -  -  -  -  -  -  - | -\*  -\*  -  -  -\*  -  -  -  - | +  +  -\*  -\*  +  -  -  -  - | +  +  +  +  +  +  +  +  + |

Keys: + = Growth, - = No growth, Fraction 1= 100 % Hexane, Fraction 2 = 80: 20 % Hexane/Ethyl acetate, Fraction 3 = 60: 40 % Hexane/Ethyl acetate, Fraction 4 = 20: 80 % Hexane/Ethyl acetate, Fraction 5= 100% Methanol, Ps= *Pseudomonas aeruginosa*, Bs= *Bacillus subtilis*, Sa= *Staphylococcus aureus*, Ec= *Escherichia coli*, -\* = MIC.

**4.5 Minimum Bactericidal Concentration of Combined Fractions of *Jatropha curcas* Oil Extracts**

Tables 4 shows the minimum bactericidal concentrations of combined fraction of *Jatropha curcas* oil extract fractions against *Staphylococcus aureus* (ATCC 6538), *Bacillus subtilis* (ATCC 6633), *Escherichia coli* (LMG 21766) and *Pseudomonas aeruginosa* (ATCC 9027). The minimum bactericidal concentration of combined fraction of *Jatropha curcas* oil extract for fraction 1&2 and fraction 2&3 against all the test organisms were 6.25 %which were not significantly different from the control (P> 0.05) whereas *Staphylococcus aureus* and *Bacillus subtilis* were 6.25 %for fractions 3&4 combination, while *Pseudomonas aeruginosa* and *Escherichia coli* were 12.50 %each. For fraction 4&5, the MBC for *Staphylococcus aureus* and *Bacillus subtilis* were 12.50 %, while *Pseudomonas aeruginosa* and *Escherichia coli* were 25 % each. For fraction 1&5, *Escherichia coli* and *Pseudomonas aeruginosa* were 12.50 %which were significantly higher (P<0.05) than the control whereas *Staphylococcus aureus* and *Bacillus subtilis* were 6.25 %which were not significantly different from the control (P> 0.05).

**Table 4: Minimum bactericidal concentrations of combine fractions of *Jatropha curcas* oil extracts against test organisms**

|  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- |
| Fractions | Test Organisms | Concentrations in % | | | | | |
|  | | | | | |
| 100 | 50 | 25 | 12.50 | 6.25 | 3.13 |
| Fraction 1&2 | Ps | - | - | - | - | -\* | + |
| Bs | - | - | - | - | -\* | + |
| Sa | - | - | - | - | -\* | + |
| Ec | - | - | - | - | -\* | + |
|  |  |  |  |  |  |  |  |
| Fraction 2&3 | Ps | - | - | - | - | -\* | + |
| Bs | - | - | - | - | -\* | + |
| Sa | - | - | - | - | -\* | + |
| Ec | - | - | - | - | -\* | + |
|  |  |  |  |  |  |  |  |
| Fraction 3&4 | Ps | - | - | - | -\* | + | + |
| Bs | - | - | - | - | -\* | + |
| Sa | - | - | - | - | -\* | + |
| Ec | - | - | - | -\* | + | + |
|  |  |  |  |  |  |  |  |
| Fraction 4&5  Fraction 1&5  Control  CH  AMP  AMP  CH | Ps | - | - | -\* | + | + | + |
| Bs | - | - | - | -\* | + | + |
| Sa | - | - | - | -\* | + | + |
| Ec  Ps  Bs  Sa  Ec  Ps  Bs  Sa  Ec | -  -  -  -  -  -  -  -  - | -  -  -  -  -  -  -  -  - | -\*  -  -  -  -  -  -  -  - | +  -\*  -  -  -\*  -  -  -  - | +  +  -\*  -\*  +  -  -  -  - | +  +  +  +  +  +  +  +  + |

Keys: + = Growth, - = No growth, Fraction 1= 100 % Hexane, Fraction 2 = 80: 20 % Hexane/Ethyl acetate, Fraction 3 = 60: 40 % Hexane/Ethyl acetate, Fraction 4 = 20: 80 % Hexane/Ethyl acetate, Fraction 5= 100% Methanol, Ps= *Pseudomonas aeruginosa*, Bs= *Bacillus subtilis*, Sa= *Staphylococcus aureus*, Ec= *Escherichia coli,* -\* = MBC.

**4.0 Discussions**

The activity of the study plant oil extract against both Gram-positive and Gram-negative bacteria can be indicative of the presence of broad spectrum antibiotic compounds or simply general metabolic toxins in the plant. The activities against different bacteria were varied with the concentration of extracts tested. High concentrations inhibited the growth of the bacterial species *Bacillus subtilis, Staphylococcus aureus,* and *Escherichia coli.* However, the inhibitory activity was reduced with the concominent decrease of extract concentration (Figure 1 and 2). The antibacterial effect of *Jatropha curcas* oil extracts increased with concentrations but, the antibacterial activities of *Jatropha curcas* (21.0±0.50 mm) and *Ricinus communis* oil extracts mean zones (19.5±0.50 mm) of inhibition against *Pseudomonas aeruginosa* (ATCC 9027) were not significant (P < 0.05). Verma *et al.* (2011) who reported similar result on antimicrobial potential of roots of *Ricinus communis* against pathogenic *Pseudomonas aeruginosa* and *Escherichia coli*.

Among the four microorganisms tested, two bacterial species, *Staphylococcus aureus* and *Bacillus subtilis* were quite sensitive to high concentrations of both *Jatropha curcas* (32±0.20 mm) and *Ricinus communis* (28.0±0.11 mm). Both *Jatropha curcas* and *Ricinus communis* had antimicrobial effects on all the selected microorganisms with less effect on *Pseudomonas aeruginosa,* but *Jatropha curcas* had higher antimicrobial activity than the *Ricinus communis*. The antibacterial effect of the combined fractions of *Jatropha curcas* and *Ricinus communis* oil increased with concentrations. The antibacterial effect of *Jatropha curcas* and *Ricinus communis* oil extracts against the test organisms were significantly different (P< 0.05).

The antimicrobial effects of both *Jatropha curcas* and *Ricinus communis* against all the test organisms also increased with concentration. This showed that inhibitory activity of plant extracts generally depends upon the concentration, type of parts used and microbes tested. This is probably because of the accumulation and concentration of secondary metabolites which are responsible for inhibitory activity and varied accordingly with the plant parts. This may be a reason for the variation in the inhibitory activities of the extracts of both *Jatropha curcas* and *Ricinus communis*. This is in agreement with the report by Odungbemi (2014) that *Ricinus communis* is recognized as a traditional solution for gastropathy i.e. amadosa, constipation, irritations, ascitis, strangury, fever, bronchitis, chest infection, skin maladies, coxalgia, colic, and lumbago. The low minimum inhibitory concentration exhibited by the oil extracts on *S. aureus* is of great significance in the health care delivery system since it could be used as an alternative to orthodox antibiotics in the management of infections due to the microorganisms, especially as they frequently develop resistance to known antibiotics. It was also observed from this work that the higher the concentration the more their activity and as the concentration decreases the lower the antimicrobial effect. Hence an acceptable and effective dosage can be prepared for the control and eradication of these pathogens.

**4.1 Conclusion**

It can be concluded that the high antibacterial activities shown by the fractions of *Jatropha curcas* and *Ricinus communis* oils against *Escherichia coli, Pseudomonas aeruginosa, Bacillus subtilis* and *Staphylococcus aureus* is an indications that the plants are potential source of standard antibacterial principles that would be cost effective.

**References**

1. Agbogidi, O.M. and Ekeke, E.A. (2011). *Jatropha curcas*: Linn an important but neglected plant species in Nigeria. *Journal of Biological and Chemical Research,* 281: 52-62.
2. Bhagat, R.B., Kulkarni, D.K. (2010). Phytochemical, antioxidant and antimicrobial anlysis of endemic and endangered *Jatropha nana* Dalz. and Gibs from Maharashtra. *Journal of Pharmaceutical Research*, 3(9):273-2076.
3. Edeoga, H.O., Okwu, D.E. and Mbaebre, B.O. (2014). Phytochemical constituents of some Nigerian plants. *African Journal of Biotechnology,* 44 (7): 685-688.
4. McGuire, N. (2015). Taming the Bean. *The American Journal of Chemical Society,*1 (4): 122-124.
5. Muzenda, E., Kabuba, J., Mdletye, P. and Belaid, M. (2012). Optimization of Process Parameters for Castor Oil Production. *Proceedings of the World Congress on Engineering,* Vol. III, London, U.K. Pp. 65.
6. Philip, M. (2013). Advanced Chemistry Physical and industrial, South Asia. Cambridge University Press. Pp.330–331.
7. Prminik, K. (2010). Properties and use of *Jatropha curcas* oil and diesel fuel blends compression ignition engines. *Journal of Renewable Energy* 28: 239-248.
8. Verma, S.K., Yousuf, S., Singh, S.K., Parsad, G.B.K.S. and Dua, V.K. (2011). Antimicrobial potential of roots of *Ricinus communis* against pathogenic microorganisms. *International Journal of Pharmacology and Biological Science*, 2(1): 545-8.
9. Odungbemi, T. (2014). Outline and pictures of medicinal plants from Nigeria, University of Lagos press, Yaba Lagos, Nigeria. Pp. 283.

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