### 2.2 Preparation of Extracts

The collected *Shorea robusta* leaves were washed several times with distilled water to remove the traces of impurities from the leaves. The leaves were dried at room temperature and coarsely powdered. The powder was extracted with 70% ethanol for 48 hours. A semi solid extract was obtained after complete elimination of alcohol under reduced pressure. The extract was stored in desiccator until used. The extract contained both polar and non-polar phytocomponents of the plant material used.

### 2.3 GC –MS Analysis

GC-MS analysis was carried out on a GC clarus 500 Perkin Elmer system comprising a AOC-20i autosampler and gas chromatograph interfaced to a mass spectrometer instrument employing the following conditions: column Elite-1 fused silica capillary column (30 x 0.25mm ID x 1µMdf, composed of 100% Dimethyl polydiloxane), operating in electron impact mode at 70eV; Helium gas (99.999%) was used as carrier gas at a constant flow of 1 ml /min and an injection volume of 0.5 µI was employed (split ratio of 10:1) injector temperature 250 ºC; ion-source temperature 280 ºC. The oven temperature was programmed from 110 ºC (isothermal for 2 min), with an increase of 10 ºC/min, to 200ºC, then 5ºC/min to 280ºC, ending with a 9min isothermal at 280ºC. Mass spectra were taken at 70eV; a scan interval of 0.5 seconds and fragments from 40 to 450 Da. Total GC running time is 36min. min. The relative percentage amount of each component was calculated by comparing its average peak area to the total areas. Software adopted to handle mass spectra and chromatograms was a TurboMass Ver 5.2.0

## 3. Results and Discussion

Plants have an almost limitless ability to synthesize aromatic substances, most of which are phenols or their oxygen substituted derivatives. Most are secondary metabolites, of which at least 12,000 have been isolated, a number estimated to be less than 10% of the total. These substances serve as plant defense mechanisms against, insects and herbivores. Flavonoids exhibit several biological effects such as anti-inflammatory, anti-fungal, anti-hepatotoxic and anti-ulcer actions [14].

### 3.1 Identification of Components

Interpretation on mass spectrum GC-MS was conducted using the database of National Institute Standard and Technology (NIST) having more than 62,000 patterns. The spectrum of the unknown component was compared with the spectrum of the known components stored in the NIST library. The name, molecular weight and structure of the components of the test materials were ascertained. The biological activities listed (Table 2) are based on Dr.Duke's Phytochemical and Ethnobotanical Databases by Dr. Jim Duke of the Agricultural Research Service/USDA.

### 3.2 GC-MS Analysis

Thirty compounds were identified in *Shorea robusta* by GC-MS analysis. The active principles with their retention time (RT), molecular formula, molecular weight (MW) and concentration (%) are presented in (Table 1 and Fig 1). The prevailing compounds were 9,12-Octadecadienoic acid (Z,Z)-, Tetra decanoic acid $$ myristic acid, Nonanedioic acid , dibutyl ester $$ Azelaic acid, 1-(+)-Ascorbic acid 2,6-dihexadecanoate, Heptadecanoic acid, Phytol isomer, Oleic acid $$ 9-octadecenoic acid (Z), Octadecanoic acid $$ stearic acid, Cis-11,14-Eicosadienoic acid, methyl ester and Hexadecanoic acid.

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The investigation concluded that the stronger extraction capacity of methanol could have been produced number of active constituents responsible for many biological activities. So that those might be utilized for the development of traditional medicines and further investigation needs to elute novel active compounds from the medicinal plants which may be created a new way to treat many incurable diseases.

## 4. Acknowledgement

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**Table 1:** Shows the components identified in methanolic extract of *Shorea robusta* (GC MS study)

| Peak | R.TIME | Area% | Name of the Compound | Molecular Formula | Molecular Weight |
|---|---|---|---|---|---|
| 1 | 6.984 | 0.71 | 1-Dimethyl(octyl)silyloxypropane | $C_{13}H_3OOS$ | 230 |
| 2 | 6.984 | 4.04 | 1,2,3-PROPANETRIO | $C_3H_8O_3$ | 92 |
| 3 | 16.358 | 0.62 | Silane, dimethyldi(but-2-enyloxy)- | $C_6H_{16}O_2Si$ | 148 |
| 4 | 16.819 | 0.41 | Dodecanoic acid | $C_{12}H_{24}O_2$ | 200 |
| 5 | 17.535 | 0.16 | Diethyl Phthalate | $C_{12}H_{14}O_4$ | 222 |
| 6 | 18.965 | 0.24 | 2-Propenal, 3-(2,4,5,6,7,7A-Hexahydr | $C_3H_6O_3$ | 90 |
| 7 | 19.650 | 1.07 | Tetradecanoic acid $$ Myristic acid | $C_{24}H_{48}O_2$ | 368 |
| 8 | 20.620 | 0.95 | 2,6,10-Trimethyl,14-ethylene-14-PE | $C_{20}H_{38}$ | 278 |
| 9 | 20.722 | 0.28 | 2-Pentadecanon, 6,10,14-Trimethyl | $C_{18}H_{36}O$ | 268 |
| 10 | 20.910 | 0.53 | 3,7,11,15-Tetramethyl-2-hexadecen-1-ol - | $C_{20}H_{40}O$ | 296 |
| 11 | 21.135 | 0.42 | 3,7,11,15-Tetramethyl-2-hexadecen-1-ol | $C_{20}H_{40}O$ | 296 |
| 12 | 21.848 | 1.17 | Oleic Acid $$ 9-Octadecenoic acid (Z) | $C_{18}H_{34}O_2$ | 282 |
| 13 | 21.892 | 0.76 | Nonanedioic acid, dibutyl ester | $C_{17}H_{32}O_4$ | 300 |
| 14 | 22.032 | 18.38 | l-(+)-Ascorbic acid 2,6-dihexadecanoate | $C_{38}H_{68}O_8$ | 562 |
| 15 | 23.095 | 1.11 | Heptadecanoic acid $$ Potassium | $C_{17}H_{34}O_2$ | 270 |


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| | | | | | |
|---|---|---|---|---|---|
| 16 | 23.724 | 1.80 | Phytol isomer | $C_{20}H_{40}O$ | 296 |
| 17 | 23.808 | 0.22 | Octadecanoic acid, methyl ester | $C_{19}H_{38}O_2$ | 298 |
| 18 | 24.00 | 25.84 | Oleic Acid | $C_{18}H_{34}O_2$ | 282 |
| 19 | 24.242 | 25.86 | Octadecanoic acid $$ Stearic acid | $C_{18}H_{34}O_2$ | 282 |
| 20 | 24.542 | 3.06 | 9,12-Octadecadienoic acid (Z,Z)- | $C_{18}H_{32}O_2$ | 280 |
| 21 | 25.047 | 0.56 | 9,12-Octadecadienoic acid | $C_{21}H_{42}O_4$ | 358 |
| 22 | 25.556 | 0.77 | Nonadecanoic ACID | $C_{19}H_{36}O_2$ | 296 |
| 23 | 26.125 | 0.46 | Hexadecanoic acid, 2-hydroxy-1,3 | $C_{37}H_{74}NO_8P$ | 691 |
| 24 | 26.817 | 0.80 | cis-13-Eicosenoic acid | $C_{15}H_{26}O$ | 222 |
| 25 | 26.916 | 1.16 | 9-Octadecenoic acid, 1,2,3-propanetriyl ester, (E | $C_{18}H_{36}O_2$ | 284 |
| 26 | 27.165 | 4.71 | Icosanoic acid $$ Arachinsaeure | $C_{20}H_{40}O_2$ | 321 |
| 27 | 29.575 | 1.27 | 2-Hydroxy-3-[(9E)-9-Octadecenoylo | $C_{39}H_{72}O_5$ | 620 |
| 28 | 29.665 | 1.26 | Solanesol | $C_{45}H_{74}O$ | 360 |
| 29 | 30.091 | 1.06 | Glycidol stearate | $C_{21}H_{40}O_3$ | 340 |
| 30 | 31.495 | 0.31 | Bis(2-ethylhexyl) phthalate | $C_{24}H_{38}O_4$ | 390 |

**Table 2:** Activity of phyto-components identified in the methanolic extracts of the *Shorea robusta* by GC-MS

| S. No | Compound name | Biological activity** |
|---|---|---|
| 1. | 9,12-Octadecadienoic acid (Z,Z)- | Antiinflammatory, hypocholesterolemic, cancer preventive, hepatoprotective, nematicide, insectifuge, antihistaminic antieczemic, antiacne, 5-Alpha reductase inhibitor, antiandrogenic, antiarthritic, anticoronary, insectifuge |
| 2. | Tetra decanoic acid $$ myristic acid | Anti oxidant, cancer preventive, hypercholesterolemic, nematicide, lubricant, cosmetic. |
| 3. | Nonanedioic acid , dibutyl ester $$ Azelaic acid | Anti microbial, anti inflammatory, anti tumor, anti hyperpigmentative, anti proliferative, anti acne, cyto toxic,<br>Anti leukemic, oxy radical scavenging activity. |
| 4. | 1-(+)-Ascorbic acid 2,6-dihexadecanoate | Anti oxidant, anti scorbutic, anti inflammatory, anti nociceptive, anti mutagenic, wound healing property. |
| 5. | Heptadecanoic acid | Antioxidant, anti fungal, surfactant |
| 6. | Phytol isomer | Anti inflammatory, anti cancer, anti microbial, diuretic. |
| 7. | Oleic acid $$ 9-octadecenoic acid (Z) | 5-α reductase inhibitor, allergenic, α-reductase inhibitor, anti inflammatory, anti androgenic, cancer preventive, anemiagenic, anti alopecic, anti leukotriene-D4, choleretic, dermatitigenic, hypocholestrolemic, insectifuge, perfumery, propecic, flavour. |
| 8. | Octadecanoic acid $$ stearic acid | 5-α reductase inhibitor, hypo cholesterolemic, suppository, cosmetic, lubricant, surfactant & softening agent, perfumery, propecic, flavour. |
| 9. | Cis-11,14-Eicosadienoic acid, methyl ester | Anti inflammatory, anti oxidant, anti arthritic, anti coronary. |
| 10. | Hexadecanoic acid | Anti oxidant, hypocholesterolemic, nematicide, pesticide, lubricant, anti androgenic, flavour, hemolytic-5-α reductase inhibitor. |

**Source: Dr.Duke's phytochemical and ethnobotanical databases [Online database].

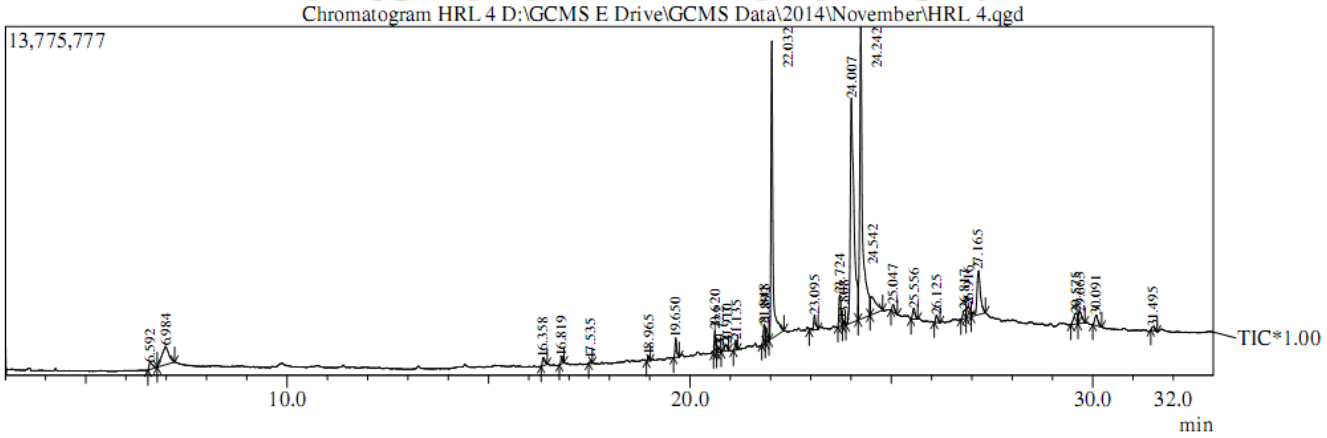


**Figure 1:** Chromatogram obtained from the GC/MS with the extract of *Shorea robusta.*

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