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# Oxidation of essential oil of *Chloroxylon swietenia* (Roxb. corom)

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## Abstract

*Hydro distilled oil from the leaves of C. swietenia is of unpleasant odour and cannot be marketed but it has medicinal importance. If oxygen content present in this, is increased, then not only its odour will turn into pleasant fragrance but also its quality will be improved for medicinal purposes. With this intention it was oxidized using $HNO_3$, $KMnO_4$, $K_2Cr_2O_7$ and $H_2O_2$ as oxidizing agent. $H_2O_2$ was found best suited for oxidation. Antimicrobial activity of oxidized essential oil was found better as compared to un oxidized essential oil which proved improvement in its quality after oxidation.*

**Keywords**: Hydro distillation, essential oil, antimicrobial.

## Introduction

*Chloroxylon swietenia* (Roxb**.** corom) belonging to Rutaceae family is commonly known as Bherul, Bhirra, Ghirya in Hindi and Satinwood in English. It is 9-15 meter high monotypic genus of timber yielding tree found in India and Ceylon. It is widely found in dry deciduous forests in India at an altitude from 1000 to 5000 meter. This plant has medicinal uses.

**Figure-1**

**The important studies on the essential oil of this plant have been made[1] but there had been no oxidation study done so far[2]**

The essential oil from the leaves of *C. sweitenia* does not have pleasing odour and cannot be marketed[3]. In the present work attempt is made to improve the odour and the quality of the essential oil by oxidation. Essential oil contain oxygenating molecules which transport the nutrients to the cells of the body. Clinical research shows that essential oils may help to create an environment in which harmful bacteria, virus, fungi etc. cannot survive. Diseases cannot exist in an oxygen rich environment[4]. If oxygenated components in the essential oil are increased, its fragrance will improve and it will not only be more useful for medicines but also be more useful for industrial and cosmetic uses.

## Material and Methods

The fresh leaves of *C. sweitenia* were collected from Pachmari forest in Hoshangabad district of Madhya Pradesh during September to December.

The essential oil (0 .91% v/w) obtained from the hydrodistillation of shade dried leaves was analysed for physico-chemical properties. Using various methods. after physicochemical analysis and identifying components essential oil was oxidised.

Various oxidants viz. potassium permanganate ($KMnO_4$), potassium dichromate ($K_2Cr_2O_7$), nitic acid ($HNO_3$) and hydrogen peroxide ($H_2O_2$) were tried for oxidising the essential oil obtained from the C. *Swietenia*. The experimental procedure adopted for the study is given below.

**Experimental:** 1.0 ml.of essential oil was taken in a conical flask ( 20ml capacity) and to it 1.00ml of 30 volume hydrogen peroxide was added. The reaction mixture was kept at room temperature ( 28$^o$C) for 24 hours. The reaction was studied at different timing i.e. 6 hours, 12 hours, 18 hours and 24 hours.

The same procedure was adopted at two different temperatures i.e. 50$^o$C and 80$^o$C. The observations are recorded in table 1, table 2 and table 3.

A similar procedure was adopted to study the oxidation of the essential oil using potassium permanganate, potassium dichromate and nitric acid at three temperatures i.e 28$^o$C, 50$^o$C and 80$^o$C. The quantity of each oxidant was taken as 0.01 mol/100ml. volume. The observations are recorded in table 4-12.

**Table-1**
**Oxidant: Hydrogen peroxide ($H_2O_2$). Temperature: 28$^o$C**

| S.N. | Time inHours | Colour of Essential Oil | Odour of Essential Oil |
|---|---|---|---|
| 1. | 6 | Yellow | No change in fragrance |
| 2. | 12 | Yellow | Change in fragrance |
| 3. | 18 | Faint Yellow | Fragrance increased |
| 4 | 24 | Faint Yellow | No change in fragrance |

**Table-2**
**Oxidant: Hydrogen peroxide ($H_2O_2$). Temperature: 50$^o$C**

| S.N. | Time in Hours | Colour of Essential Oil | Odour of Essential Oil |
|---|---|---|---|
| 1. | 6 | Yellow | Agreeable change in fragrance |
| 2. | 12 | Faint Yellow | Fragrance increased significantly |
| 3. | 18 | Faint Yellow | No change in fragrance |
| 4. | 24 | Faint Yellow | No change in fragrance |

**Table-3**
**Oxidant: Hydrogen peroxide ($H_2O_2$). Temperature: 80$^o$C**

| S.N. | Time in Hours | Colour of Essential Oil | Odour of Essential Oil |
|---|---|---|---|
| 1. | 6 | Faint Yellow | Agreeable change in fragrance |
| 2. | 12 | Faint Yellow | Fragrance increased significantly |
| 3. | 18 | Faint Yellow | No change in fragrance |
| 4. | 24 | Faint Yellow | No change in fragrance |

**Table-4**
**Oxidant: Potassium permanganate ($KMnO_4$). Temperature: 28$^o$C**

| S.N. | Time in Hours | Colour of Essential Oil | Odour of Essential Oil |
|---|---|---|---|
| 1. | 6 | Pink | Fragrance increased ( not agreeable ) |
| 2. | 12 | Pink | No change in fragrance |
| 3. | 18 | Pink | No change in fragrance |
| 4. | 24 | Pink | No change in fragrance |

**Table-5**
**Oxidant: Potassium permanganate ($KMnO_4$). Temperature 50$^oC$**

| S.N. | Time in Hours | Colour of Essential Oil | Odour of Essential Oil |
|---|---|---|---|
| 1. | 6 | Pink | No significant change |
| 2. | 12 | Pink | No significant change |
| 3. | 18 | Faint pink | No significant change |
| 4. | 24 | Faint pink | No significant change |

**Table-6**
**Oxidant: Potassium permanganate ($KMnO_4$). Temperature: 80$^oC$**

| S.N. | Time in Hours | Colour of Essential Oil | Odour of Essential Oil |
|---|---|---|---|
| 1. | 6 | Pink | No change in fragrance |
| 2. | 12 | Faint pink | No change in fragrance |
| 3. | 18 | Faint pink | No change in fragrance |
| 4. | 24 | Faint pink | No change in fragrance |

**Table-7**
**Oxidant: Potassium dichromate ($K_2Cr_2O_7$). Temperature: 28$^oC$**

| S.N. | Time in Hours | Colour of Essential Oil | Odour of Essential Oil |
|---|---|---|---|
| 1. | 6 | Yellow | No Change in fragrance |
| 2. | 12 | Yellow | **C**hange in fragrance |
| 3. | 18 | Yellow | **C**hange in fragrance |
| 4. | 24 | Faint yellow | No change in fragrance |

**Table-8**
**Oxidant: Potassium dichromate ($K_2Cr_2O_7$). Temperature: 50$^oC$**

| S.N. | Time in Hours | Colour of Essential Oil | Odour of Essential Oil |
|---|---|---|---|
| 1. | 6 | Yellow | No change in fragrance |
| 2. | 12 | Faint green | Slight change in fragrance |
| 3. | 18 | Faint green | No change in fragrance |
| 4. | 24 | Faint green | No change in fragrance |

**Table-9**
**Oxidant: Potassium dichromate ($K_2Cr_2O_7$). Temperature: 80$^oC$**

| S.N. | Time in Hours | Colour of Essential Oil | Odour of Essential Oil |
|---|---|---|---|
| 1. | 6 | Faint green | Not significant |
| 2. | 12 | Faint green | Not significant |
| 3. | 18 | Faint green | No change in fragrance |
| 4. | 24 | No change In colour | No change in fragrance |

**Table-10**
**Oxidant: Nitric Acid ($HNO_3$). Temperature: 28$^oC$**

| S.N. | Time in Hours | Colour of Essential Oil | Odour of Essential Oil |
|---|---|---|---|
| 1. | 6 | Pale yellow | No change in fragrance |
| 2. | 12 | Pale yellow | No change in fragrance |
| 3. | 18 | Pale yellow | No change in fragrance |
| 4. | 24 | Pale yellow | No change in fragrance |

**Table-11**
**Oxidant: Nitric Acid ($HNO_3$). Temperature: 50$^oC$**

| S.N. | Time in Hours | Colour of Essential Oil | Odour of Essential Oil |
|---|---|---|---|
| 1. | 6 | Pale yellow | No change in fragrance |
| 2. | 12 | Pale yellow | No significant fragrance |
| 3. | 18 | Pale yellow | No significant fragrance |
| 4. | 24 | Pale yellow | No change in fragrance |

**Table-12**
**Oxidant: Nitric Acid ($HNO_3$). Temperature: 80°C**

| S.N. | Time in Hours | Colour of Essential Oil | Odour of Essential Oil |
|---|---|---|---|
| 1. | 6 | Pale yellow | No significant fragrance |
| 2. | 12 | Pale yellow | No significant fragrance |
| 3. | 18 | Pale yellow | No significant fragrance |
| 4. | 24 | Pale yellow | No significant fragrance |

**Determination of amount of hydrogen peroxide $H_2O_2$ used for the oxidation of 1.0 ml of essential oil of chloroxylon swietenia**

**Experimental:** 1.0 ml of essential oil of *C. swietenia* was taken in to a 50ml conical flask and 5.0ml volume hydrogen peroxide ($H_2O_2$) was to added it. This mixture was kept at 50°C temperature. After 12 hours 10ml. of potassium iodide (KI, 10%) solution were added to the mixture. The mixture was titrated against, standard hypo ($Na_2S_2O_3$) (N/20) solution using starch indicator. Same procedure was repeated with 5.0 ml of 30 volume $H_2O_2$ without essential oil (blank titration).

**Table-13**
**Observation Table of Volume of Hypo used for the reaction with Hydrogen peroxide (Without Essential oil) [Blank]**

| Volume of Hydrogen peroxide | Volume of Hypo | | |
|---|---|---|---|
| | Initial Volume (a) | Final Volume (b) | Volume of Hypo used ( b-a) |
| 5.0 ml. | 0.0 ml. | 11.3. ml. | 11.3. ml. |
| 5.0 ml. | 0.0 ml. | 11.2. ml. | 11.2. ml. |
| 5.0 ml. | 0.0 ml. | 11.2 ml | 11.2 ml |

Volume of Hypo used for the titration with 5.0 ml. Of $H_2O_2$ (Blank) = 11.2 ml.

**Table-14**
**Observation Table of Volume of Hypo used for the reaction with Hydrogen peroxide (With 1 ml Essential oil)**

| S.N. | Volume of Hydrogen Peroxide | Volume of Hypo | | |
|---|---|---|---|---|
| | | Initial Volume (a) | Final Volume (b) | Volume of Hypo used ( b-a) |
| 1. | 5.0 ml. | 0.0 ml. | 9.7 ml. | 9.7. ml. |
| 2. | 5.0 ml. | 0.0 ml. | 9.5 ml. | 9.5. ml. |
| 3. | 5.0 ml. | 0.0 ml. | 9.5 ml | 9.5 ml |

Volume of Hypo used for the titration with 5.0 ml. Of $H_2O_2$ (with 1.0 ml. Essential oil) = 9.5 ml.

On calculating the volume and amount of hydrogen peroxide used for the oxidation 1.0.ml. of essential oil of *C. swietenia* were found to be 0.76 ml. **a**nd 0.003 gm respectively from the observation TABLE 13 and 14.

The unoxidised and oxidised essential oils were analysed for physico chemical constants and constituent**s** present in them. To analyse components present in the oxidised essential oil, Thin layer Chromatography, Column Chromatography, Gas liquid Chromatography, Gas Chromatography, Mass-Spectroscopy and Infra Red Spectroscopy techniques were used. Components present in both the essential oils were compared, which is tabulated as under in table 15.

**Table-15**
**Components of Unoxidised and Oxidised Essential Oils: A Comparision**

| Component | Unoxidised essential oil (Concentration of component) % | Oxidised essential oil (Concentration of component) % |
|---|---|---|
| α- pinene | 0.11 | 0.40 |
| Camphene | 0.78 | 0.42 |
| Limonene | 2.78 | 28.95 |
| β-Pinene | 0.08 | **Nil** |
| $\Delta^3$ Carene | 3.17 | 1.94 |
| Myrcene | 0.83 | **Nil** |
| β-Phellandrene | 0.10 | 1.76 |
| P-Cymeme | 0.93 | **Nil** |
| α-Terpinene | 9.29 | 4.30 |
| α-Terpineol | 12.5 | 4.34 |
| Methyl heptenone | 12.29 | **Nil** |
| Citral-a | 4.05 | **Nil** |
| Citral-b | 2.22 | **Nil** |
| Geraniol | 1.05 | 1.97 |
| Linalool | 1.75 | 0.42 |
| β -Caryophyllene oxide | 18.40 | 47.73 |
| Nerol | 6.54 | **Nil** |
| Geranyl acetate | 3.34 | 0.22 |
| β - Caryophyllene | 5.40 | **Nil** |
| α- Caryophyllene | 2.83 | **Nil** |
| Methyl cinnamate | 3.22 | 0.07 |
| α-cadinene | 6.20 | **Nil** |

Three new components, Copaene, Hexahydrodimethyl naphthalene and Cyclobuta 1,2,3,4, dicyclopentene were reported by GC-MS analysis of oxidised oil of *C. swietenia.*

The change in components and their percentages of concentration were due to possibility of isomerisation, re-arrangement**,** elimination, addition and substitution reactions.

Oxidised essential oil was screened for its antimicrobial activity against four gram positive bacteria and sixteen gram-negative bacteria using paper disc agar diffusion method[5] and against thirteen fungi.

In the light of above observations it may be suggested that oxidised essential oil of *C. swietenia* may be used as antifungal and antibacterial agent against some bacteria viz. *Shigella shiga, Sh. flexneri. Vibrio cholerae Ogawa, Bacillus mycoides, B. pumilus* and *Vibrio cholerae Ianwa.* The activity of some fungi viz. *Aspergillus oryzae, A. terreus, Curvularia prasadii, Candida albicans* and *Trichoderma viride* also can be suppressed using this if the in vitro studies hold good under in vivo conditions.

## Results and Discussion

The essential oil of *C. sweitenia* was oxidised using $H_2O_2$. The odour of the oil was slightly improved and became agreeable. The time and temperature required for oxidation were found to be 12 hrs. and 50$^oC$ respectively. The amount $H_2O_2$ used to oxidise 1.0 ml of the essential oil was found to 0.76ml. GLC and GC-MS analysis reported the presence of eighteen components in the oxidised oil out of which three components could not be identified**.** β-caryophyllene oxide was found to be the major components of oxidised oil. The improvement of odour was due to change in the percentages of components after oxidation, which may probably be due to addition**,** elimination and rearrangement reactions and isomerisation of components. The increase in the concentrations of oxidised components i.e. Geraniol and β-caryophyllene oxide indicated that the oxygen content had increased after oxidation therefore the odour and quality of oil was improved.

## Conclusion

Oxidised essential oil of *C. sweitenia* can be used as antibacterial and antifungal agent against some specific bacteria and fungi if the in vitro studies hold good under in vivo conditions and can be marketed with good fragrance and quality for medicinal uses.

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