



### In-Vitro Hydrogen Peroxide Scavenging Activity of Ethanol Extract of *Canthium coromandelicum*

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**ABSTRACT**

**INTRODUCTION:** *Canthium coromandelicum* is a shrub widely indicated for dysentery. An antioxidant is molecule that inhibits the oxidation of other molecules. Antioxidants are widely used in dietary supplements and have been investigated for the prevention of diseases such as cancer, coronary heart disease and altitude sickness. In this study we have attempted to evaluate the free radical scavenging activity of *canthium coromandelicum* using in-vitro hydrogen peroxide.

**OBJECTIVE:** The objective of the present study was to evaluate the free radical scavenging activity and antioxidant potential of *Canthium coromandelicum*.

**MATERIAL & METHODS:** Hydrogen peroxide scavenging activity was determined according to the method of Ruch et al (1989). The principle of this method is that there is a decrease in absorbance of H<sub>2</sub>O<sub>2</sub> upon oxidation of H<sub>2</sub>O<sub>2</sub>. Hydrogen peroxide solution (40mM) was prepared in phosphate buffer (pH 7.4). *Canthium coromandelicum* extract (100 µg/mL) in distilled water was added to hydrogen peroxide solution (0.6 mL, 40mM). Absorbance of hydrogen peroxide at 540 nm was determined 10 minutes later against a blank solution containing the phosphate buffer without hydrogen peroxide. The resultant absorbance was recorded at 540 nm using spectrophotometer.

The percentage inhibition was calculated using the formula

$$\text{Percentage inhibition} = \frac{\text{Abs control} - \text{Abs sample}}{\text{Abs control}} * 100$$

**RESULTS:** At concentrations of 20, 40, 60, 80 µg/ml percentage of inhibition observed was 10.68±0.10, 20.39±0.37, 43.56±0.09 and 74.03±0.67 respectively.

**DISCUSSION & CONCLUSION:** Thus this invitro study suggest that *Canthium coromandelicum* possesses antioxidant properties. Prior phytochemical analysis *Canthium coromandelicum* shows the presence of flavonoids, tannins and glycosides (Mohan et.al). The antioxidant property of *Canthium coromandelicum* may be attributed to the presence of flavonoids and tannins. However pharmacokinetic and safety profile of *Canthium coromandelicum* requires pre-clinical testing prior to its clinical application in human beings.

**KEYWORDS**

*Canthium coromandelicum*, Nitric Oxide, In-Vitro

**INTRODUCTION:**

Various herbs have been tried in Indian conventional system of medicine for the treatment of number of conditions. The therapeutic value of these herbs lies in some chemical constituents that produce a specific physiological action on the human body. The most important of these bioactive substances of plants are alkaloids, tannins, flavonoids and phenolic compounds (Okeke and Elekwa, 2003). A very large variety of herbs have been evaluated for new source of natural anti-oxidant.

*Canthium coromandelicum* is a shrub, usually with side thorns a little above the leaf axils. Leaves are ovate and smooth. Short, few flowered racemes arise in leaf axils. Flowers are small and yellow. Tube is short, with 4-5 petals. Fruits are oval, furrowed on each side. Occurs throughout India, Indo-China, west and south China, Malaysia. Flowers between July-August [1].

**Fig 1: *Canthium coromandelicum* plant**



Prior phytochemical analysis *Canthium coromandelicum* shows the presence of flavonoids, tannins and glycosides (Mohan et.al). *Canthium coromandelicum* leaves are used for the treatment of diarrhoea. *Canthium coromandelicum* is a very significant medicinal plant, various phytochemical, antimicrobial and wound healing studies have already been carried out with *Canthium coromandelicum* leaf extract [2, 3].

In this study we have attempted to find the free radical scavenging activity of *Canthium coromandelicum* using in-vitro hydrogen peroxide assay.

**MATERIALS & METHODS:**

**Principle:**

Hydrogen peroxide assay is based on the principle that there is a decrease in absorbance of H<sub>2</sub>O<sub>2</sub> upon oxidation of H<sub>2</sub>O<sub>2</sub>.

**Plant source:** *Canthium coromandelicum* leaves were obtained from Shri Shail Herbs, Nagpur, Maharashtra, India during July 2015

**Reference antioxidant:** Quercetin

**Solvent:** Ethanol

**Preparation of extract:**

*Canthium coromandelicum* leaves were first washed with distilled water. About 5g of dried leaves were powdered and dissolved in 50ml of ethanol and kept overnight. The obtained extract was filtered. The ethanol was then removed under pressure to yield a concentrated extract.

**Fig.2: Dried leaves of *Canthium coromandelicum***



**Fig.3: *Canthium coromandelicum* extract**



**PROCEDURE:**

**Hydrogen Peroxide Scavenging Activity:** The ability of ethanolic extract of *Canthium coromandelicum* to scavenge hydrogen peroxide was determined according to the method of Ruch et al (1989). Hydrogen peroxide solution (40mM) was prepared in phosphate buffer (pH 7.4). *Canthium coromandelicum* extract (100 µg/mL) in distilled water was added to hydrogen peroxide solution (0.6 mL, 40mM). Absorbance of hydrogen peroxide at 230 nm was determined 10 minutes later against a blank solution containing the phosphate buffer without hydrogen peroxide.

**Fig.4: Spectrophotometer**



**Calculation:**

The percentage inhibition was determined using the formula

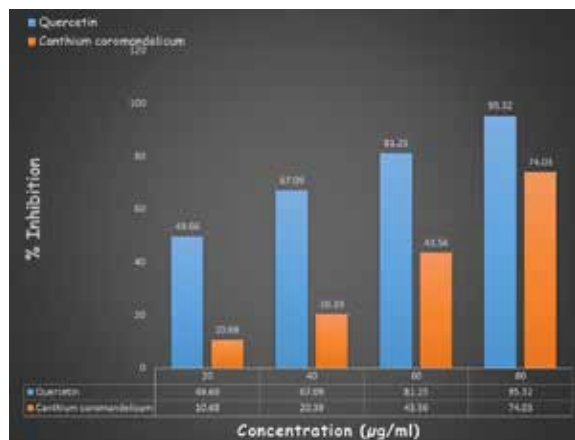
$$\% \text{ H}_2\text{O}_2 \text{ Scavenged} = \frac{\text{Abs control} - \text{Abs sample}}{\text{Abs control}} * 100$$

**RESULTS AND DISCUSSION:**

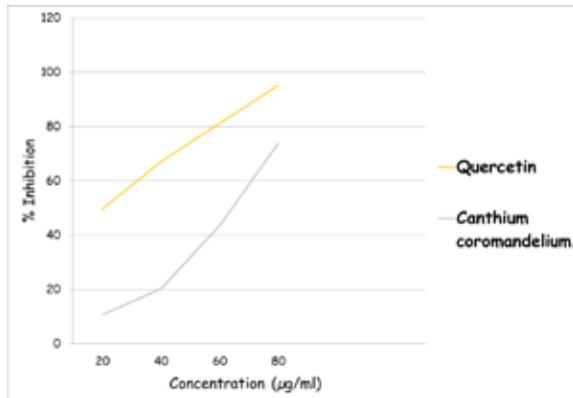
**TABLE 1: COMPARISON OF % INHIBITION OF NITRIC OXIDE BY QUERCETIN AND CANTHIUM COROMANDELICUM AT VARIOUS CONCENTRATIONS**

Concentration (µg/ml)	% Inhibition Quercetin	% Inhibition <i>Canthium coromandelicum</i>
20	49.66 ± 1.12	10.68±0.10
40	67.09 ± 0.16	20.39±0.37
60	81.25 ± 2.47	43.56±0.09
80	95.32 ± 1.07	74.03±0.67

**Fig 5: Comparison of % inhibition between Quercetin and *Canthium Coromandelicum***



**Fig 6: Comparison of % inhibition between Quercetin and Canthium Coromandelicum**



% inhibition of Canthium coromandelicum at concentration of 20, 40, 60, 80 µg/ml are 10.68±0.10, 20.39±0.37, 43.56±0.09 and 74.03±0.67 respectively.

The results from this study clearly indicate that Canthium coromandelicum has powerful antioxidant capacity under in vitro conditions. Canthium coromandelicum notably inhibited hydrogen peroxide in concentration dependent gradient. Many plants and its derivatives are considered to be a good sources of natural antioxidants. They have found wide medical uses such as against cancer, diabetic mellitus, cardiovascular diseases, aging etc. Finding natural antioxidants compounds will help us to introduce new drug candidates for antioxidant therapy. The antioxidant property of Canthium coromandelicum may be due to the existence of flavonoids and tannins. From this study we conclude that Canthium coromandelicum possesses antioxidant properties. Nevertheless pharmacokinetic and safety profile of Canthium coromandelicum requires pre-clinical screening prior to its clinical application in human beings.

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