

## PHARMACOLOGICAL SCREENING OF COMMIPHORA MUKUL LEAF EXTRACT

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## ABSTRACT

Commiphora mukul leaves were extracted by petroleum ether and it was isolated and purified by various spectral analyses. The spectral analysis revealed that terpenoids, steroids, flavonoids and tannins were found. The isolated compound was further evaluated for pharmacological screening methods. The isolated product was tested for acute toxicity studies and selected the dose. The selected dose was preferred for pharmacological screening methods. This dose was used in smooth muscle relaxant activity, but no significant activity was observed. The isolated compound was showed significant analgesic activity and anti-convulsant activity.

**KEYWORDS:** Commiphora mukul anti-convulsant activity.

## 1. INTRODUCTION

Commiphora mukul is a flowering plant in the family Burseraceae. It is a shrub or small tree, attaining maximum height of 4 m, with thin papery bark and thorny branches. The leaves are simple or trifoliate, the leaflets ovate, 1-5 cm long, 0.5-2.5 cm broad, irregularly toothed. Oleogum resin obtained from Commiphora mukul used for the medicinal preparation and commercially known as Indian bdellium or Guggul and the extract of this gum, called guggulipid, guggulipid or guglipid, located in ducts of the soft under bark.

The yellowish oleoresin collected in the form of pale yellow, aromatic fluid, flows out from bark that turn into a stalactic pieces and around 700 to 900 g resin collected from each four- to six-foot-tall tree. This Indian bdellium is antiseptic, ecboic, appetizing, aphrodisiac, emmenagogue, expectorant and used as menorrhagia, anaemia, leucorrhoea, rheumatism, nervous diseases, bone-fractures, obesity, disorder of lipid metabolism and peptic ulcer.<sup>[1,2]</sup> As per constituents, it contains 6.9% moisture, 0.6% volatile oil, 61% resin, 29.6% gum and 3.2% insoluble substances. In flowers quercetin, 3-O- $\alpha$ -L-arabinoside, 3-O- $\beta$ -D-galactoside-, 3-O- $\alpha$ -L-rhamnoside- and 3-O- $\beta$ -D-glucoronide, elagic acid and pelargonidin 3, 5-di-O-glucoside were obtained.<sup>[3,4]</sup>

## 2. AIM AND OBJECTIVES

- Extraction, Isolation and Pharmacological screening of Commiphora mukul leaf extract
- Spectral study of isolated compound
- Determination of Acute toxicity Studies and fixation of Dose

- Evaluation of smooth muscle relaxant activity from Commiphora mukul leaves extract with petroleum ether as solvent in mice.
- Evaluation of analgesic activity from Commiphora mukul leaves extract with petroleum ether as solvent in mice by Eddy's hot plate method
- Evaluation of anti convulsant activity from Commiphora mukul leaves extract with petroleum ether as solvent in mice by maximal electric shock induced convulsion

## 3. MATERIALS AND METHODS

## 3.1. Petroleum ether as solvent

Petroleum ether is the petroleum fraction consisting of C<sub>5</sub> and C<sub>6</sub> hydrocarbons and boiling in the range 35 to 60°C; commonly used as a laboratory solvent. The term ether is used only figuratively, signifying extreme lightness and volatility.

## Properties

The very lightest, most volatile liquid hydrocarbon solvents that can be bought from laboratory chemical suppliers may also be offered under the name petroleum ether. Petroleum ether consists mainly of C<sub>5</sub> and C<sub>6</sub> aliphatic hydrocarbons and is usually low on aromatics. It is commonly hydro desulfurized and may be hydrogenated to reduce the amount of aromatic and other unsaturated hydrocarbons. Petroleum ether bears normally a descriptive suffix giving the boiling range. Thus, from the leading international laboratory chemicals suppliers it is possible to buy various petroleum ethers with boiling ranges such as 30 to 50°C, 40 to 60°C, 50 to 70°C, 60 to 80°C, etc.

### 3.2. Soxhlet apparatus

A Soxhlet extractor is a piece of laboratory apparatus involved in 1879 by Franz von Soxhlet. It was originally designed for the extraction of a lipid from a solid material.

#### Assembly

1. The source of material containing the compound to be extracted is placed inside the thimble.
2. The thimble is loaded into the main chamber of Soxhlet extractor.
3. The extraction solvent to be used is placed in distillation flask.
4. The flask is placed on heating element.
5. The Soxhlet extractor is placed atop the flask.
6. A reflux condenser is placed atop the extractor.

### 3.3. Extraction procedure

- Collect Commiphora mukul leaves from medicinal plant garden in Vishwa Bharathi College of pharmaceutical sciences and then dried for 25 days. After completion of drying, the leaves were powdered by means of hands then extract with petroleum ether solvent.
- We have kept the powder by packing it in accurately weighed quantity and pour exactly 500mL of petroleum ether. Continue the process for 24 hours and collect the final sample and it was filtered and purified. The extracted solvent was subjected to evaporation and the solid sample was collected.

### 3.4. Phytochemical screening of Commiphora mukul leaf extract with petroleum ether as solvent.

Table-1: Qualitative chemical tests for Phytoconstituents

S.NO.	Name of the test	Petroleum ether extract
1.	Terpenoids	+
2.	Steroids	+
3.	Saponins	-
4.	Alkaloids	-
5.	Carbohydrates	-
6.	Flavonoids	+
7.	Tannins	+
8.	Glycosides	-

### 3.5. Spectral study of isolated compound of Commiphora mukul Leaf Extract

#### 3.5.1 NMR SPECTRA

The following is the interpretation of the NMR and it is very true values have been referred with a standard.

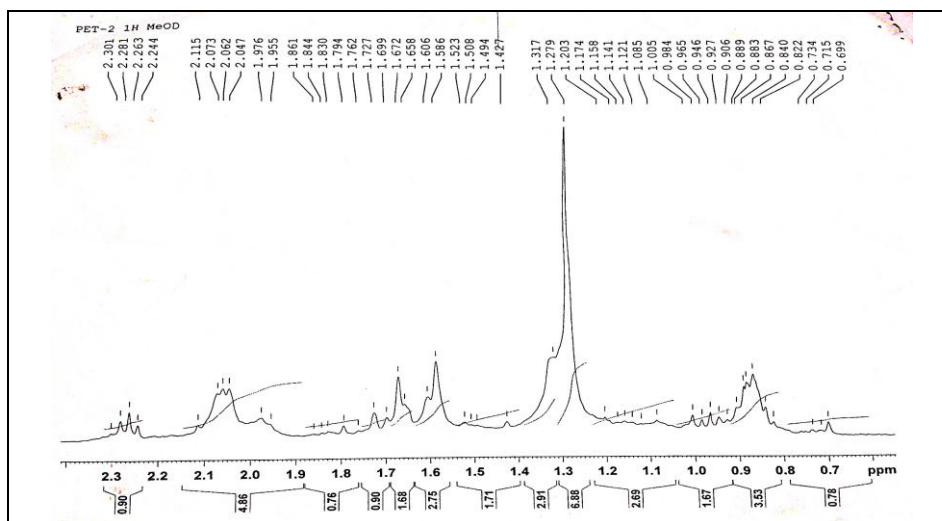


Fig: 1  $^1\text{H}$  NMR SPECTRA-ANALYTICAL DATA INTERPRETATION

#### Interpretation results (Table-2)

S.no	$\delta$ (chemical shift)-ppm	Nature of bond
1.	0.699-0.715	C-H Bending (alkyl group/methyl group)
2.	0.734-0.867	C-O Stretching (oxygen attached at $\alpha$ -position)
3.	0.833-0.906	C-O stretching
4.	0.906-0.927	C-O Bending
5.	0.946-0.965	C-O Bending
6.	0.984-1.174	C-H Bending (alkyl/methyl group presence)

7.	1.211-1.279	R-CH <sub>2</sub> group present and the molecule is undergoing a vibrational transition
8.	1.427-1.523	C-H Stretching (R <sub>3</sub> C-H)
9.	1.586-1.699	C-H Bending (Presence of allylic nature) (C is next to $\pi$ -bond)
10.	1.762-1.976	C-H Stretching (presence of methyl group)
11.	2.047-2.115	C is next to a phenol (benzyl aromatic ring)
12.	2.244-2.307	C-O Bending (presence of an alcoholic bond and the carbon is attached at $\beta$ -position)

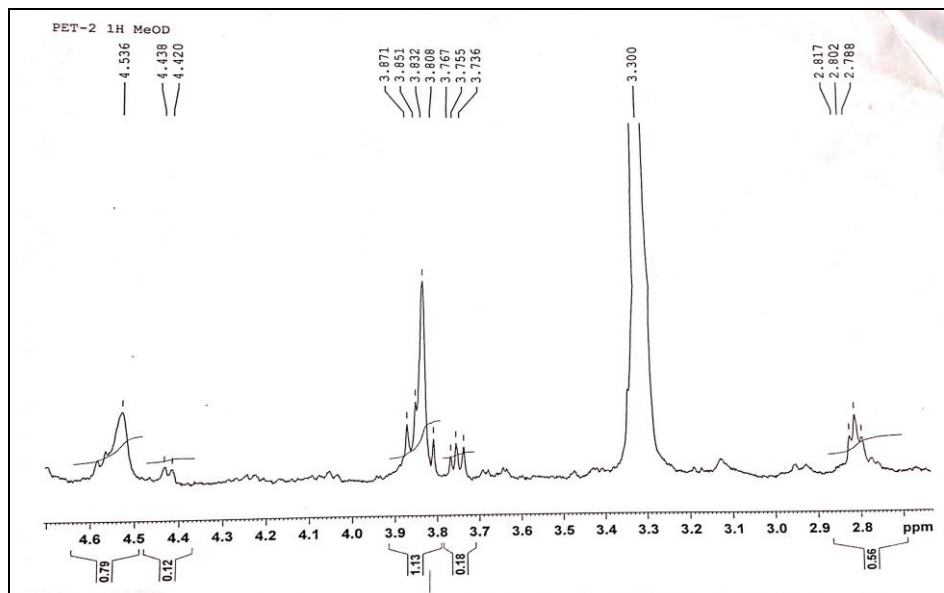


Fig: 2 <sup>1</sup>H NMR SPECTRA-ANALYTICAL DATA INTERPRETATION

#### Interpretation results (Table-3)

S.no	$\delta$ (chemical shift)-ppm	Nature of bond
1.	2.788-2.817	Presence of C=O and the molecule undergone stretching (carbon is attached to oxygen) (R-CH <sub>2</sub> )
2.	3.300	C-O stretching (carbon is attached to oxygen) ( $\alpha$ to oxygen)
3.	3.736-3.871	C-H scissoring ( $\alpha$ to oxygen)
4.	4.420-4.536	Presence of steroid skeletal nuclei; C-H bending

#### 3.5.2. MASS SPECTRA

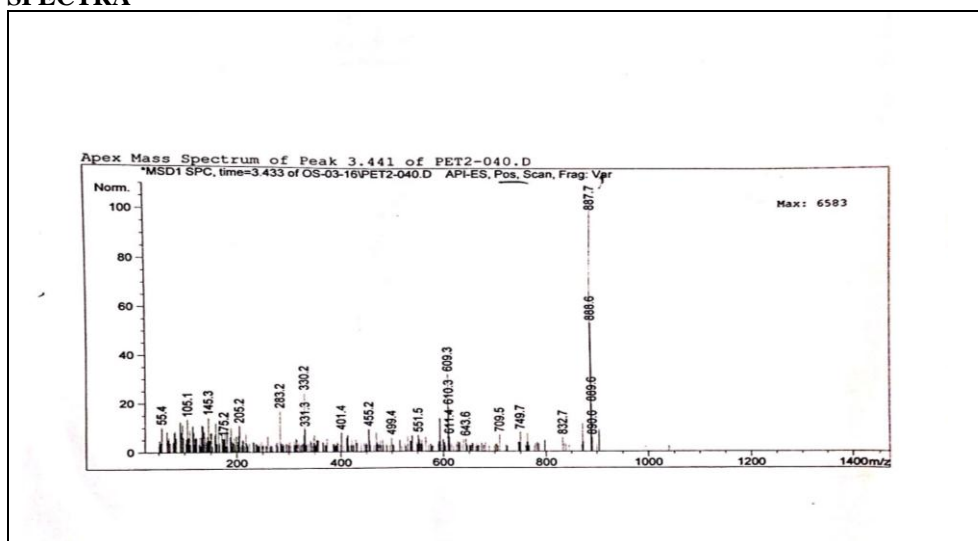


Fig: 3. THE MASS SPECTRUM FOR GUGGUL STERONE HAS 40 PEAKS.

## Interpretation results (Table-4)

S.No.	Peak value	Interpretant Data
1.	55.4	$\text{CH}_3\text{CH}_2^+$
2.	105.1	$\text{CH}_2\text{CH}_2\text{CH}_3$
3.	145.3	$\text{CH}_3\text{CH}_2\text{OH}$
4.	175.2	$\text{CH}_2\text{CH}_2\text{O}$
5.	205.2	$\text{CH}_2\text{CHCHOH}$
6.	283.2	$(\text{CH}_2\text{CH}_3)_2$
7.	331.3	$\text{CH}_2\text{CH}^+$
8.	401.4	$\text{CHCH}^+$
9.	455.2	$\text{CH}_2\text{C}=\text{O}$
10.	499.4	$\text{CH}_3\text{CH}_2\text{CH}_2$
11.	551.5	$\text{CH}_3\text{C}-\text{OH}$
12.	611.4	$\text{CH}_3\text{CHCHCHCH}_2$
13.	643.6	$\text{CHOH}^+$
14.	709.5	$\text{CH}_2\text{CHCHCH}_2^+$
15.	749.7	$\text{CH}_3\text{CH}_2\text{OH}^+$
16.	832.7	$\text{CH}_2\text{OH}^+$
17.	890.6	$\text{CH}_2\text{CHCH}_3^+$

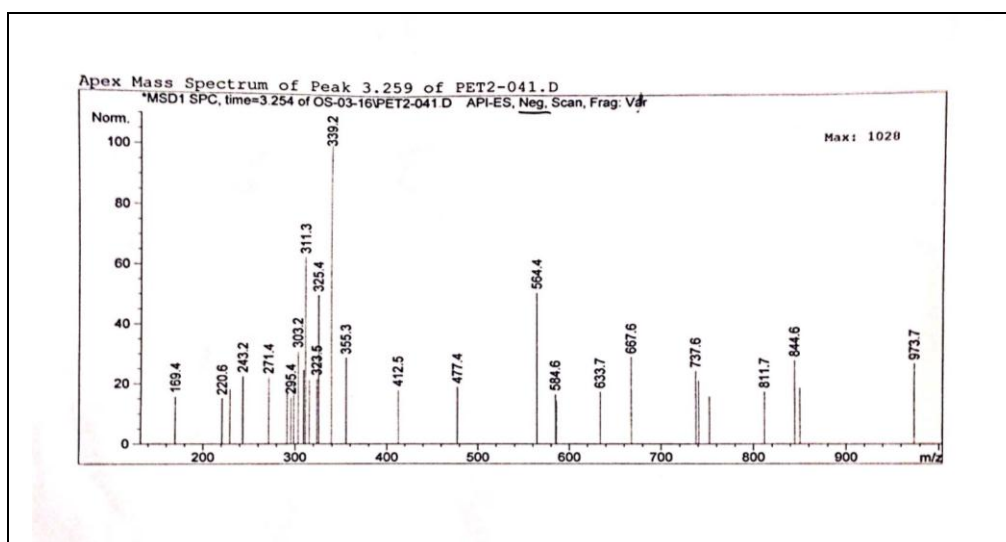


Fig: 4. MASS SPECTRUM FOR GUGGUL STERONE

## Interpretation results (Table-5)

S.No.	Peak value	Interpretant Data
1.	169.4	$\text{CH}_3\text{CH}_2\text{OH}$
2.	220.6	$\text{CH}_2\text{CHCHOH}$
3.	24.2	$\text{CH}_2\text{CHCHOH}$
4.	271.4	$(\text{CH}_2\text{CH}_3)_2$
5.	295.4	$(\text{CH}_2\text{CH}_3)_2$
6.	303.2	$(\text{CH}_2\text{CH}_3)_2$
7.	311.3	$\text{CH}_2\text{CH}^+$
8.	323.5	$\text{CH}_2\text{CH}^+$
9.	339.2	$\text{CH}_2\text{CH}^+$
10.	355.3	$\text{CH}_2\text{CH}^+$
11.	412.5	$\text{CHCH}^+$
12.	477.4	$\text{CH}_2\text{C}=\text{O}$
13.	564.4	$\text{CH}_3\text{C}-\text{OH}$
14.	584.6	$\text{CH}_3\text{CHCHCHCH}_2$
15.	633.7	$\text{CH}^+-\text{OH}$
16.	667.6	$\text{CH}^+-\text{OH}$
17.	737.6	$\text{CH}_3\text{CH}_2\text{OH}^+$

18.	811.7	CH <sub>2</sub> OH <sup>+</sup>
19.	844.6	CH <sub>2</sub> OH <sup>+</sup>
20.	937.7	CH <sub>2</sub> CHCH <sub>3</sub> <sup>+</sup>

#### 4. ACUTE ORAL TOXICITY STUDIES

1. The petroleum ether extract of Commiphora mukul leaves was isolated, purified and carried out the spectral analysis.

2. This isolated product was used to determine the acute oral toxicity study and determine the safe doses of compound. Based on the toxicity, the doses were selected for further pharmacological screening methods.

3. The dose of product was calculated by using up and down procedure according to OECD guidelines 423.

4. The initial dose is 0.2 ml having 200mg.

5. The second dose used for screening is 500 mg

6. The lethal dose (LD<sub>50</sub>) of isolated product was 1000mg

7. The maximal dose is 2000mg is considered as toxic dose. At this dose all the animals were died.

The preferable doses were selected for further pharmacological screening methods.

Table 6:

S.No.	Selected dose	Results
1.	200mg	No toxicity was observed
2.	500mg	No toxicity was observed
3.	1000mg	Lethal dose (LD <sub>50</sub> )
4.	2000mg	Toxicity observed

#### 5.1. PHARMACOLOGICAL SCREENING METHODS

##### 5.1.1. Evaluation of smooth muscle relaxant property of petroleum ether extract of leaves of Commiphora mukul in mice.

##### Principle

One of the important pharmacological actions of anti-anxiety agents of benzodiazepine class of drugs is muscle relaxing property. The skeletal muscle relaxation together with taming or calming effect these agents reduces anxiety and tension. The loss of muscle-grip is an indication of muscle relaxation. This effect can be easily studied in animals using inclined plane or rotating rods. The difference in the fall off time from the rotating rod between the control and diazepam- treated animal is taken as an index of muscle relaxation. The angle of the slope of the inclined plane, or the rate of rotation of the rod should be adjusted such that a normal mouse can stay on the rod for an appreciable period (3-5 min) of time.

##### Requirements

**Animal:** mice (20-25 g).

**Drugs:** diazepam (dose 4mg/kg, ip; prepare a stock solution containing 0.4mg/ml of the drug and inject 1ml/100 g of body weight of the mouse. Diazepam is suspended in 1% w/v gum acacia or carboxy methylcellulose).

**Equipment:** Rota-rod apparatus (techno) or devise your own rotating-rod as shown in the diagram.

##### Experimental work

1. Weigh the animals and number them.
2. Turn on the rota-rod by one on the rotating rod. Select an appropriate speed (20-25 rpm is ideal).
3. Place the animal one by one on the rotating rod. (If the rod is divided into several compartments, one can place more than one mouse at a time). Note down the fall off time when the mouse falls from the rotating rod. A normal (untreated) mouse generally falls off within 3-5 minutes.
4. Inject diazepam (4mg/kg) to all the animals. After 30 mints repeat the experiment as done in Step-3. Note the fall off time.
5. Compare the fall off time of animals before and after diazepam treatment.<sup>[5]</sup>
6. The same procedure is repeated by using the petroleum ether extract of commiphora mukul leaves.
7. The calculated dose of 0.2 ml of extracted drug was given orally to the mice one by one.
8. They were placed on the Rota-rod by one by and observed the fall off time of each animal.

#### Smooth muscle relaxant property of petroleum ether extract of leaves of Commiphora mukul in mice

Table 7:

S.No.	Drug	Dose in mL	Animal body weight (grams)	Fall-off time in seconds		%decrease in activity/time
				Before drug administration	After drug administration	
1.	Control	-	20	80	-	No activity
2.		-	20	90	-	
3.		-	22	80	-	
4.		-	18	90	-	
Average values			20	85	-	

**Table 8: Smooth muscle relaxant property of diazepam**

S.No.	Drug	Dose in mL	Animal body weight (g)	Fall-off time in seconds		%decrease in activity/time
				Before drug administration	After drug administration	
1.	standard (diazepam)	0.006	20	100	60	36.82
2.		0.006	22	90	40	
3.		0.006	18	80	50	
4.		0.006	20	70	60	
Average values			20	85	52.5	

$$\% \text{ decrease in activity/time} = \frac{\text{before drug administration} - \text{after drug administration}}{\text{Before drug administration}} \times 100$$

$$= \frac{85-52.5}{85} \times 100$$

$$= 36.82$$

**Table 9: Smooth muscle relaxant property of Commiphora mukul leaf extract**

S.No.	Drug	Dose in mL	Animal body weight (grams)	Fall-off time in seconds		%decrease in activity/time
				Before drug administration	After drug administration	
1.	Test	0.2	20	110	105	No activity
2.		0.2	20	120	110	
3.		0.2	20	80	75	
4.		0.2	20	90	89	
Average values			20	100	94.75	

$$\% \text{ decrease in activity/time} = \frac{\text{before drug administration} - \text{after drug administration}}{\text{Before drug administration}} \times 100$$

The petroleum ether extract of commiphora mukul leaves no significant smooth muscle relaxant property.

### 5.1.2. Evaluation of Analgesic activity from petroleum ether extract of Commiphora Eddy's hot plate mukul leaves in mice by method.

**Principle:** in this method the heat is used as a source of pain. Animals are individually placed on a hot plate maintained at constant temperature (55°C) and the reaction of animals, such as paw licking or jumping response is taken as the end point. Analgesics increase the reaction-time. The method was first described by Eddy and Leimbach (1953).

#### Requirements

**Animals:** mice (20-25g).

**Drugs:** Diclofenac sodium (dose 5mg/kg, s.c., prepare a stock solution containing 0.5mg/ml and inject 1ml/100 g of body weight of mouse).

**Equipment:** Eddy's hot plate (techno).

#### Experimental work

1. Weigh and number the mice.
2. Take the basal reaction-time by observing hind paw licking or jump response (whichever appears first) in

animals when placed on the hot plate maintained at constant temperature (55°C). Normally animals show such response in 6-8 sec. A cut off period of 15 sec is observed to avoid damage to the paws.

3. Inject Diclofenac to animals and note the reaction time of animals on the hot plate at 15, 30, 60 and 120 min after the drug administration. As the reaction time increase with Diclofenac, 15 sec is taken as maximum analgesic and the animals are removed from the hot plate to avoid injury to the paws.
4. Calculate percent increase in reaction-time (as index of analgesia) at each time interval.<sup>[6]</sup>
5. The same procedure is repeated by using the petroleum ether extract of commiphora mukul leaves.
6. The calculated dose of 0.2 ml of extracted drug was given orally to the mice one by one.
7. The analgesic activity of mice was observed by placing the drug given mice by paw licking and jump response by maintaining the temperature at 55°C.

**Table 10: Analgesic activity from petroleum ether extract of Commiphora mukul leaves in mice by Eddy's hot plate method.**

S.No.	Animal(Mice) Body.wt (gm)	Drug	Paw licking & jump response in sec at particular time interval											
			0min		30min		60min		90min		120min		150min	
			P	J	P	J	P	J	P	J	P	J	P	J
1.	20	Control	4	8	4	9	5	9	3	5	2	6	3	7



2.	20		3	5	3	6	3	7	2	5	2	6	2	7
3.	20		2	7	3	6	2	3	1	7	1	8	2	6
4.	20		3	6	2	5	2	6	1	4	3	7	3	8
Average values of jump responses			6.5		6.5		6.25		5.25		6.75		7.00	

Table 11: Analgesic activity of Diclofenac sodium

S.No.	Animal Body.wt (gm)	Drug	Paw licking & jump response in sec at particular time interval											
			0min		30min		60min		90min		120min		150min	
			P	J	P	J	P	J	P	J	P	J	P	J
1.	20	Standard (Diclofenac) (3mL/60kg b.w) (0.006mL)	6	12	5	13	5	10	5	12	6	13	7	14
2.	20		7	12	7	13	6	13	7	13	7	14	7	15
3.	20		6	10	6	11	6	11	6	12	7	13	7	15
4.	20		7	10	7	11	7	12	7	12	7	14	7	15
Average values of jump responses			11		12		11.5		12.25		13.5		14.75	

Table 12: Analgesic activity of Commiphora mukul leaf extract

s.no	Animal Body.wt (gm)	Drug	Paw licking & jump response in sec at particular time interval											
			0min		30min		60min		90min		120min		150min	
			P	J	P	J	P	J	P	J	P	J	P	J
1.	20	Test	8	12	7	14	6	14	8	15	8	15	7	14
2.	20		6	10	5	11	5	12	5	12	7	15	8	15
3.	20		6	11	7	12	6	12	8	12	8	12	8	15
4.	20		8	13	4	8	3	7	8	9	4	13	8	15
Average values of jump responses			11.5		11.25		11.25		12		13		14.75	

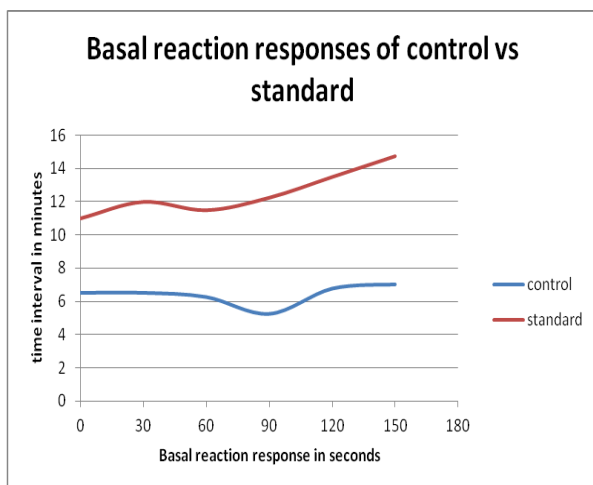


Fig: 5

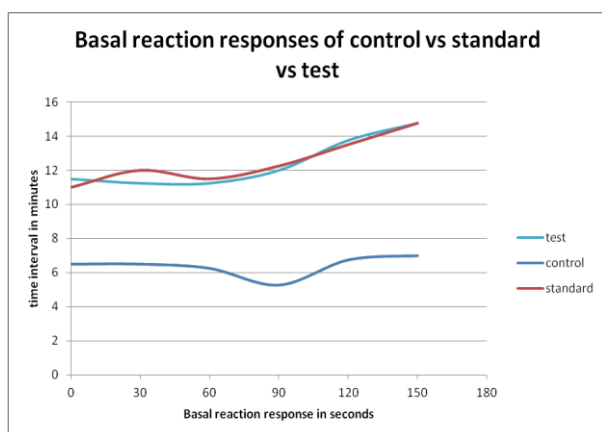


Fig: 6

### 5.1.3. Evaluation of Anti Convulsant activity of petroleum ether extract of Commiphora mukul leaves in mice.

#### Principle

Different types of epilepsies i.e., grand mal, petit mal or psychomotor type, can be studied in laboratory animals. The maximal electro-shock (MES) induced convulsions in animals represent grand mal type of epilepsy. Similarly, chemo-convulsions due to pentylene tetrazol which produce clonic-type of convulsions resemble petit mal type of convulsions in man. These are the two procedures used to study convulsions and to test anti-convulsant drugs in laboratory animals.

In MES-convulsions electro shock is applied through the corneal electrodes. Through optic stimulation cortical excitation is produced. The MES-convulsions are divided into five phases such as (a) tonic flexion, (b) tonic extensor, (c) clonic convulsions, (d) stupor and (e) recovery or death. A substance is known to possess anti-convulsant property if it reduces or abolishes the extensor phase of MES convulsions. This procedure may be used to produce convulsions both in rats and mice. It is advised that the students should have complete background of the pharmacology of anti-epileptic drugs before performing this experiment.

#### Requirements

**Animal:** mice (20-25g).

**Drugs:** diazepam (dose 25mg/kg; prepare a stock solution containing 5mg/ml of the drug and inject 0.5ml/100 g body weight of the animal).

**Equipment**

Electro-convulsometer, corneal electrode (apply 40 mA current for 0.2 sec), stop-watch.

**Experimental work**

1. Weigh and number the animals. Divide them into two groups each consisting of 4-5 mice. One group is used as control and the other for drug (diazepam) treatment.
2. Hold the animal properly, place corneal electrode on the cornea and apply the prescribed current. Note different stages of convulsions i.e. (a) tonic flexion, (b) tonic extensor, (c) clonic convulsions, (d) stupor and (e) recovery or death. Note the time (sec) spent by the animal in each phase of the convulsions. Repeat with other animals of control group.
3. Inject diazepam intra-peritoneally to a group of 4-5 mice. Wait for 30 min and subject the animals to electro-convulsions as described in step-2.
4. Note the reduction in time or abolition of tonic extensor phase of MES-convulsions.
5. Repeat the same procedure by using the petroleum ether extract of commiphora mukul leaves.<sup>[7-9]</sup>
6. The calculated dose of 0.2 ml of extracted drug is given orally to the mice one by one.
7. The electric shock of 40 mA is given with corneal electrodes for 0.2 sec and observed the following characteristics.

**Table 12: Anti convulsant activity of petroleum ether extract of Commiphora mukul leaves in mice.**

S.No.	Animal weight	Drug	dose	Time in sec in various phases of convulsions				
				flexion	extensor	clonus	stupor	Recovery/Death
1.	20g	Control	-	3sec	11sec	3sec	125sec	Died
2.	20g		-	2sec	13sec	1sec	85sec	Recovery
3.	20g		-	3sec	12sec	0sec	100sec	Died
4.	20g		-	1sec	10sec	0sec	95sec	Died

**Table 13: Anti convulsant activity of diazepam**

S.No.	Animal weight	Drug	Dose	Time in sec in various phases of convulsions				
				flexion	extensor	clonus	stupor	Recovery/Death
1.	20g	Standard (diazepam)	0.006mL	3sec	10sec	1sec	85sec	Recovery
2.	20g		0.006mL	2sec	9sec	1sec	75sec	Recovery
3.	20g		0.006mL	2sec	10sec	1sec	88sec	Recovery
4.	20g		0.006mL	2sec	10sec	0sec	90sec	Recovery

**Table 14: Anti Convulsant activity of Commiphora mukul leaf extract**

S.No.	Animal weight	Drug	Dose	Time in sec in various phases of convulsions				
				flexion	extensor	Clonus	stupor	Recovery/Death
1.	20g	Test	0.2mL	4sec	13sec	0sec	80sec	Recovery
2.	20g		0.2mL	3sec	10sec	0sec	95sec	Recovery
3.	20g		0.2mL	2sec	9sec	1sec	72sec	Recovery
4.	20g		0.2mL	3sec	10sec	0sec	85sec	Recovery

**6. RESULTS AND DISCUSSION**

The collected Commiphora mukul leaves were dried perfectly under shade and powdered into fine form by means of hand crushing. The crude drug was weighed for 20gm and kept in apparatus. The calculated volume of solvent was poured in soxhlet apparatus for extraction for 48 hours. The extracted drug in solvent was subjected to evaporation and dried it and then it was processed for purification and isolation by spectral analysis. In commiphora mukul leaf extract having chemical constituents those are; Terpenoids, steroids, flavonoids and tannins are present in commiphora mukul leaf extract. Saponins, alkaloids, carbohydrates and glycosides are absent in commiphora mukul leaf extract. The mice were collected from animal house, Vishwa Bharathi College of pharmaceutical sciences for experimental purpose.<sup>[10-15]</sup>

The smooth muscle relaxant activity was tested by rota-rod apparatus in mice with drug (test sample) and standard drug. The selected animals were mice. Weigh the animals and number them. Divide the animals into three groups, each group contains four mice. The first group is considered as control animals which animals were tested for smooth muscle relaxant property without drug by keeping them on rota-rod apparatus maintained for 25-30rpm. The second group is considered as standard animals. The standard drug diazepam is given intraperitoneally in dose of 0.006mL. Observe the smooth muscle relaxant activity when the animal fell down from the rota-rod. The third group is considered as test animals to which the extracted drug was administered orally in dose of 0.2ml. Keep each animal on rota-rod and observe the fall-off time in seconds taking 25-30rpm. Compare the values with standard group of animals and the results were evaluated.



The analgesic activity in mice was carried out by eddy's hot plate method maintaining the temperature at 55°C. The Diclofenac sodium was used as standard drug for analgesic activity studies. The selected animals were mice and divided into three groups, each group contains four mice. The first group of animals was considered as control animals. Each animal was tested on eddy's hot plate without drug administration. The second group is considered as standard animals and treated by standard drug Diclofenac through intraperitoneal at the dose of 0.006mL. Observe the paw licking and jump responses (basal reaction responses) at particular time interval. The responses were observed in seconds. The third group of mice is considered as testing animals. The calculated dose of 0.2mL of petroleum ether extract of commiphora mukul leaves was administered orally to the mice. The paw licking and jump responses were observed by keeping them on eddy's hot plate. The observed values in seconds were compared as that with standard group animals, results were evaluated.

The anticonvulsant activity was evaluated by applying maximal electric shock induced convulsions in mice by using electro convulsometer. Kept the corneal electrodes on cornea of mice at the voltage of 40mV. Induce the various phases of convulsions note the time in seconds of various phases and observe the state of animal whether it might recovery or died. The selected animals were mice weigh and number the animals. Divide the animals into three groups, each group containing four animals. The first group is considered as control animals. They were tested without drug by holding animal in position keeping the corneal electrodes for 0.2sec and observed the phases of convulsions in mice. The second group of animals was considered as standard group. The standard drug is diazepam, given to animals intraperitoneally at the dose of 0.024 units. The corneal electrodes were kept on cornea for 0.2 sec. The various phases observed in mice. The third group of animals was considered as test animals. The above procedure is followed, by administered isolated drug at dose of 0.2mL, orally the various phases were observed. The observed values were compared with standard group. Based on the result of standard the isolated product of commiphora mukul leaf extract has showed the anticonvulsant activity.

## 7. CONCLUSION

Commiphora mukul leaves were extracted by petroleum ether and it was isolated and purified by various spectral analysis. The spectral analysis revealed that terpenoids, steroids, flavonoids and tannins were present. The isolated compound was further evaluated for pharmacological screening methods. The isolated product was tested for acute toxicity studies and fixed the safe dose. The selected dose was preferred for pharmacological screening methods. This dose was used in smooth muscle relaxant activity, but no significant activity was observed. The isolated compound was

showed significant analgesic activity and significant anti-convulsant activity.

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