International Journal of Pharmaceutical Research and Applications
Volume 7, Issue 6 Nov-Dec 2022, pp: 64-69 www.ijprajournal.com ISSN: 2456-4494

# Evaluation of Laxative Activity of Caesalpinia Sappan Wood Extracts in Rats.

Bharathi KN[1], Darsana Nair[2]

*Department of Pharmacology , M Pharm , Visveswarapura Institute of Pharmaceutical Science , 22nd main road , Siddanna layout , Banshankari stage 2 , Bengaluru , Karnataka , 570070 ,India*

Submitted: 25-10-2022 Accepted: 05-11-2022

**ABSTRACT**: - This study was undertaken to examine the laxative activity of ethanolic and aqueous extract of Caesalpinia Sappan wood using drug induced and low fiber diet induced constipation model and Castor oil induced enter poolingin rats .The Gaviscon was used as standard laxative and loperamide was used for inducing constipation in drug induced constipation model.Low fiber diet included 24.5 percent casein milk, 10.0 percent Sucrose, 7.0 percent mineral Mixture, and 1.0 percent Vitamin combination.After inducingConstipation,extracts were administered orally, in the doses of 30 and 60 mg/kg b.w., onceto respective groups.The number and weight of the faecal pellet was determined . Treatment with 60 mg/kg b.w.($p<0.01$) aqueous extract and both doses of ethanolic extracts significantly ($p<0.01$, $p<0.001$) increased number and weight of faecal pellets compared to negative control group, exhibiting dose dependent laxative activity.Ethanolic extract was found to be more potent than aqueous extract and comparable with standard.In the low fiber dietinduced constipation model , treatment with extracts significantly ($p<0.001$) increased the weight of the faecal pellet compared to negative control group indicating laxative property.

There was no difference between the doses and extracts with respect to potency.In castor oil induced enter poolingmodel, castor oil was administered to induce diarrhea.Treatment with extracts inhibited castor oil induced diarrhea in rats .The chemical constituents present in Caesalpinia sappan wood such as sappanchalcone and anthraquinones is responsible for laxative activity.

**KEYWORDS:**Caesalpinia Sappan ,Laxative, Sappanchalcone , low fiber diet .

## I. INTRODUCTION

Laxatives are a type of medicine that can treat constipation. Constipation is a condition in which there is difficulty in emptying the bowels, usually associated with hardened faeces. Laxatives can be taken orally in the form of liquids, tablets, or capsules. They can also be taken through the rectum as, suppositories or enemas. In Kerala,boiled infusion of Caesalpinia sappan wood isserved as drinking water(karikaali vellam) since it is having curing actions of wounds , rheumatism , diarrhoea ,laxative and allergic reactions [2]. But it lacks scientific evidence . Most of the laxative herbs such as Senna,Chamomile, Liquorice root, Dandelion, Ginger contain Sappanchalcone,anthraquinones glycosides, barbaloinor, substances that have a stimulant effect on the intestines. The same chemical constituents are also present inCaesalpinia sappanwood. Hence in the present study we intended to find out Laxative activity of aqueous and ethanolic wood extracts of Caesalpinia sappan in Rats[1][3][4].

## II. METERIALS AND METHODS

### PLANT MATERIAL

Caesalpinia sappan wood was procured from the local market in Kerala. The sample was authenticated by the university of Trans – Disciplinary Health Science and technology , Bengaluru , 560064.

The wood of Caesalpinia sappan were washed with water for removal of unwanted presence and dried at room temperature.

### PREPARATION OF EXTRACT

Extraction was done using Reflux condenser.

Dried wood was powdered and soaked in distilled water for 24 hours ( 1:4 ) ratio in round bottom flask for aqueous extraction and in ethanol for 24 hours (1:4 ratio) for ethanolic extraction and heated using heating mantle for 3 hours. The filtratewas collected through watsman filter paper and evaporated by usingwater bath.

International Journal of Pharmaceutical Research and Applications
Volume 7, Issue 6 Nov-Dec 2022, pp: 64-69 www.ijprajournal.com ISSN: 2456-4494

## III. EXPERIMENTATION.

### EXPERIMENTAL ANIMAL

Male Albino rats of wistar strain , weighing 150-230grams , maintained on a 12±1 hours day and night schedule, fed with standard diet and water ad libitum were used in the study.The study protocol was approved by the Institutional Animal Ethical Committee,Visveswarapura Institute of Pharmaceutical Science ,Bangalore , 560070 . (Registration no : 152/po/ReBiBt /S/99/CPCSEA). IAEC approval number: VIPS / IAEC/25-08-2021/09-KNB.

### DRUG INDUCED CONSTIPATION IN RAT MODEL

MaleAlbinoRats ofwistar strain , were assigned randomly to seven group of six animal each.Food was withdrawn for 12 hours prior to experimentation , with water ad libitum.Extracts and standard were administered orally once .

Group Iand IIanimals received vehicle,served as positive and negativecontrolrespectively .Group III to VI received 30 and 60 mg/kg b,w., aqueous andethanolicextracts respectively,Group VIIreceived standard Gaviscon 10mg/ kg b.w.,.

All groups except group 1 received loperamide3mg/kg b.w.p.o., 30 min prior to treatment with extracts and standard . The animals were placed in metabolic cages immediately following drug treatment. The output of faeces was quantified in all groups by evaluating faeces Weight after 6 hours[5][6].

### LOW FIBER DIET INDUCED CONSTIPATION IN RATS

36 Male AlbinoRats ofwistarstrain, were divided into six group of six animals each.Food was withdrawn for 12 hours prior toexperimentation, with water ad libitum.The low fiber diet included 24.5 % casein milk, 10.0 % Sucrose, 7.0 % mineral Mixture,and 1.0 % Vitamin combination(zincovit) . The low fiber mixture was given to all groups,through feeding bottle for one hour except group I, which served as positive control,and placed in metabolic cages.After one-hour animals were administered with single doses of extracts orally.Group I and IIserved as positive andnegative control respectively, received only vehicle.Group III to VI received 30 and 60 mg/kg b,w., aqueous andethanolic extracts respectively. Fecal matter was collected and weighed at the end of 24 hours[8].

### CASTOR OIL INDUCED ENTER POOLING

Male AlbinoRats ofwistarstrain were assigned randomly to six group of six animals each.Food was withdrawn for 12 hours prior toexperimentation, with water ad libitum.All the groups received1 ml of castor oil orally except group I and placed in metabolic cages.After one hour, animals were administered with single doses of extracts orally. Group I and IIreceived vehicle ,served as positive and negative control respectively.Group III to VI received 30 and 60 mg/kg b,w., aqueous andethanolic extracts respectively.Theoutput of faecalmatter was quantified in all groups by evaluating faeces weight after 2 hours[7].

## IV. OBESERVATIONS AND RESULTS

The yield of aqueous and ethanolic extracts of Caesalpinia sappan wood was found to be6.875 % and 6.12 % respectively . The following phytochemical constituents were present in the extract when analysed qualitatively(TableNo. 1) .

**Table number 1**; Phytochemical constituents of Caesalpinia sappanwood extracts.

| CHEMICAL CONSTITUENTS | AQUEOUS EXTRACTS | ETHANOLIC EXTRACTS |
|---|---|---|
| Flavonoids | + | + |
| Phenolic compounds | + | + |
| Anthraquinones | + | + |
| Sappanchalcone | + | + |
| Steroids | + | + |

International Journal of Pharmaceutical Research and Applications
Volume 7, Issue 6 Nov-Dec 2022, pp: 64-69 www.ijprajournal.com ISSN: 2456-4494

| | | |
|---|---|---|
| Glycosides | + | + |

+ = Present; – = absent.

**Table number2;**Effect of Caesalpinia sappanwood extract in drug induced constipation in rats.

| **Groupings and Treatment** | **Weight of feces 'g'** | **Number of feces** |
|---|---|---|
| I.Positive Control | 1.78±0.36 | 1.60 ± 0.35 |
| II . Negative control | 1.06±0.65 # | 1.02± 0.6 # |
| III. Aqueous extract of Caesalpinia sappanwood30 mg/kg | 1.55±0.32 | 1.5 ± 0.31 |
| IV. Aqueous extract of Caesalpinia sappanwood 60mg/kg | 2.27±0.25 ** | 2.32 ± 0.27 ** |
| V. Ethanolic extract of Caesalpinia sappan wood 30mg/kg | 2.24±0.72 ** | 2.30 ± 0. 70 ** |
| VI. Ethanolic extract of Caesalpinia sappan wood 60mg/kg | 2.58±0.82 *** | 2.60 ± 0.90 *** |
| VII. Gaviscon(std dose) | 2.89±0.38 *** | 2.92 ± 0. 35 *** |

n = 6, values are mean ± S.E.M, one way ANOVA followed bypost hockDunnett's test
#p<0.05as compared to positive control.
** P<0.01, *** p<0.001 as compared to negative control

As shown in Table no 2, Loperamide induced constipation, as there was significant (p<0.05 ) decrease in the stool number and weight of faeces in group IIrats when compared to group I .Treatment with extracts and standard significantly increasedfaecal parameters compared to negative control except aqueous 30 mg/kg treated rats.Extracts exhibited dose dependent laxative activity. Ethanolic extract was found to be more potent than aqueous extract and comparable with standard.

**Table number3 ;**Effect of Caesalpinia sappanwood extracts in Low fiber induced constipation in rats.

| **Grouping and Treatment** | **Weight of feces 'g'** |
|---|---|
| I. Positive Control | 2.69± 0.16 |
| II . Negative control | 0.71 ± 0.24*** |
| III. Aqueous extract of Caesalpinia sappanwood30 mg/kg | 3.22 ± 0.12+++ |

International Journal of Pharmaceutical Research and Applications
Volume 7, Issue 6 Nov-Dec 2022, pp: 64-69 www.ijprajournal.com ISSN: 2456-4494

| | |
|---|---|
| IV. Aqueous extract of Caesalpinia sappanwood 60mg/kg | 3.33± 0.30+++ |
| V. Ethanolic extract of Caesalpinia sappan wood 30mg/kg | 3.44 ± 0.38+++ |
| VI. Ethanolic extract of Caesalpinia sappan wood 60mg/kg | 3.46 ± 0.28 +++ |

n = 6, values expressed as mean ± S.E.M, one way ANOVA followed bypost hockDunnett's test , ***P<0.001 as compared positive control , +++ p <0.001 as compared to negative control.

In low fiber diet induced constipation, group II animals compared to positive control as there was significant (***P<0.001) decrease in fecal weight .Treatment with extracts significantly (P<0.001) increased the weight of the faecal weight compared to negative control indicating laxative property. There was no difference between the doses and extracts with respect to potency as shown in table no 3.

**Table number4;**Effect of Caesalpinia sappanwood extracts in castor oil induced enter pooling in rats .

| **Grouping and Treatment** | **Weight of wet feces 'g'** |
|---|---|
| I.Positive Control | 2.37 ± 0.41 |
| II.Negative control (castor oil) | 3.99 ± 0.27 * |
| III. Aqueous extract of Caesalpinia sappanwood30 mg/kg | 3.32 ± 0.44 |
| IV. Aqueous extract of Caesalpinia sappanwood 60mg/kg | 3.03 ± 0.21 |
| V. Ethanolic extract of Caesalpinia sappan wood 30mg/kg | 2.71 ± 0.46 |
| VI. Ethanolic extract of Caesalpinia sappan wood 60mg/kg | 3.07 ± 0.26 |

n = 6, values are mean ± S.E.M, one way ANOVA followed by post hock Dunnett's test
P <0.05 compared to positive control

Castor oil induced diarrhea in negative control group of rats as there was significant (P <0.05) increase in fluid content and weight of feces compared to positive control . administered to induce diarrhea. Treatment with extracts inhibited castor oil induced diarrhea in rats as there was no significant increase in fecal parameters compared to positive control.There was no difference between the doses and extracts with respect to potency as shown in table no 4.

## V. DISCUSSION AND CONCLUSION

There are many causes of constipation – lifestylechoices, medications, medical conditions, and pregnancy. Many herbal infusions are used to resolve constipation.one such infusion is karikaali vellam . In Kerala,water infused with Caesalpinia

International Journal of Pharmaceutical Research and Applications
Volume 7, Issue 6 Nov-Dec 2022, pp: 64-69 www.ijprajournal.com ISSN: 2456-4494

sappan wood is used for drinking purpose, forhaving curing actions of wounds,rheumatism, diarrhoea ,laxative and allergic reactions .Since it lacks scientific evidence, in our study aqueous and ethanolic extracts of Caesalpinia sappan wood was evaluated for laxative activity in rats . The doses of Caesalpinia sappan wood extracts was selected according to the previous studiesconducted.Drug induced and , low fiber induced constipation in rats andcastor oil induced enter pooling in rats were used as screening models for evaluation of laxative activity.Loperamide is well known to stimulate the extension of stool evacuation time and to delay intestinal luminal transit through the inhibition of water secretion , as well as smooth movement in the intestinal wall . Furthermore, Loperamide has been used to induce constipation in a variety of studies to determine the cause of constipation. Mechanism of Loperamide is , it binds to the opiate receptor in the gut wall**.** Consequently, it inhibits the release of acetylcholine and prostaglandins, thereby reducing peristalsis, and increasing intestinal transit time. Loperamide increases the tone of the anal sphincter, thereby reducing incontinence and urgency.

In the present study, the laxative effects of Caesalpinia sappan was evaluated based on changes in fecal parameters (numbers, weight ) , asignificantdecrease in fecal discharge(fecal pellet number and weight) was observed during constipation induced by loperamide treatment. Treatment with extracts of Caesalpinia sappan wood except aqueous 30 mg/kg, significantly ( $P<0.01$, $p<0.001$) increased fecal pellet number and weight, indicating dose dependent laxative property. Ethanolic extract was found to be more potent than aqueous extractand comparable with standarddrug Gaviscon(Table 2) .The effect of loperamide induced constipation was reverted bythe extracts.The anthraquinone glycoside present in the Caesalpinia sappan is responsiblefor this action ,by increasing the tone of thesmooth muscle in wall of the large intestine.

Low fibre diets increase cecal retention time, leading to hypomotility of the gut, and reduces the amount of cecotrophs produced. Indigestible fibers like cellulose and lignin (crude fiber digestibility of 15%) are the best way of preventing enteritis as they stimulate most hindgut motility.Maintaining a low-fiber diet significantly decreased stool frequency, weight, and water content and significantly delays carmine egestion.

In Lowfibre diet induced constipation model, treatment with extracts significantly ($P<0.001$) increased the weight of the faecal matter compared to negative control indicating laxative property. There was no difference between the doses and extracts with respect to potency as shown in table no 3.Chemical constituents such as sappanchalcone , anthraquinone glycosides present in Caesalpinia sappan has decreased faecal retention time and increased motility of the gut .

Castor oil is one of the oldest drugs. When given orally, it has a laxative effect and induces labor in pregnant females. The effects of castor oil are mediated by ricinoleic acid, a hydroxylated fatty acid released from castor oil by intestinal lipases. Despite the wide-spread use of castor oil in conventional and folk medicine, the molecular mechanism by which ricinoleic acid acts remains unknown. Here we show that the $EP_3$ prostanoid receptor is specifically activated by ricinoleic acid and that it mediates the pharmacological effects of castor oil. In the present study,2ml /kg b.w.p.o, of castor oil, induceddiarrhea in negative control group of rats as there was significant ($P<0.05$) increase in fluid content and weight of feces compared to positive control.Treatment with extracts inhibited castor oil induced diarrhea in rats as there was no significant increase in fecal parameters compared to positive control (table no 4). This shows that extracts at doses used are not precipitating diarrhoea. This can be advantage in chronic treatment of constipation as some of the medications can cause diarrhoea.The phytochemical constituentsuchas anthraquinones, sappanchalcones, glycosides present in Caesalpinia sappanwoodare responsible for laxative activity**.** Similar phytochemical constituentspresent in herbssuch as Alexandrian senna, rhubarb, and hybrids, aloe, cascara, alder buckthorn, glossy buckthornetc have shown to possess laxative activity.Anthraquinones are organic compounds, in the form of simple anthrones or bianthrones.The free anthraquinone aglycones exhibit little therapeutic activity. The sugar residue facilitates absorption and translocation of the aglycone to the site of action. The anthraquinone and related glycosides are stimulant cathartics and exert their action by increasing the tone of thesmooth muscle in wall of the large intestine. The glycosides are absorbed from the small intestine and re excreted in the large intestine where they cause irritation of colon mucosa increases the motility to produce laxative effect. In conclusion, wood extracts of Caesalpinia sappanat doses administered, showed a laxative effect in rats.Ethanolic extract was found to be more potent, and exhibited dose depended laxative activity only in drug induced constipation model , all extracts were equally effective in low

International Journal of Pharmaceutical Research and Applications
Volume 7, Issue 6 Nov-Dec 2022, pp: 64-69 www.ijprajournal.com ISSN: 2456-4494

fiber diet models. Extracts also inhibited castor oil induced diarrhea,Thus it is demonstrated that wood extracts of Caesalpiniasappan administration can cure constipation without causing diarrhea . Results of the study can be extrapolated to humans,thus substantiates the use of Caesalpinia sappanwood infusion in parts of India (kerala)for constipation.

**SOME OF THE ADVANAGES FROM THE ABOVE RESULTS**

a)Caesalpinia sappan can be used to treat constipation in rats.

b)Caesalpinia sappn relives constipation without inducing diarrhea.

## REFERENCES

[1]. Nirmal NP, Rajput MS, Prasad RG, Ahmad M. Brazilin from Caesalpinia sappan heartwood and its pharmacological activities: A review. Asian Pacific Journal of Tropical Medicine. 2015 Jun 1;8(6):421-30.

[2]. DWIVEDI N, NAIR RA, INDIRADEVI A. Traditional Ayurvedic Herbal Drinking Water to Cure Various Diseases in Kerala (India).

[3]. Klaschik E, Nauck F, Ostgathe C. Constipation—modern laxative therapy. Supportive care in cancer. 2003 Nov;11(11):679-85.

[4]. Cai C, Zhao M, Tang L, Tu P. Study on chemical constituents from heartwood of Caesalpinia sappan. Zhongcaoyao= Chinese Traditional and Herbal Drugs.

[5]. Branch RL, Butt TF. Drug-induced constipation. Adverse Drug Reaction Bulletin. 2009 Aug 1(257):987-90.

[6]. Kojima R, Doihara H, Nozawa K, Kawabata-Shoda E, Yokoyama T, Ito H. Characterization of two models of drug-induced constipation in mice and evaluation of mustard oil in these models. Pharmacology.

[7]. Tunaru S, Althoff TF, Nüsing RM, Diener M, Offermanns S. Castor oil induces laxation and uterus contraction via ricinoleic acid activating prostaglandin EP3 receptors. Proceedings of the National Academy of Sciences. 2012 Jun 5;109(23):9179-84.

[8]. Makizaki Y, Maeda A, Oikawa Y, Tamura S, Tanaka Y, Nakajima S, Yamamura H. Alleviation of low-fiber diet-induced constipation by probiotic Bifidobacterium bifidum G9-1 is based on correction of gut microbiota dysbiosis. Bioscience of microbiota, food and health. 2018:18020.