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J. Nat. Prod. Plant Resour., 2012, 2 (3):360-367  
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ISSN : 2231 – 3184  
CODEN (USA): JNPPB7

### The effect of methanol leaves extract of *Ficus Glumosa* on gastrointestinal motility and on castor oil induced diarrhea in laboratory animals

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#### ABSTRACT

*Ficus glumosa* belongs to the family moraceae and the genus ficus. This study was aimed at investigating the methanol extract of the stem bark of *Ficus glumosa* for antidiarrhoeal activity using castor oil induced diarrhoea in mice. The effects of this extract on perfused isolated rabbit ileum were also evaluated. The methanol extract produced a dependent relaxation of the rabbit ileum. The extract protected the mice against diarrhoea the dose of 20mg/kg bodyweight produced the highest protection (60%). Also as regards to the other two doses (40 and 80 mg/kg) produced 40 and 20 % protections respectively. The acute toxicity test revealed the median lethal dose (LD<sub>50</sub>) values for the extract is found to be 2.154 mg/kg bodyweight. These results obtained revealed that the methanol leaves extract possess pharmacological activity against diarrhoea and may possibly explain the use of the plant in traditional medicine.

**Key words:** *Ficus glumosa*, Methanol, Diarrhoea, Rabbit ileum, Castrol oil.

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#### INTRODUCTION

Diarrhea is an alternation in the normal pattern of defecation that usually results in the increased frequency, volume and softness of feces. It is characterized by the passage of watery and loose stools. It may be acute or chronic depending on the duration and response to treatment<sup>1</sup>. The use of plants as medicine is an ancient practice common to all societies especially the African society. This practice continues to exist in the developing nations. It is on this basis that researchers keep on working on medicinal plants in order to produce/develop the best for physiological uses as medicines<sup>2</sup>.

*Ficus glumosa* is a small- to medium-sized tree, 5-10 m tall, or it may Become a large tree reaching 24 m and 2 m in girth. Branches widely Spreading, more or less horizontal, often supported by stilt roots. Bark Local names: Afrikaans (Afrikaanse rotsvy, Afrikaans rostvy); English (African rock fig); Hausa (kawuri); Somali (berde); Tigrigna (chekomte, check lang)

*Ficus glumosa* occurs on rocky outcrops, where it splits rocks; along dry water courses or in open country; frequently in valleys, where it reaches its greatest size. The species also occurs in fringe forest in savannah areas, especially in swampy ground, and in swamp forest in coastal areas. Originally collected in Ethiopia, it occurs in many parts of tropical Africa and is typically found in dry country in wooded grassland and bush.

The aim of this experiment is to investigate the effect of methanol leaves extract of *Ficus glumosa* on gastrointestinal motility and on castor oil induced diarrhea in rodents.

## MATERIALS AND METHODS

### COLLECTION AND IDENTIFICATION OF PLANT MATERIAL

*Ficus glumosa* plant stem bark was collected from Samaru-Zaria in the Month of May, 2011 and was authenticated by A.U. Gallah of the Biological Sciences Department, Ahmadu Bello University, Zaria, Nigeria where a voucher specimen (No.0662) was deposited.

### Preparation of plant materials

The *Ficus glumosa* was air dried under shade and then ground to powder. 3 kg of *Ficus glumosa* was macerated with methanol (70%) 48 hours. It was filtered using filter paper, beaker and funnel. The filtrate was then transferred to an evaporating dish and was evaporated using water bath. The extract was obtained after evaporation from the water bath and refrigerated till the day of the experiment. Solutions of the extracts were prepared freshly for each study.

### Animals

A rabbit (1 kg) and 37 Swiss albino mice weighed between 18- 22 g were used for the study. The animals were maintained in the Animal House Facility of the Department of Human physiology, Ahmadu Bello University, Zaria, Nigeria. The animals were fed on standard small animal feeds (Excel feed, Ilorin, Nigeria) and water *ad libitum*. This research was carried out in Ahmadu Bello University in accordance with the rules governing the use of laboratory animals as accepted internationally.

### Drugs

All chemicals and drugs were obtained commercially and were of analytical grade.

### Preliminary phytochemical screening

The extract was subjected to preliminary phytochemical analysis using standard protocol <sup>3</sup>.

### Acute toxicity study

The method described by <sup>4</sup> was adopted. Briefly, 12 mice were used for each extract. In the first phase, three doses of the methanol leaves extract (10, 100 and 1000 mg/kg were administered to three groups each containing three mice). In the second phase, more specific doses 1600, 2900 and 5000mg/kg were administered to four groups each containing one mouse. The median Lethal dose (LD50) was determined as the geometric mean of the highest non lethal dose and the lowest lethal dose of which there is 1/1 and 0/1 survival.

### Experimental designs

#### (a) Effects on isolated rabbit jejunum

The rabbit was sacrificed by cervical dislocation. Segments of the ileum, about 3.0 cm long were removed and dissected free of adhering mesentery. The intestinal contents were removed by flushing with Tyrode solution of the following compositions in millimoles (mM): NaCl, 136.8; KCl, 2.7; CaCl, 1.3; NaHCO<sub>3</sub>, 12.0; MgCl, 0.5; NaPO<sub>4</sub>, 0.14; glucose, 5.5. The tissue was mounted in a 25 ml organ bath containing Tyrode solution maintained at 35°C and aerated with air. An initial tension of 0.5 g was applied to the segments and 60 min equilibration period was allowed while the physiological solution was changed every 15 min. At the end of the equilibration period, the effect of acetylcholine, adrenaline and methanol extract of leaves was investigated. The contact time for each concentration was 1 minute which was followed by washing three times. The tissue was allowed a resting period of 15 min before the next addition.

#### (b) Effects of castor oil-induced diarrhoea on mice

The mice were fasted for 12 h prior to the commencement of the study and were randomly divided into five groups each containing five mice. The grouping is as follows:  
30 min post treatment, castor oil (0.2 ml / mouse) was administered to all the groups. All treatments were given intragastrically. The animals were then placed in individual cages on a clean Whatmann filter paper size 1. Four hours after the castor oil administration, the cages were inspected for the presence of characteristic diarrhoeal droppings; absence of which was regarded as protection <sup>5</sup>.

**Statistical Analysis**

The results was analyzed by determine the percentage protection against diarrhea versus the control.

**RESULTS****Acute toxicity studies**

<b>Phase 1</b>	
Dose	No of mice/death
10mg/kg	0/3
100mg/kg	0/3
1000mg/kg	0/3
<b>Phase 2</b>	
Dose	No of mice/death
1600mg/kg	0/3
2900mg/kg	1/3
5000mg/kg	1/3
$LD_{50} = \sqrt{1600 \times 2900} = 2154 \text{mg/kg}$	

The acute toxicity studies, phase 1 shows no lethal dose for 10, 100, and 1000mg/kg of methanolic leaves extract of *Ficus glumosa*. While in the phase 2, 1600, 2900 and 5000mg/kg were administered. 1600mg/kg had no lethal dose but 2900 and 5000mg/kg bodyweight both had lethal doses where the mice in each group died. The lowest lethal dose is 2900mg/kg and the highest non-lethal dose is 1600mg/kg, thus the LD50 = 2,154 mg/kg bodyweight

**Table : Phytochemical constituents of the methanol extract of *Ficus glumosa*.**

Phytochemical constituents	inference
Flavonoids	+
Saponins	+
Carbohydrate	—
Cardiac glycosides	+
Tannins	+
Triterpenes	+
Anthraquinone	—
Keys= + Presence - Absent	

**Table 2: Effect of methanol extract of . on castor oil *Ficus glumosa* induced diarrhea in mice**

Treatment	Dose (mg/kg)	No. of mice with diarrhoea	Protection (%)
Normal saline	10	5/5	0
Loperamide	5	0/5	100
<i>Ficus glumosa</i>	20	3/5	60
<i>Ficus glumosa</i>	40	2/5	40
<i>Ficus glumosa</i>	80	1/5	20

Mice administered with 20, 40 and 80mg/kg of extract of had diarrhea *Ficus glumosa* in 3/5, 2/5 and 1/5 respectively, (60,40 and 20% protection respectively).

The mice administered with loperamide, 5mg/kg as positive control after castor oil-induced diarrhea show no sign of diarrhea (100% protection) while the negative control normal saline group had diarrhea.

Figure 1: Effect of methanol leaves extract of (20mg/ml) *Ficus glumosa* on isolated rabbit ileum

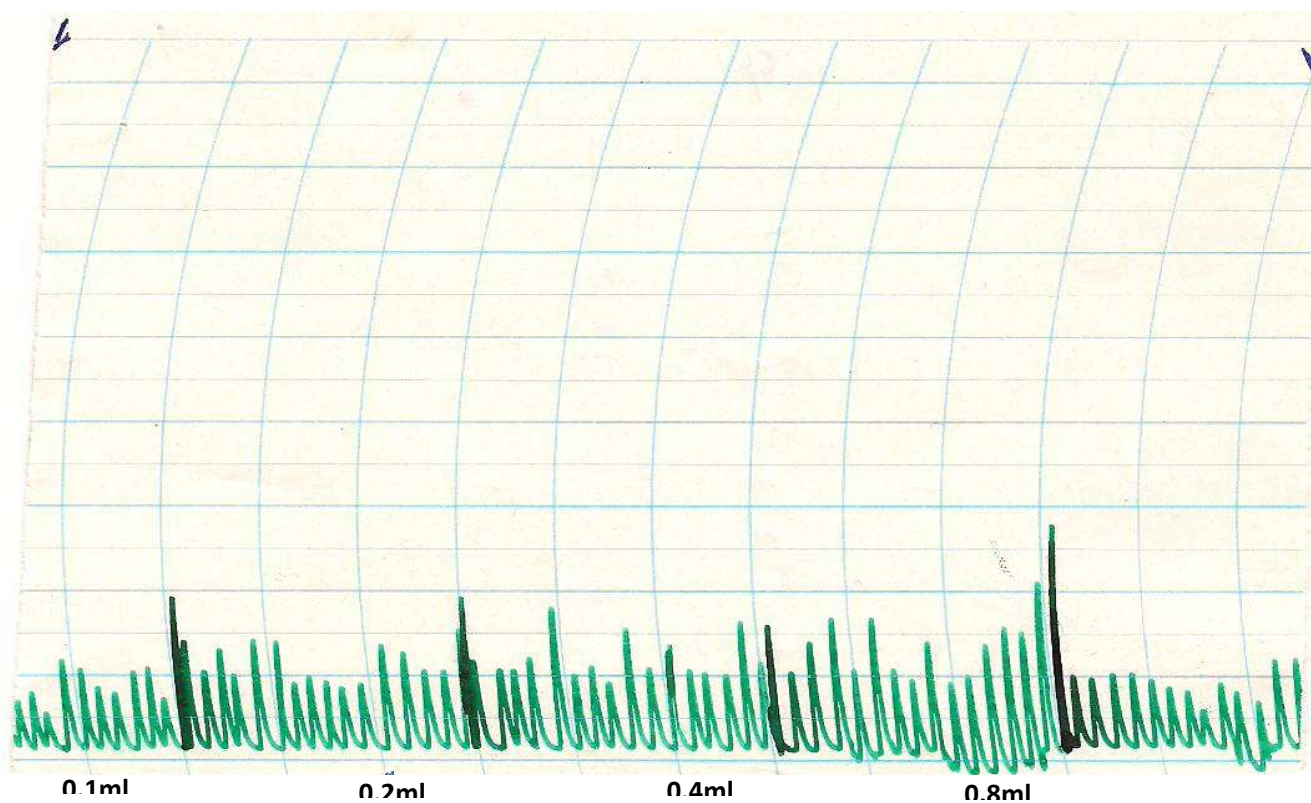


Figure 2: Effect of methanol leaves extract of *Ficus glumosa* (40mg/ml) on isolated rabbit ilium

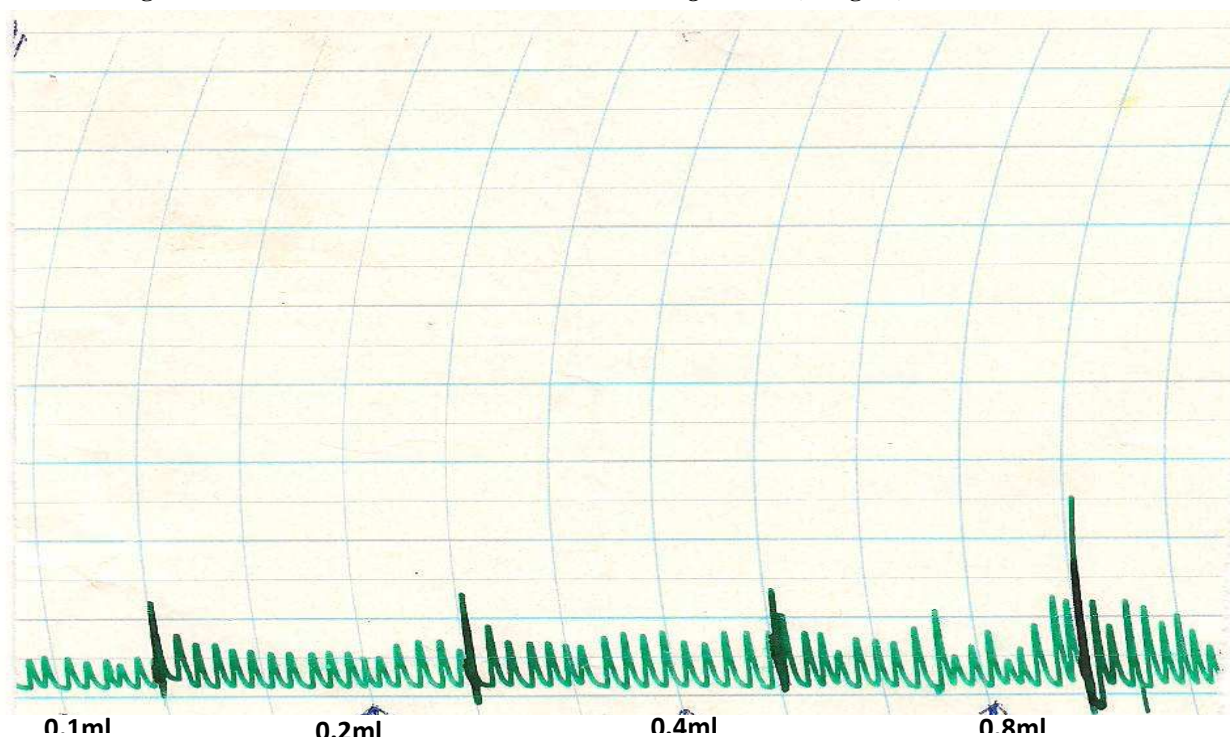




Figure 3: Effect of methanol leaves extract of *Ficus glumosa* (80mg/ml) on isolated rabbit ileum

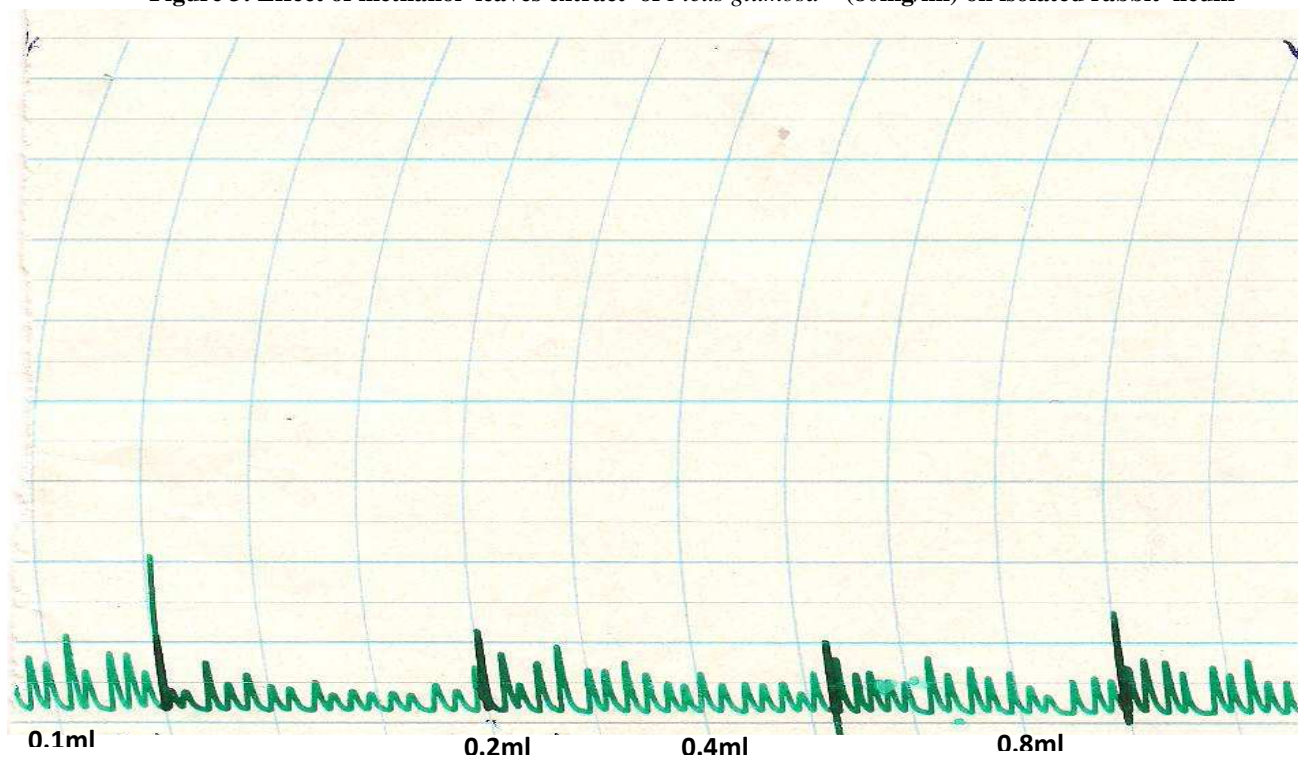
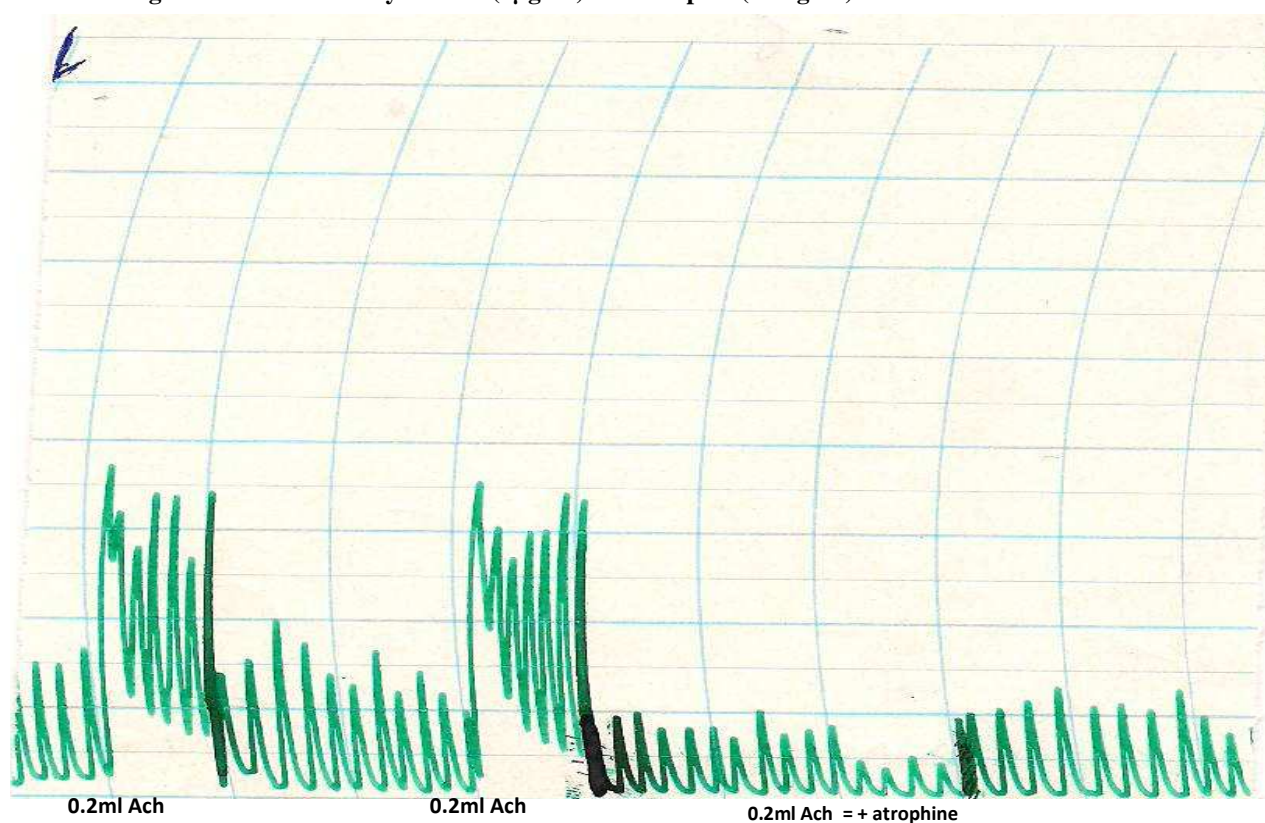
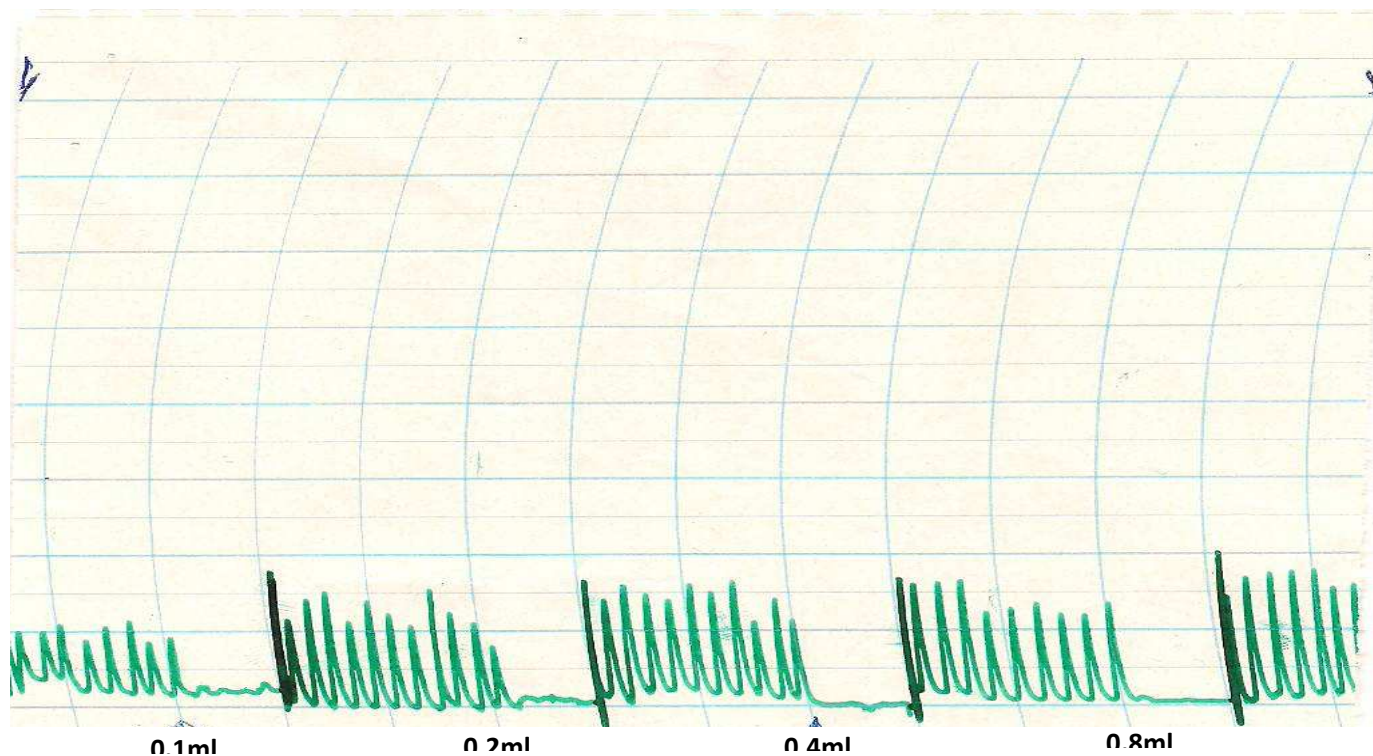


Figure 4: Effect of Acetylcholine (2 $\mu$ g/ml) and atropine (25mg/ml) on isolated rabbit ileum.

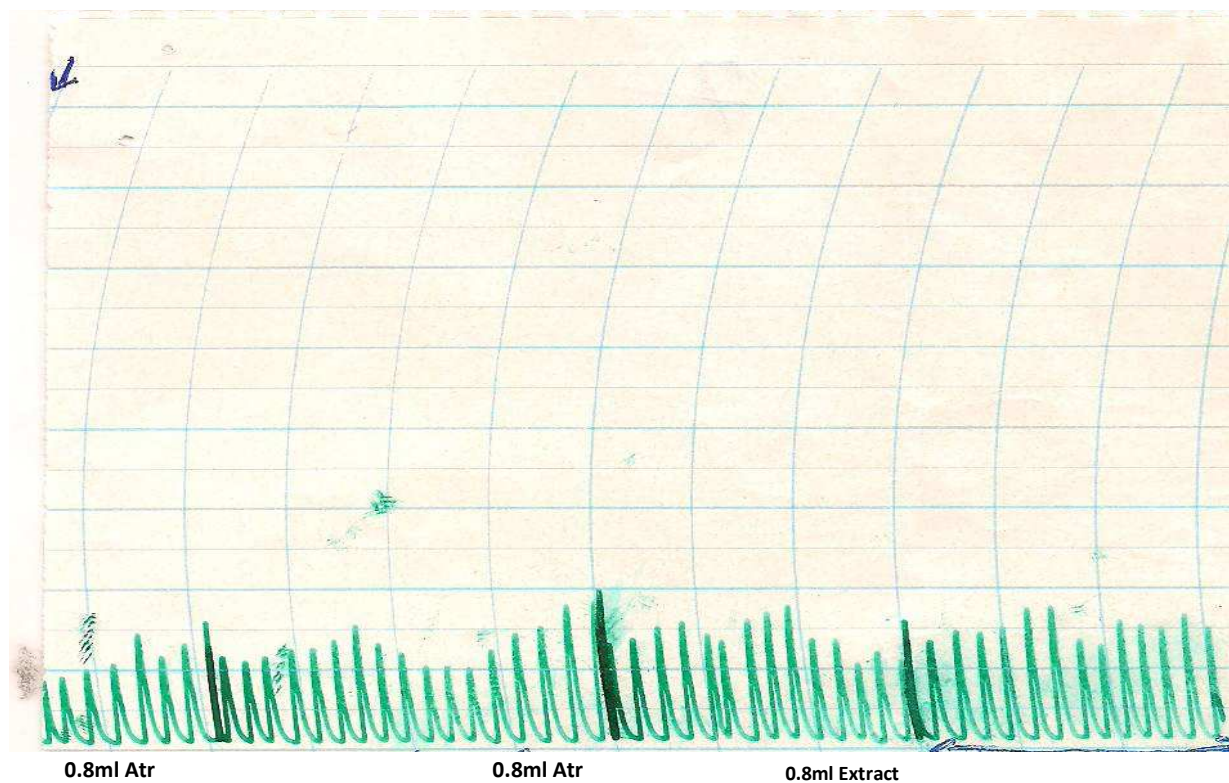


**Figure 5: Effect of Adrenaline (10 $\mu$ g/ml) on isolated rabbit ileum**



Drug and extract interactions on the rabbit jejunum. There was relaxation of the tissue .

**Figure 6: Effect of extract (80mg/ml) and atropine (Atr. 25mg/ml) on isolated rabbit ileum.**



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## DISCUSSION

Castor oil stimulates peristaltic activity in the small intestine, leading to changes in the intestinal mucosa. Its action also stimulates the release of prostaglandins.

Castor oil is a triglyceride in which approximately 90% of the fatty acid chains are ricinoleic acid. Oleic and linoleic acids are the other significant components. Castor oil causes diarrhea due to its active metabolite, ribonucleic acid<sup>6,7</sup>. Ricinoleic acid, a monosaturated 18-carbon fatty acid is unusual in that it has a hydroxyl functional group on the 12<sup>th</sup> carbon. This functional group causes ricinoleic acid (castor oil) to be unusually polar.<sup>8</sup>. *Ficus glumosa* significantly reduced the intestinal transit time as observed by the decrease in intestinal motility of the isolated rabbit ileum. The median lethal dose of the extract was 2,154mg/kg. Phytochemical screening revealed the presence of flavonoids, tanins, saponins, cardiac glycosides and triterpenes.<sup>9</sup>. Hence, tannins and triterpenes may be responsible for the mechanism of action of *Ficus glumosa* anti diarrheal activity.<sup>10</sup>.

The anti diarrheal activity of the extract may also be due to the presence of denatured proteins which form protein tannates. protein tannates makes the mucosa more resistant and hence, reduce secretion.<sup>11</sup>. This can be due the fact that the extract increased the reabsorption of water by decreasing intestinal motility in the isolated rabbit ileum. Loperamide, apart from regulating the gastrointestinal tract is also reported to slow down transit in the small intestine, reduce colon flow rate and consequently any effect on colonic motility<sup>12</sup>. The mice in the control group were given 10ml/kg bodyweight normal saline and all the mice in the group had stool with diarrhea which indicate there was no protection. While the mice that were given 5mg/kg bodyweight loperamide as positive control after castor oil-induced diarrhea showed no sign of diarrhea indicating 100% protection against Castrol oil induced diarrhea. In relation to the extract treated groups there was 60% protection against diarrhea in the groups treated with the doses of 20. In relation to the dose of 40 mg/kg body weight there was 40% protection. Also as regards to the group given the highest dose that is 80 mg/kg body weight there as 20 % protection against Castrol oil induced diarrhea.

In this experiment, the methanol extract of *Ficus glumosa* exhibited a significant anti diarrheal activity. Based on this observation, it is plausible in nature which may be suggested that the anti diarrheal effect of the extract may be due to the inhibition of prostaglandin biosynthesis. Flavonoids are known to modify the production of cyclooxygenase 1 and 2 (COX-1, COX-2) and lipooxygenase (LOX)<sup>11</sup>. Certain flavonoids inhibit inflammatory processes by inhibiting key enzymes involved in the synthesis of prostaglandins processes.

The effects of the plant extract on the rabbit ileum were dose related. The leaves extract relaxed spontaneous the rabbit ileum which is dose dependent.

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