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Medicinal Potential of *Phyllanthus emblica* (Linn.) Fruits Extracts: Biological and Pharmacological Activities

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Authors' contributions

This work was carried out in collaboration between all authors. Authors SMMH, RS, SM and NMAA designed the study, performed the statistical analysis, wrote the protocol and author SMMH wrote the first draft of the manuscript. All authors read and approved the final manuscript.

Original Research Article

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ABSTRACT

Phyllanthus emblica (Linn.) is a common plant and fruits very popular in Bangladesh. It is a common ingredient of many traditional and herbal medicines. The intention of the present study was to explore the scientific relation with the traditional use of the fruit of *Phyllanthus emblica* (Linn.). Antimicrobial screening, analgesic activity, antidiarrheal activity and the brine shrimp lethality test for cytotoxic activity screening are the selected pharmacological activities. Phytochemical analysis of ethanolic fruits extract confirms that the fruit contain flavonoids, alkaloids, tannin, steroids, reducing sugar and gum. Experimental screening confirms that the fruit extract produced 19.07% and 38.67% writhing inhibition at the oral dose of 250 and 500mg/kg-body weights respectively. That means ethanol extract of the fruit of *Phyllanthus emblica* (Linn.) has an analgesic property. The ethanolic fruit extract of *Phyllanthus emblica* (Linn.) also significantly inhibited ear edema formation in xylene induced ear edema, considered as direct evidence that supporting the anti-inflammatory activity of ethanolic fruit extract of *Phyllanthus emblica* (Linn.) The fruit extract ha a remedy for a different bacterial disease

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is supported by the antibacterial screening tests. During the antidiarrheal activity screening at the dose of 500mg/kg-body weight, *Phyllanthus emblica* (Linn.) showed a moderate antidiarrheal activity in castor oil induced test in mice and caused an increase in latent period i.e. delayed the onset of diarrheal episode and decreased the frequency of defecation. T-test of these responses showed that the result is significant throughout the observation period. The ethanolic fruits extract have cytotoxic activity and test sample showed different mortality rate at different concentrations. The LC_{50} values were found to be 60µg/ml for the crude extract. The 90% mortality (LC_{90}) values were 100µg/ml respectively. According to the results of the present investigation, we can conclude that the ethanolic fruits extract of *Phyllanthus emblica* (Linn.) has significant analgesic, anti-inflammatory, antimicrobial, anti-diarrheal and cytotoxic effects. This study also suggests us to isolate the active compound(s) responsible for those pharmacological properties.

Keywords: Phyllanthus emblica (Linn.); antimicrobial; analgesic; antidiarrheal; anti Inflammatory; Bioassay.

1. INTRODUCTION

Phyllanthus emblica (Linn.) or *Emblica officinalis* (Gaertn) belongs to the Family Euphorbiaceae. Bengali/vernacular names are Amloki, Amla; Aila (Sylhet) and tribal names are Ambari (Garo); Amloti (Chakma); Soi sha (Marma); Sowan Lu (Bawm); Khulu (Murong). English names are Emblic Myrobalan, Indian Gooseberry [1]. Fruit a globose drupe, about 2.5 cm across, obscurely 6-lobed. Occurs in the dry forests of Chittagong, Chittagong Hill Tracts, Cox's Bazar, Sylhet, Dhaka-Tangail (Sal forest) and Dinajpur; also cultivated elsewhere [1].

Fruits are diuretic, refrigerant, carminative, astringent, tonic, stomachic, laxative, antacid and rich in Vitamin C, improves appetite, useful in vomiting and burning urination, diseases of the heart and liver, piles, stops nasal haemorrhage. It promotes children's resistance to cough and cold; used as a hair tonic. Dried fruits are useful in haemorrhoids, diarrhoea, dysentery, anemia, jaundice and dyspepsia. The fruits are also said to be beneficial in insomnia, skin problems, gall pain, leucorrhoea and tympanites. Sherbet prepared from the fruit along with lemon juice is used for arresting acute bacillary dysentery. Fruits are a valuable component of "Trifala" used in different Ayurvedic preparations. Flowers are cooling and aperient. Bark is astringent [1]. Ethanolic extract of the leaves possess good antibacterial properties and mild antifungal properties [2,3]. Phyllambin isolated from fruit potentiates pharmacological action of adrenaline, has mild depressant action on central nervous system and possesses spasmolytic action [1]. Water extract of the fruit causes moderate relaxation of the isolated guinea-pig ileum, but it does not interact with the activity of Acetylchloline [4].

Fruit is a rich natural source of vitamin C. It also contains tannins and colloidal substances, phyllembic acid, lipids, gallic acid, ellagic acid, trigalloylglucose, terchebin, corilagin and emblicol. Phyllembin and mucic acid have been isolated from the fruit pulp. Seeds contain fixed oil, phosphatides, tannins and essential oil. Bark, fruits and leaves are rich in tannin. They also contain lupeol, β-sitosterol and ellagic acid. Bark also contains leucodelphinidin. Seed oil also contains linoleic acid (64.8%), closely resembled linseed oil [1,5,6].

The selected pharmacological investigations are antimicrobial screening, evaluation of analgesic activity, antidiarrhoeal activity screening and the brine shrimp lethality test by

using the ethanolic fruit extract of *Phyllanthus emblica* (Linn.). The present study investigates the phytoconstituents in order to correlate the folkloric claims with the bioactive compounds present in the plant and investigates the cytotoxicity potential of the plant on brine shrimp (*Artemia salina*) larvae. Since toxicological evaluation of plant extracts seeks to determine its possible collateral effects to ensure the safety of its use, brine shrimp larvae being sensitive to toxic substances are commonly used for toxicity assays in pharmacology [7,8].

2. MATERIALS AND METHODS

2.1 Plant Material Collection and Identification

Fruits of *Phyllanthus emblica* (Linn.) were collected from the district of Chittagong and were identified by the experts and preserved in the herbarium, department of botany, University of Chittagong. (Acc. No. SBU1521).

2.2 Plant Extracts Preparation

The fruit were collected and clean to separate from undesirable plant parts. They were air-dried for one week. The fruits were milled into a coarse powder by using laboratory milling equipment. Cold extraction was performed during extract preparation. About 350 gm of powered material was taken in a clean, flat bottomed glass container and 900 ml of 80% ethanol was added in container. The container with its contents was sealed and kept for a period of 10 days accompanying occasional shaking and stirring to enhance the efficient extraction. The whole mixture then filtered through a piece of clean, white cotton and finally it was filtered through Whatmann filter paper [Bibby RE200, Sterilin Ltd., UK]. The filtrate (ethanol extract) obtained was evaporated under ceiling fan and on a water- bath until dried and % yield of fruit calculated 4.57% [9]. The extract was stored at 4ºC in refrigerator until use.

2.3 Experimental Animals

Young Swiss-albino mice aged 4-5 weeks old and average weight 20-25 gm was employed for the experiment. The mice were purchased from the Animal Research Branch of the International Centre for Diarrhoeal Disease and Research, Bangladesh (ICDDR,B). They were kept in standard environmental condition (RH 55% to 60%, room temperature 25±2ºC and 12 h light/ dark cycle) for one week for adaptation after their purchase and fed ICDDR,B formulated rodent food and water. The experimental study was performed under the guidelines of Institutional Animal Ethics Committee [10].

2.4 Chemicals and Drugs

Drug Diclofenac Sodium, Kanamycin, Loperamide were from square pharmaceuticals limited and Vincristine Sulphate (VINCRIRST ®) from Techno Drugs Ltd. Used other reagents and analytical kits were laboratory reagent grade and from Merck Specialities Private Limited, India.

2.5 Phytochemical Screening

Different preliminary phytochemical tests were performed to identify different phytochemical constituents in the extract as well as powdered drug following standard methods as described [11-14].

2.6 Analgesic Activity

Acetic acid induced writhing in mice model was employed for screening analgesic activity of the ethanolic fruit extract of *Phyllanthus emblica* (Linn.) [15-20]. Young Swiss-albino mice, the experimental animals were randomly selected and divided into four groups denoted as group-I, group-II, group III and group-IV, consisting of 5 mice in each group. Group I received 1% tween 80 in water (10 ml/kg) as control, group II received standard drug Diclofenac sodium (25 mg/kg) while group III and IV received 250 mg/kg and 500 mg/kg extract of sample respectively. Each mouse was weighed properly and the dose of the test samples and control materials were adjusted accordingly. Test samples and diclofenac sodium were given orally by means of a feeding needle and the control was given intraperitoneally. After 30 minutes, the writhing inducing chemical, acetic acid solution (0.7%, 10 ml/kg) was administered intraperitoneally to each of the animals group. After an interval of five minutes the number of squirms (writhing) was counted for 15 minutes. The mice didn't always perform full writhing. The incomplete writhing was taken as a half writhing, so two half writhing were taken as one full writhing.

2.7 Anti-inflammatory Activity

To determine the anti-inflammatory activity of the plant extract Xylene induced air edema in mice model was employed and the method was described by Dev et al. [21]. Before experiment all the experimental mice were divided into four groups and treated with different agents like group I received 1% tween 80 in water (10 ml/kg) as control, group II received standard drug diclofenac sodium (25 mg/kg) while group III and IV received 250 mg/kg and 500 mg/kg extract respectively. One hour after administration of the above dose, 0.01 ml of xylene was injected to the anterior and posterior surfaces of the right ear of the each mouse. After one hour of xylene injection, all mice were sacrificed. Both treated and untreated ears of mice were cut downed by using a 7mm diameter cork borer as circular sections and weighed. The weight difference between untreated and treated ear sections was calculated [19-22].

2.8 Antimicrobial Activity

Antimicrobial screening was performed using disc-diffusion method. Sample disc 500 µg/disc and Standard Kanamycin (30 µg/disc) discs were used as positive control and blank discs were used as negative controls. The sample discs, standard antibiotic discs and control discs were placed gently on marked zones in the agar plate's pre-inoculated with test bacteria, protozoa and fungi. The plates were then kept in a refrigerator at 4ºC for about 24 hours to allow sufficient diffusion of materials from discs to surrounding agar medium. The plates were then inverted and kept in an incubator at 37ºC for 24 hours. All organisms used in experiment are listed in Table 1. Antibacterial activity of the crude ethanolic extract was determined by disc diffusion method [23-25]. The selected organisms were gram negative bacteria were *Salmonella typi, Shigella dysenteriae, Shigella sonnei, Vibrio cholera, Hafnia, Plesiomonas* and gram positive bacteria were *Staphylococcus aureus, Staphylococcus*

epidermis, Staphylococcus saprophyticus, Staphylococcus pyogenas and fungi (*Candida albicans, Fusarium solanii*).

2.9 Antidiarrheal Activity

Castor oil induced diarrheal model was followed for this experiment. The employed mice were screened initially by giving 0.3 ml of castor oil and only those showing diarrhea were selected for the final experiment. The test animals were selected randomly and divided into three groups and each group having five mice. They were accurately weighed & properly marked of the experimental groups. As group-I or the control received only distilled water containing 1% Tween-80 (10 ml/kg). Group-II or the positive control received standard anti-motility drug, Loperamide (3mg/kg) as oral suspension. The test group III was treated with suspension of fruits extract of *Phyllanthus emblica* (Linn.) at the oral dose of 500 mg/kg-body weight. The method, described by Chatterjee was followed for this study [26]. Test samples, control and Loperamide were given orally by means of a feeding needle. Castor oil dose of 0.5ml per mouse was administered to all mice and after 1 hour of administration, the mice were fed with the samples, control and Loperamide. Individual animals of each group were placed in separate cages with adsorbent paper beneath the case and examined for the presence of diarrhea every hour in four hours study after the castor oil administration. Stool or fluids from mouse are the sign of induction diarrhea. Number of stools or any fluid material that stained the adsorbent paper were counted at each successive hour and noted for each mouse. The latent period of each mouse also counted. At the beginning of each hour old papers were replaced for the new ones. During an observation period, the total number of faecal output including diarrheic faces excreted by the animals was recorded. A numerical score based on stool consistency was assigned as follows: normal stool=1 and watery stool=2 [27-29].

2.10 Brine Shrimp Lethality Bioassay

This bioassay indicates cytotoxicity as well as a wide range of pharmacological activities such as antimicrobial, anti-viral, pesticidal and anti-tumor etc. of the compounds. For cytotoxic activity of the ethanolic extracts of *Phyllanthus emblica* (Linn.) stock solution was prepared having a concentration of 5µg/µl. Simulated sea water was prepared by dissolving 38gm of sodium chloride per liter of distilled water and filtered. This simulated sea water was used for a hatching of brine shrimp. Test solutions of different concentration (5, 10, 20, 40, 60, 80 and 160 µg/ml) were prepared from the stock solution of plant extract and living shrimps were kept to each of the solution. After 24 hrs the test tubes were observed and the number of survived nauplii in each test tube was counted and the results were noted. From this, the percentage of lethality of brine shrimp nauplii was calculated at each concentration for each sample [30-34].

$$\% \text{ mortality} = \frac{\text{no. of dead nauplii}}{\text{initial no. of live nauplii}} \times 100\%$$

3. RESULTS

3.1 Phytochemical Screening

Phytochemical analysis of *Phyllanthus emblica* (Linn.) showed the presence of flavonoids, alkaloids, tannin, steroids, reducing sugar and gum. The results were given in Table 1.

Table 1. Test result for chemical groups of *Phyllanthus emblica* (Linn.)

S/no.	Compound groups	Present/absent
1	Flavonoid	+
2	Alkaloid	+
3	Saponin	-
4	Tannin	+
5	Steroid	+
6	Reducing Sugar	+
7	Gum	+

EE =Ethanolic Extract, + = Present, - = Absent

3.2 Analgesic Activity

Effect of ethanolic fruits extract of *Phyllanthus emblica* (Linn.) on acetic acid induced writhing test on mice model showed mild analgesic activity. The summery of study design, results are given on Table 2.

Table 2. Analgesic activity of *Phyllanthus emblica* (Linn.) on acetic acid induction

Animal group and treatment	Writhing count	%Writhing	%Writhing inhibition	(t-test) p values
Group-I 1% tween-80 solution in water	36.2 ± 2.035	100	--	--
Group-II Diclofenac sodium 25 mg/kg	7.8 ± 1.113	21.55	78.45	(12.243)P<0.001
Group-III Extract (250 mg/kg)	29.3± 1.21	80.93	19.07	(5.87) P<0.01
Group-IV Extract (500 mg/kg)	22.2 ± 2.395	61.33	38.67	(4.454)P<0.01

Values are expressed as mean ± SEM, SEM= Standard error of mean, n=No. of mice, %=Percentage

3.3 Anti-inflammatory Activity

All the results of anti-inflammatory effect of ethanolic fruits extract of *Phyllanthus emblica* (Linn.) on xylene induced ear edema model were summarized on Table 3.

Table 3. Anti-inflammatory effect of ethanolic fruits extract of *Phyllanthus emblica* (Linn.) on xylene induced ear edema model

Group	n	Increased weight	Inhibition rate %
Blank (Xylene 0.01mL. injection)	5	10.5±0.22	00
Positive control (Diclofenac sodium 10mg/Kg)	5	6.5±0.23*	35
Test 1, Extract (250 mg/kg)	5	7.25±0.25*	18
Test 2, Extract (500 mg/kg)	5	6.75±0.22*	32.5

** P<0.01, vs. blank control group*

3.4 Antimicrobial Activity

The fruits extract of *Phyllanthus emblica* (Linn.) showed mild antimicrobial activity and may be a remedy for different bacterial diseases is supported by the antibacterial and antifungal screening tests. Antimicrobial activity results are on Table 4.

Table 4. *In vitro* antimicrobial activity of ethanolic fruit extract of *Phyllanthus emblica* (Linn.) by following disc diffusion method

Strains	Diameter of zone of inhibition in mm	
	Ethanol extract (500 µg/disc)	Kanamycin (30 µg/disc)
Bacterial gram negative		
Salmonella typhi	9.3	25.5
Vibrio cholerae	R*	24.3
Shigella dysenteriae	7.1	28
Plesiomonas	R*	27.1
Sheigella sonnie	10.24	25
Hafnia	8.4	29.6
Bacterial gram positive		
Bacillus subtilis	11.2	25.6
Bacillus megaterium	8.5	24.8
Staphylococcus aureus	R*	30
Staphylococcus epidermis	7.1	22.2
Staphylococcus saprophyticus	R*	23.8
Staphylococcus pyogenas	9.7	29.4
Fungal strain		
Candida albicans	11.0	27.35
Fusarium solanii	9.5	26.8

R= Resistant or No growth

The fruit of *Phyllanthus emblica* (Linn.) performing as a remedy for different bacterial diseases is supported by the antibacterial screening tests.

3.5 Antidiarrheal Activity

Phyllanthus emblica (Linn.) showed a moderate antidiarrheal activity in castor oil induced test in mice model. Antidiarrheal activity results are summarized in Table 5 and Fig. 1.

Table 5. Effect of *Phyllanthus emblica* (Linn.) on castor oil induced diarrhea in mice

Groups	Period of study (hr)	Mean latent Period±S.E.	Mean no. of stools	S.E.	t-test (p value)
Control	C1	0.399 ±0.0396	13.2	0.583	--
	C2		14.4	0.678	--
	C3		9	0.447	--
	C4		6.6	1.077	--
Positive Control (Loperamide)	P1	0.929 ±0.0475	7	1	5.356[a]
	P2		8.4	0.748	5.887[a]
	P3		5.2	2.154	1.727[c]
	P4		3.4	1.777	1.540[c]
Extract of *Phyllanthus emblica* Linn. 500mg/kg	E1	0.497 ±0.0419	8.2	1.281	3.5549[b]
	E2		9.6	1.077	3.7605[b]
	E3		7.8	0.860	1.5328[c]
	E4		4.4	0.696	1.7156[c]

Values are t-test. (n=5), [c]p< 0.2, [a] p< 0.001, [b] p< 0.01 vs. Control. Student's t-test

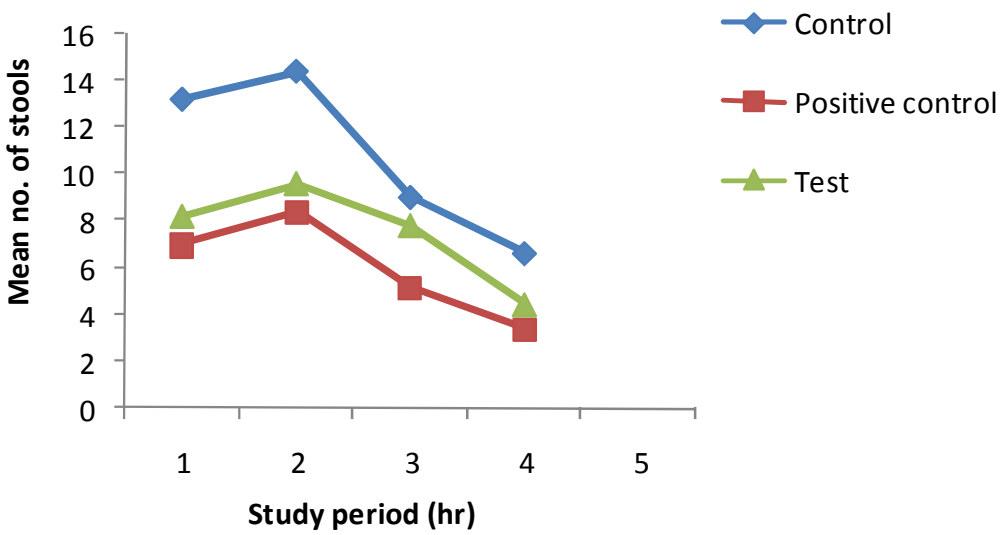

Fig. 1. Effect of Loperamide and *Phyllanthus emblica* (Linn.) on castor oil induced diarrhoea in mice through the observation period (4hr)

3.6 Cytotoxic Activity

The mortality rate of brine shrimp increased with the increase in concentration of the sample and plot of percent mortality versus log concentration on the graph paper produced an approximate linear correlation between them. From the graph Fig. 2 the concentrations at which 50% mortality (LC_{50}) of brine shrimp nauplii occurred were obtained by extrapolation. The values were found to be 60µg/ml for the crude extract. The 90% mortality (LC_{90}) values were 100µg/ml respectively. Experimental results are as table 6A, 6B and figure 2.

Table 6A. Result of Brine shrimp lethality bioassay of ethanolic fruit extract of *Phyllanthus emblica* (Linn.)

Test sample	Conc. (µg/ml)	Log (Conc.)	No. of alive shrimp	% mortality	LC_{50} (µg/ml)	LC_{90} (µg/ml)
Ethanolic fruits extract of *Phyllanthus emblica* (Linn.)	20	1.30	6	40		
	40	1.60	6	40		
	60	1.77	5	50		
	80	1.90	3	70	60	100
	100	2	1	90		
	120	2.07	2	80		
	140	2.14	0	100		

Table 6B. Test significance analysis of brine shrimp lethality bioassay of ethanolic extract of fruit of *Phyllanthus emblica* (Linn.)

Ethanolic extract of *Phyllanthus emblica* (Linn.)		
Name of applied test	**Chi-value**	**Sig.**
Linear-by-Linear Association	5.44	0.02
Pearson's R	0.95	0.001

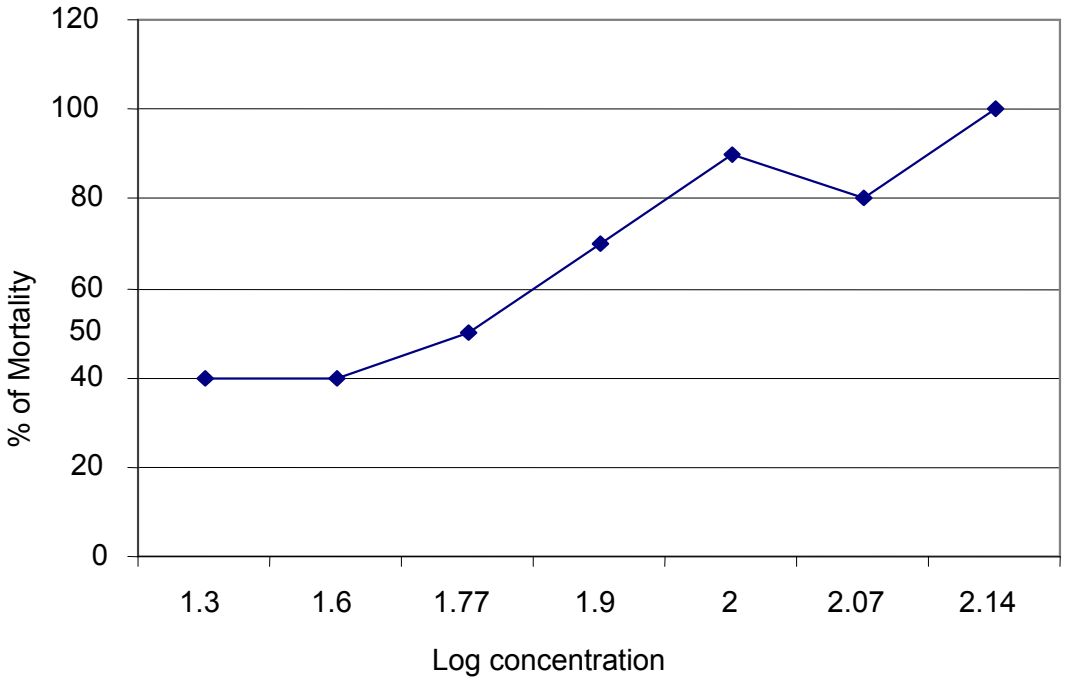

Fig. 2. LC_{50} and LC_{90} of *Phyllanthus emblica* (Linn.)

4. DISCUSSION

Preliminary phytochemical analysis of ethanolic fruit extract of *Phyllanthus emblica* (Linn.) confirms the presence of flavonoids, alkaloids, tannin, steroids, reducing sugar and gum. Further studies on isolation of its active components by using bioactivity guided approach may helps to determine the particular type of components. Present days it is very important for biologically active new components.

The ethanol extract of the fruit of *Phyllanthus emblica* (Linn.) was subjected to acetic acid induced writhing method in mice for preliminary analgesic activity screening. The results of analgesic activity screening Table 2 showed that the ethanol extract showed analgesic activity depending upon the nature of their active ingredients in the extracts. The fruit extract produced 19.07% and 38.67% protection or writhing inhibition at the oral dose of 250 and 500 mg/kg-body weights respectively. Acetic acid causes pain and localized inflammation by the action of prostaglandins production, mainly prostacyclines and prostaglandin-E (PG-E) and increasing the level of PGE2 and PGF2α in the peritoneal fluid [34-40]. Prostacyclines and prostaglandin-E (PG-E) stimulate the Aδ-fibres that cause a sensation of sharp well

localized pain [34]. There is various peripherally acting common analgesic drugs such as ibuprofen, aspirin, diclofenac sodium and indomethacin, inhibit acid induced writhing by inhibition of prostaglandin synthesis [41]. So we can conclude that any agent that reduces the number of writhing will demonstrate analgesic effect by inhibition of prostaglandin synthesis, a peripheral mechanism of pain inhibition [42] like the fruit extract of *Phyllanthus emblica* (Linn.) in acetic acid-induced writhing method suggests that the reduction of pain might be occurred due to the presence of analgesic properties in the extract via inhibition of prostaglandin synthesis. However further study should be done for its isolated, purified active components. From the above observations it is obvious that the ethanol extract of the fruit of *Phyllanthus emblica* (Linn.) has an analgesic activity.

Xylene is responsible for vasodilation and edematous changes of skin as signs of acute inflammation [43]. The increased thickness of the ear tissues is due to histopathological changes. In the present investigation, the plant extract significantly inhibited the xylene-induced increases in ear weight in a dose related manner. This inhibition capacity of the plant extract can be regarded as the evidence of anti-inflammatory efficacy through reducing vasodilation and so that improving edematous condition. In this study the ethanolic fruit extract of *Phyllanthus emblica* (Linn.) significantly inhibited ear edema formation in xylene induced ear edema model mice. This inhibition can be considered as direct evidence that supporting the anti-inflammatory activity of ethanolic fruit extract of *Phyllanthus emblica* (Linn.)

The ethanolic fruit extract of *Phyllanthus emblica* (Linn.) has a remedy for a different bacterial disease is supported by the antibacterial screening tests results.

During the antidiarrhoeal activity by castor oil induced model, at the dose of 500 mg/kg-body weight, the extract of *Phyllanthus emblica* (Linn.) compared to the control group, offered about 0.5389 of the mean latent period for diarrhoeal episode and the result was significant ($P<0.2$). The mean numbers of stool at the 1^{st}, 2^{nd}, 3^{rd} and 4^{th} hour of study were 8.2., 9.6, 7.8 and 4.4 respectively.

Phyllanthus emblica (Linn.) showed a moderate antidiarrhoeal activity in castor oil induced test in mice at the dose of 500 mg/kg-body weight as compared to the standard antidiarrhoeal agent loperamide. *Phyllanthus emblica* (Linn.) caused an increase in latent period i.e. delayed the onset of diarrhoeal episode and decreased the frequency of defecation. The decreased frequency of defecation and increase mean latent period of test group than control group and comparison with positive control group can claim that *Phyllanthus emblica* (Linn.) might possess antidiarrheal activity. T-test of these responses showed that the result is significant throughout the observation period.

Test sample showed different mortality rate at different concentrations. The mortality rate of brine shrimp was found to be increased with the increase in concentration of the sample and plot of percent mortality versus log concentration on the graph paper produced an approximate linear correlation between them. From the graph figure the concentrations at which 50% mortality (LC_{50}) of brine shrimp nauplii occurred were obtained by extrapolation. The values were found to be 60µg/ml for the crude extract. The 90% mortality (LC_{90}) values were 100µg/ml respectively. The approximate significance value for Chi-Square Tests (Linear by liner association) is equal 0.02 and also significance value for Pearson's R is 0.001 which are significant at 5% level of significance. Therefore mortality rate of shrimp due to Ethanolic extract concentration is not due to chance. So we can conclude there is a statistically significant relationship between Ethanolic extract concentration and mortality rate

of shrimp. The crude extracts *Phyllanthus emblica* (Linn.) were found to show high lethality against the brine shrimp nauplii. These results tend to suggest its possible antitumor, antibacterial or pesticidal activities. However, further researches are necessary particularly with its purified fraction.

The preliminary phytochemical analysis of the plant extract showed the presence of reducing sugars, alkaloids, flavonoids, tannins, steroids, gums and glycosides. The previous scientific studies have been reported that alkaloids, flavonoids and tannins are known to inhibit prostaglandin synthetase that is responsible for its antinociceptive and anti-inflammatory effects [20,44-50]. Therefore antinociceptive and anti-inflammatory effect of the extract may be due to the presence of flavonoids, tannins, and alkaloids either singly or in combination. Presence of tannins, alkaloids, flavonoids, sterol and reducing sugars in the medicinal plants have also been known to indicate antidairrheal activity [51-52]. In general antidiarrhoeal activity of tannins and flavonoids has been recognized for the inhibition of intestinal motility, antimicrobial action and antisecretory effects [48]. In addition, the astringent properties of tannins are known to cause antinociceptive, antilammatory and antidiarrheal effects [20,44,53]. Therefore antinociceptive and anti-inflammatory effect of the extract may be due to the presence of flavonoids, tannins, and alkaloid either singly or in combination. Besides alkaloids, flavonoids or tannins may also be responsible for anti-diarrheal potential of the plant extract.

5. CONCLUSION

According to the results of the present investigation, we can conclude that the ethanolic fruits extract of *Phyllanthus emblica* (Linn) has significant analgesic, anti-inflammatory and anti-diarrheal effects that support to the traditional use of this plant for the treatment of related diseases. This study also suggests us for the further detail investigation of mechanisms of the pharmacological effects and also to isolate the active compound(s) responsible for those properties.

ETHICAL APPROVAL

All authors hereby declare that all experiments have been examined and approved by the appropriate ethics committee and have therefore been performed in accordance with the ethical standards laid down in the 1964 Declaration of Helsinki.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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