

Elucidation of *In-vitro* thrombolytic, membrane stabilizing, cytotoxic activities and phytochemical nature of *Bruguiera cylindrica* leaves

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ABSTRACT

Objectives: The present study was intended to investigate possible phytochemical nature (group determinant of plant extract) and to reveal *in-vitro* thrombolytic, membrane stabilizing and cytotoxic activities of crude methanolic extracts of *Bruguiera cylindrica* leaves. **Methods:** Collected plant parts were cleaned, dried, and ground to obtain powdery mass. Various methanolic extracts of *B. cylindrica* was subjected for evaluating membrane stabilizing potentials at a hypotonic solution and heat induce condition, where thrombolytic activity assessment was done by employing Streptokinase as standard. Finally, Cytotoxicity activity was determined by Brine shrimp Lethality bioassay. **Results:** Phytochemical screening of methanolic extracts revealed the presence of carbohydrate(s), glycoside(s), phenol, tannin, protein(s), gum and mucilages. Crude methanolic extracts with different doses were used for *in-vitro* thrombolytic study and 10 mg/ml concentration of plant extracts showed moderate clot lysis ($14.51 \pm 1.87\%$) while the standard (streptokinase) showed $59.73 \pm 0.97\%$ clot lysis. The methanolic extracts exhibited significant anti-inflammatory properties at 10mg/ml

concentration by both hypotonic solution and heat induced haemolysis of erythrocyte membrane i.e. $18.53 \pm 0.91\%$ and $23.60 \pm 0.89\%$ respectively. In contrast to vincristine sulphate the crude methanolic extracts showed moderate cytotoxic properties with LC_{50} value of 16.628. **Conclusions:** Our present study suggest that, further studies may carry on this plant extracts to build up an acceptable report in the medical science.

Key Words: *B. cylindrica*, phytochemical screening, Thrombolytic, Membrane stabilizing, Cytotoxic.

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INTRODUCTION

Nature always acts as a great source of rescue for human being by providing different remedies from its plants, animals, and other sources to cure all ailments of mankind. Many species of plants containing substances of medicinal value, are comprised from the plant kingdom, which are yet to be explored. Because of their possible medicinal value a large number of plants are continually being screened.¹ World Health Organization (WHO) addressed medicinal plants as an accessible, affordable and culturally appropriate source of primary health care for more than 80% of Asia's population. Not only Asia, but also the whole world is used to depend on natural sources for health related problems. Depending on indigenous systems of medicine, Indian Sub-continent who has a long tradition of herbal treatment are able to integrate two third of the plants for used in modern medicine and health care system of rural population. All of this data acknowledged for the plants are used for the treatment of disease over a decades thus become take the major consideration in present pharmacotherapy under the title of phytotherapy. Though medicinal plants have been used in traditional medicine for hundreds of years with reputation as efficacious remedies but there are very small scientific data to substantiate their efficacy. That is why most of the research work was undertaken to open up the arcane of herbal medicine and also generate data for biochemical mechanism through which they exert their effect.² We know since ancient times, medicinal plants have been used for the treatment of several diseases. On this aspect present study was carried out over *Bruguiera cylindrica*, which a medicinal plant belonging to the family *Rhizophoraceae*. It is found in South East Asia with the range extending from India and Sri Lanka through the Philippines, Malaysia, In-

donesia and Papua New Guinea to Queensland, Australia.³ It is also one of the commonest mangroves in Singapore.⁴ In the Malay language it is known as Bakau Putih. The purpose of the present study was to explore phytochemical nature, *in-vitro* clot lysis, anti-inflammatory and cytotoxic activities of the crude methanolic extracts of *B. cylindrica* leaves.

Thromboembolic disorders such as pulmonary emboli, deep vein thrombosis, strokes and heart attacks etc. and these are considered as the main causes of morbidity and mortality in developed countries.⁵ To dissolve clots many therapeutic agents are available such as alteplase, anistreplase, streptokinase, urokinase, and tissue plasminogen activator (TPA). The purpose of a fibrinolytic drug is exert their effect through dissolving thrombin in acutely occluded coronary arteries thereby to restore blood supply to ischemic myocardium and also to limit necrosis and to improve prognosis.⁶ Although in India, Bangladesh and other developing countries UK and SK are widely used as compared to other thrombolytic drugs due to their lower cost⁷ but, the use of these drug is associated with high risk of bleeding intracranial hemorrhage, severe anaphylactic reaction and lacks specificity⁸ Some recent study also support that these drugs are not suitable for used in patients who have undergone surgery or those with a history of nervous lesions, gastrointestinal bleeding or hypertension.⁸ Again, Thrombolytic therapy with recombinant t-PA is effective in acute myocardial infarction, but the treatment is limited by a fairly slow reperfusion rate and frequent early reocclusions.⁹ Fucoidan, a newly discovered thrombolytic agent, extracted from brown seaweeds with anticoagulant and antithrombotic effects mediated by direct thrombin inhibition has been reported recently.¹⁰ However, traditional medi-

cines are wide-spoken as green medicine for their safe and dependable health care paradigms. The traditional herbal medicines takes an uprising interest since couple of decades due to their tremendous pharmacological activities, economic viability and less side effects in different healthcare management system.¹¹ Thus, incredible efforts have also been directed towards the discovery and development of natural products with antiplatelet,¹² anticoagulant,¹³ antithrombotic¹¹ and thrombolytic activity of the plants not documented. Again, one of the most important pathological disorder is inflammation. It is a part of non-specific immune response that occurs in reaction to any type of bodily injury, is a complex biological response of vascular tissues to harmful stimuli.¹⁴ Cancer or tumor is the most common cause of death in both developed and developing countries. There are many methods are available to describe how cancer spread throughout the body. One method showed cancer is preliminary effect on specific part of our body and then invade to the other parts of our body very quickly and ultimately causes death of the patient.¹⁵ So it is very necessary to identify or diagnosis of cancer at early stage otherwise if it is spread other part of the body then difficult to treat. However, there are several approaches of cancer treatments are available including surgery, radiation therapy and chemotherapy. All of these approaches are aimed to destroy cancerous cell from the body. Each approach possesses several side effect.¹⁶ That is why it is now demand of the present era to discover drug with fewer side effect. There are many chemotherapeutic agents is being invented to treat various cancer. They utilize sometimes in individual form or in conjunction with other drugs in the form of chemotherapy. But most of these drugs are synthetic and shown numerous side effects. We know that plant is always the safer source for treating any kind of disease. By considering this universal truth our present study was undertaken to discover drug from natural source with fewer side effect for treating different types of cancer.

MATERIALS AND METHODS

Collection of plant materials

For this present investigation the leaves of *B. cylindrica* were collected from surrounding area of Sonadia deep, Cox's Bazar, in June, 2014. The plant was identified by expert of Bangladesh National Herbarium, Mirpur, Dhaka, Bangladesh (Accession number- DACB: 38325).

Extraction

After collection of leaves of *B. cylindrica* were thoroughly washed with water. Then the collected plant materials were chopped, dried, and powdered. About 500g of the powdered materials were soaked in 1.5 litre of methanol at room temperature for two weeks. Then the solution was filtered using filter cloth and Whatman's filter paper and concentrated with a rotary evaporator. It rendered a brown granular. The brown granular was designated as crude methanolic extract.

Phytochemical Evaluation

Small quantity of freshly prepared methanolic extracts of *B. cylindrica* leaves were subjected to preliminary quantitative phytochemical investigation for the detection of phytochemicals such as alkaloids, carbohydrates, glycosides, phytosterols, proteins, flavonoids, tannins, saponins, phenols, gums and mucilages, fats & fixed oils using the following standard methods given by Roopashree *et al.*, 2008.¹⁷

Thrombolytic Activity

Standard drug Streptokinase (SK)

Commercially available lyophilized Alteplase (Streptokinase) vial (Beacon pharmaceutical Ltd) of 15, 00,000 I.U., was collected and 5 ml sterile distilled water was added and mixed properly. This suspension was used

as a standard from which 100µl (30,000 I.U) was used for in vitro clot lysis assay method described by Prasad *et. al* 2007.¹⁸

Preparation of test sample

To evaluate the thrombolytic activity of the plant extracts test solution was prepared. The plant extracts were dissolved in methanol and shaken vigorously on a vortex mixer to prepare different concentration (2, 4, 6, 8 and 10 mg/ml respectively) of the test sample. The suspension was kept overnight and decanted to remove the soluble supernatant, which was filtered through a 0.22 micron syringe filter. Thus test samples were prepared for thrombolytic screening.

Thrombolytic potential analysis

Thrombolytic potentials of *B. cylindrica* leaves was carried out by the method of Prasad *et al.*¹⁸ 5 ml of venous blood were drawn from healthy volunteers without a history of oral contraceptive or anticoagulant therapy, which were distributed in five different pre weighed sterile micro centrifuge tubes (0.5 ml/tube) and incubated at 37°C for 45 minutes. After clot formation, the serum was completely removed without disturbing the clot and each tube having clot was again weighed to determine the clot weight (clot weight = weight of clot containing tube – weight of tube alone). Each micro-centrifuge tube containing clot was properly labeled and 100 µl of the test samples from each doses (2, 4, 6, 8 and 10 mg/ml respectively) were added to the tubes accordingly.¹⁸ As a positive control, 100 µl of streptokinase (SK) and as a negative control, 100 µl of distilled water were separately added to the control tubes. All the tubes were then incubated at 37°C for 90 minutes and observed for clot lysis. After incubation, the fluid was removed and tubes were again weighed to observe the difference in weight after clot disruption. Difference obtained in weight taken before and after clot lysis was expressed as percentage of clot lysis as shown below:

$$\% \text{ of clot lysis} = (\text{wt. of released clot} / \text{wt. of clot before treatment}) \times 100$$

Membrane Stabilizing Activity

In vitro membrane stabilizing screening of *B. cylindrical* was performed by following the methods developed by Sikder *et al.*,¹⁹

Preparation of test sample

Crude methanolic extracts of *B. cylindrica* leaves with different concentration (2, 4, 6, 8, & 10 mg/ml respectively) were prepared as the test samples for membrane stabilizing study.

Drug

Standard Acetyl Salicylic Acid (ASA) or Aspirin was used as standard drug for comparison with methanolic extracts of *A. marina*.

Red Blood Cells (RBC) Collection

5 ml of whole blood was collected from healthy human volunteers in a test tube containing an anticoagulant (EDTA 2.2 mg/ml of blood) under standard conditions of temperature 23±2°C and relative humidity 55±10%.

Preparation of Phosphate Buffer Solution

A buffer is an aqueous solution that has a highly stable pH. A pH of about 7.4 with buffer strength of 10 mM was obtained using 0.0352% monosodium phosphate dehydrate and 0.1099% disodium phosphate anhydrate. The buffer was made by adding 0.352 gm monosodium phosphate dehydrate and 1.099 gm disodium phosphate anhydrate to 1000 mL water.

Preparation of Isotonic Solution

A solution that has a concentration of electrolytes, nonelectrolytes or a combination of the two that will exert equivalent osmotic pressure as that solution with which it is being compared. Either 0.16M sodium

chloride (NaCl) solution (approximately 0.95% salt in water) or 0.3M nonelectrolyte solution is approximately isotonic with human red blood cells. For the preparation of 500 ml isotonic solution of 154 mM strength, 4.5045 gm NaCl was added and mixed.

Preparation of Hypotonic Solution

A solution of lower osmotic pressure than that of a reference solution or of an isotonic solution is called hypotonic solution. For the preparation of 500 ml hypotonic solution, having strength of 50 mM, 1.4625 gm NaCl was added and mixed.

Erythrocyte Suspension

For the preparation of erythrocyte suspension, the collected RBC was centrifuged, supernatant was removed and the blood cells were washed three times with isotonic solution (154 mM NaCl) in 10 mM sodium phosphate buffer (pH 7.4) through centrifuge action for 10 min at 3000 rpm using the same volume as supernatant. Finally it was resuspended in the same volume of this isotonic buffer solution.

Brine Shrimp Lethality Bioassay

The measurement of toxicity plays a vital role in drug discovery and is a useful tool in biological, especially ecological investigations.²⁰ It also serves as a tool for screening plant extracts of possible medicinal value. In this study, we used simple brine shrimp bioassay test of Meyer with slight modification by using *Artemia salina* as test organism, which was collected from a pet shop.

Brine shrimp hatching

Sea water was prepared by dissolving 38 gm sea salt (pure NaCl) in one liter of distilled water, which is then filtered to get clear solution of 3.8% concentration.²¹ In a suitable plastic or glass vessel sea water was taken and shrimp eggs were added to one side of the vessel and allowed to hatch for 24 hours till the mature nauplii were found. Continuous oxygen and light supply were provided to support the hatching process.

Sample preparation

Test solution was prepared by mixing 4mg of methanolic extract with 200µL of DMSO in a suitable vial by using vortex mixer. 100µL of sample was taken from the vial and mixed with 5ml of sea water in a test tube. Thus the concentration we obtained was 400µg/ml. Then a series of solution at a concentration of (200 µg/ml, 100 µg/ml, 50 µg/ml, 25 µg/ml, 12.5 µg/ml, 6.25 µg/ml, 3.125 µg/ml, 1.5625 µg/ml, 0.78125µg/ml) were prepared by serial dilution method.

Counting of noupilii

After 24 hours, the number of survived nauplii in each vial was counted by using magnifying glass. From this data the percent (%) of mortality of brine shrimp nauplii was calculated for each concentration.

Statistical analysis

All the above assays were conducted in triplicate and repeated threes for consistency of results and statistical purpose. The data were expressed as Mean \pm SD and analyzed by one way analysis of variance (ANOVA) followed by Dunnett 't' test using SPSS software of 10 version. $P < 0.05$ was considered statistically significant.

RESULTS

Phytochemical analysis

Phytochemical screening of methanolic extracts of *B. cylindrica* leaves displayed the presence of carbohydrate(s), glycoside(s), phenol, tannin, protein(s), gum and mucilages which are presented in Table-1.

Table 1: Preliminary phytochemical screening of the methanolic extracts of *B. cylindrica* (leaves).

Sl. No	Phytochemicals	Methanolic Extract
1	Alkaloids	-
2	Carbohydrates	+
3	Cardiac glycosides	+
4	Saponins	+
5	Phytosterols	+
6	Phenols	+
7	Tannins	+
8	Flavonoids	-
9	Proteins and amino acids	+
10	Terpenes	+

Here, "+" stands for Presence of Phytochemicals and "-" stands for Absent of Phytochemicals

Thrombolytic activity

The effective clot lysis percentage by five different concentrations of the plant extract, positive control (Streptokinase) and negative control (water) with statistical representation is tabulated in Table 2. From table 2,

Table 2: Effects of different doses of crude methanolic extracts of *B. cylindrica* leaves and the standard on *in-vitro* clot lysis.

Treatment	% of clot lysis (mean \pm SD)	P - value
ME (2mg/ml)	6.77 \pm 2.18	< 0.05
ME (4mg/ml)	9.90 \pm 1.33	< 0.05
ME (6mg/ml)	11.74 \pm 1.22	< 0.05
ME (8mg/ml)	12.77 \pm 2.28	< 0.05
ME (10mg/ml)	14.51 \pm 1.87	< 0.05
Blank (water) and Streptokinase		
Treatment	% of clot lysis	P - Value
SK	59.73 \pm 0.97	< 0.001
Blank	5.63 \pm 0.79	

it is evident that the percentage of clot lysis was 59.73 \pm 0.97% when 100 µl of streptokinase (30,000 I.U.) was used as a positive control, while in case of water (negative control) the percentage of clot lysis was negligible (5.63 \pm 0.79%). The mean difference in clot lysis percentage between positive and negative control was very significant ($p < 0.001$). When clots were treated with 100 µl each of different concentrations (2, 4, 6, 8 & 10 mg/ml respectively) of the test sample moderate clot lysis activity, i.e., 6.77 \pm 2.18%, 9.90 \pm 1.33%, 11.74 \pm 1.22%, 12.77 \pm 2.28%, and 14.51 \pm 1.87% respectively, was observed and when compared with the negative control (water) the mean of percentage (%) of clot lysis was significant ($p < 0.05$). Percentage of clot lysis after treatment with different concentrations of the methanolic extract and appropriate controls is shown in Figure 1.

Membrane stabilizing activity

The anti-inflammatory activities of the Crude methanolic extracts of *B. cylindrica* leaves are displayed in Table 3 & 4. The plant extracts dose dependently increased in anti-inflammatory studies, whereas 10 mg/ml concentration more significantly showed 18.53% & 23.60% inhibition of hemolysis by hypotonic solution and heat induced hemolysis of erythrocyte membrane respectively (Fig-2). Acetyl salicylic acid was used as standard in membrane stabilization. ASA (0.10 mg/mL) revealed 53.25% & 47.81% inhibition of hemolysis respectively induced by hypotonic so-

lution and heat induced hemolysis correspondingly.

Table-3: Membrane stabilizing activities of *B. cylindrica* leaves with different concentration on hypotonic solution induced haemolysis of erythrocyte membra

Treatment	Concentration	Optical density of samples In hypotonic solution (Mean ± SD)	% inhibition of haemolysis
Control	---	3.809±0.74	
ME	2 mg/ml	3.495±0.54	8.243±0.49
ME	4 mg/ml	3.328±0.28	12.62±0.84
ME	6 mg/ml	3.215±0.77*	15.60±0.90
ME	8 mg/ml	3.149±0.89*	17.32±0.67
ME	10 mg/ml	3.103±0.76**	18.53±0.91
Acetyl salicylic acid	0.10 mg/ml	1.781±0.23***	53.25±0.80

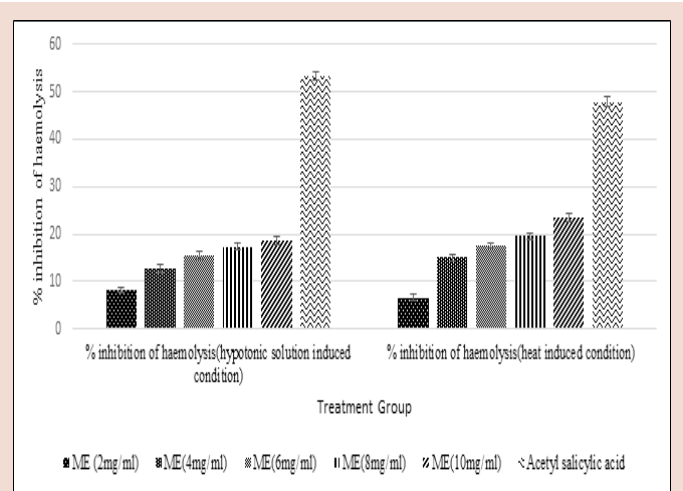


Figure 2: Effects of different conc. of *B. cylindrica* on hypotonic solution and heat induced haemolysis of erythrocyte membrane.

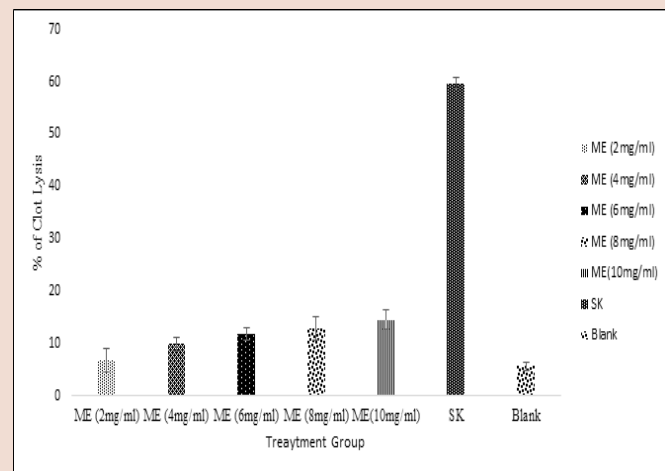


Figure 1: In-vitro thrombolytic activity of the crude methanolic extracts of *B. cylindrica* (leaves), positive control (streptokinase) and negative control (water).

Table-4: Effects of crude methanolic extracts of *B. cylindrica* leaves on heat induced haemolysis of erythrocyte membrane.

Treatment	Concentration	OD of sample ±SD		% inhibition of Haemolysis
		Heated Solution	Unheated Solution	
Control	---	1.084±0.029		
ME	2 mg/ml	0.827±0.127	0.809±0.21	6.54±0.67
ME	4 mg/ml	0.780±0.004	0.733±0.70	15.01±0.56
ME	6 mg/ml	0.713±0.114	0.634±0.32	17.55±0.47
ME	8 mg/ml	0.587±0.096	0.466±0.42*	19.57±0.77
ME	10 mg/ml	0.401±0.078	0.190±0.91* *	23.60±0.89
Acetyl Salicylic Acid	0.10mg/ml	0.690±0.065	0.329±0.39* *	47.81±0.98

Brine shrimp lethality bioassay

By using brine shrimp bioassay, developed by Meyer we could understand the cytotoxic potential and anti-tumor properties. In our current

study we used methanol and aqueous soluble extracts of *B. cylindrica*. The plant extracts showed various rate of mortality at different concentration which are given in Table-5. By plotting the log of concentration

Table 5: Results of test samples of *B. cylindrica* leaves on brine shrimp lethality bioassay.

Sample	LC ₅₀ (µg/ml)	Regression Equation	R2
Vincristine Sulphate (Positive control)	0.861	y = 34.025x + 52.581	0.9521
Aqueous Extract	8.396	y = 35.233x + 1.05	0.982
Methanol Extract	16.628	y = 37.649x + 4.0363	0.9913

against percent of mortality for all test sample, we found a linear correlation (Fig 3-5). On the basis of this correlation the LC₅₀ (the concentration at which 50% of mortality of brine shrimp nauplii occurred) was determined for each of the studied sample named methanol and aqueous soluble extracts. The LC₅₀ of standard (vincristine sulfate) was also ascertained. We also found that, there was no rate of mortality obtained, in case of control study.

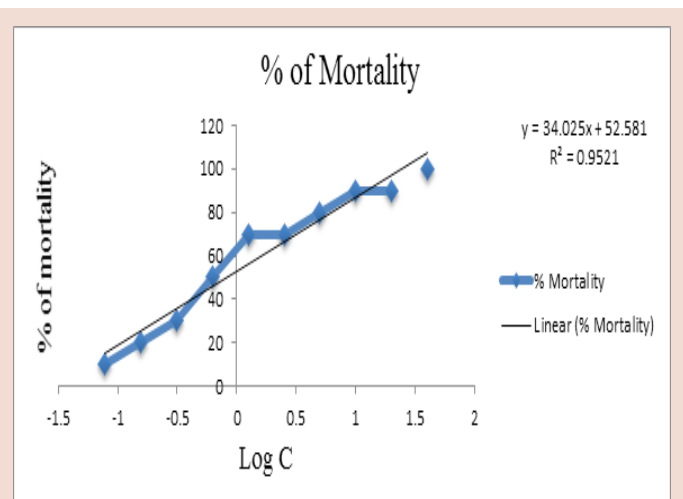


Figure-3: Effect of Vincristine Sulphate on brine shrimp lethality bioassay.

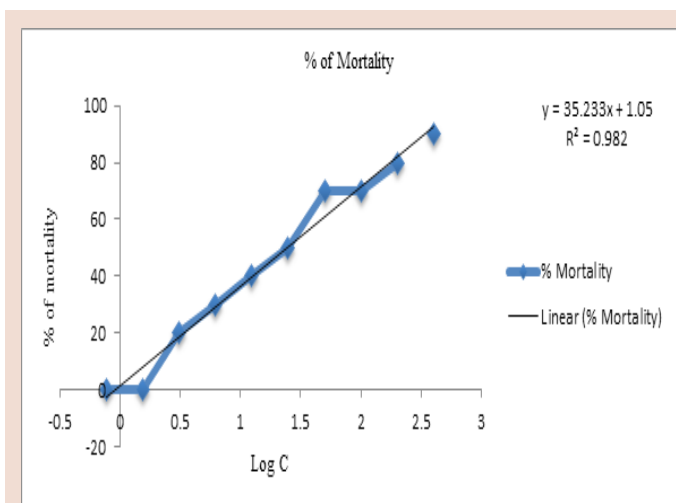


Figure-4: Effect of Aqueous soluble extract of *B. cylindrical* on brine shrimp lethality bioassay.

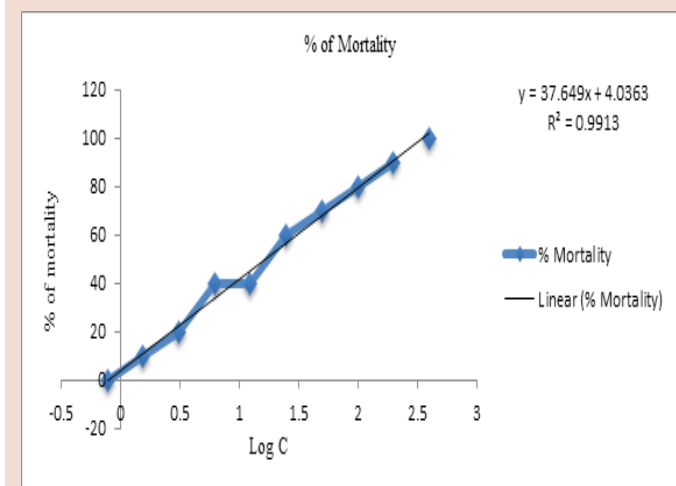


Figure-5: Effect of Methanol extracts of *B. cylindrical* on brine shrimp lethality bioassay.

DISCUSSION

Very recently phytopharmacological investigation able to create a new field to discover plant derivative drugs and renew the attention in herbal medicines, where 30% of the pharmaceuticals are prepared from plants derivatives.²¹ Some severe outcomes such as stroke and myocardial infarction manifested due to the failure of hemostasis and consequent formation of blood clots in the circulatory system. Fibrinolytic agents such as urokinase, tissue plasminogen activator and streptokinase used for clinical intervention for pathological development of blood clots. Many research works have been undertaken to discover antithrombotic (anti-coagulant and antiplatelet) effect of plants and natural food sources in order to prevention of coronary events and stroke.⁶ In the present study Methanolic extract of *B. cylindrical* showed significant thrombolytic activity, this effect may be possible due to phytoconstituents such as tannin, alkaloid, and saponin present in the plant extracts affecting activation of plasminogen both by fibrin-dependent and fibrin-independent mechanisms similar to Streptokinase which causes extra production of plasmin which breaks down fibrin the major constituent of thrombi, to dissolve unwanted blood clots. Several studies support our present findings.²² Present study evidence that the methanolic extract dose depend-

ently protect the human erythrocyte membrane against lysis induced by hypotonic solution and heat induced condition. During inflammation phagocytes release many lysosomal enzymes and hydrolytic components to the extracellular space, which assists a variety of disorders by inducing damages of the surrounding organelles and tissues.²³ Studies evidence that non-steroidal anti-inflammatory drugs act through stabilization of lysosomal membranes by inhibiting these lysosomal enzymes. Again, lysis of the RBC membranes accompanied by the oxidation when exposed to harmful substances such as hypotonic medium, heat, etc through lysis of hemoglobin²⁴ Thus mechanism of anti-inflammatory activity of the plant extract is assessed by considering their potentials in inhibition of hypotonicity and heat induced RBC membrane lysis, because human RBC membranes are considered similar to lysosomal membrane components²⁵ One can also assumed that the possible mode of action of the extract and standard anti-inflammatory drugs may be connected with binding to the erythrocyte membranes through consequent alteration of surface charges of cells. Some research works were able to reveal the name of some responsible chemical components present in the extracts, which are well known for their anti-inflammatory activity.²⁶ Both in vitro and in vivo studies in experimental animals showed that the flavonoids exert stabilizing effects largely on lysosomes while tannin and saponins are also capable of stabilizing the erythrocyte membrane with an ability of binding with cations and other biomolecules²⁷ Our present research reveals that plant methanolic extracts showed potent RBC membrane stabilization activity with a good protection against both hypotonic solution and heat-induced lysis. Plants are important resources for the development of new chemotherapeutic agents. Again, brine shrimp lethality bioassay (BSLB) is used widely in the preliminary screening of the crude extracts to evaluate the toxicity towards brine shrimps, thus providing an indication of possible cytotoxic properties of the test materials²⁸ For evaluating cytotoxic activity of our present plant extract this method was chosen because this is easiest to conduct than any other methods and also said that the cytotoxic compounds generally exhibit significant activity in the BSLB; for all of this aspect this method become one of the recommended guideline for the detection of antitumour compounds and pesticides due to its low cost²⁹ This bioassay also found to exhibit positive relationship with the human solid tumour cell lines. It is now become a proverb that if Correlation between cytotoxicity and activity against the brine shrimp nauplii using extracts have been established than cytotoxic effects of the plant extracts will enunciate for further cell line assay. Not only Bangladesh but also the country of the third world this methods got the main consideration for conducting cytotoxic activity of plant extract. BSLB also hold the basement while conducting our present study to enunciate the cytotoxic activity of methanolic extract of *B. cylindrical*. Present study data showed various rate of mortality at different concentration as the report of Anderson *et al.*³⁰ though there was no rate of mortality obtained. We want to conclude here with a message that significant lethality (as LC_{50} value less than 100 ppm or $\mu\text{g/ml}$) of the plant extract to brine shrimp is indicative of the presence of potent cytotoxic and probably insecticidal compounds which warrants further investigations.

CONCLUSION

The findings of the present study provide convincing evidence that methanolic extract of *B. cylindrical* possesses remarkable thrombolytic, membrane stabilizing and cytotoxic activity, with a demands of further biochemical studies for isolating the bioactive compounds and determine the precise mechanisms responsible for the noticed biological activities of this plant.

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CONFLICT OF INTEREST

The authors declare they have no competing interests.

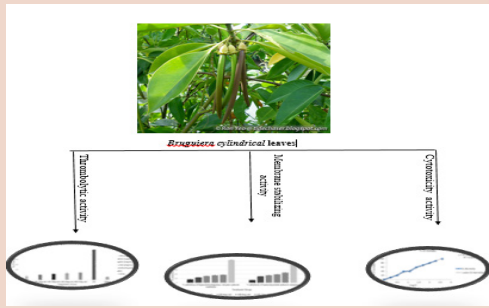
ABBREVIATIONS

RBC=Red Blood Cell, NaCl= Sodium Chloride, EDTA= Ethylene Diamine Tetra Acetic acid, UV= Ultra Violet, OD= Optical Density, ANOVA= Analysis of Variance, SD= Standard Deviation, WHO= World Health Organization, ME= Methanolic Extract, ASA= Acetyl Salicylic Acid, NSAID= Nonsteroidal Anti-inflammatory Drug.

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PICTORIAL ABSTRACT



SUMMARY

Bruguiera cylindrica which is an ethno-medicinal plant belonging to the family Rhizophoraceae. The selected plant had different phytochemical constituents. Thrombolytic potential was determined by clot lysis assay method. Membrane stabilizing activity was performed by hypotonic solution and heat induced haemolysis when cytotoxic activity was evolved by brine shrimp lethality bioassay method. The methanolic extract showed moderate thrombolytic, membrane stabilizing and cytotoxic activities.

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