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EFFICACY OF LEAVES POWDER OF *BAMBUSA ARUNDINACEA* ON ANTITHROMBOTIC ACTIVITY

Abirame S.*[1], V. Paheerathan V.[2] and Pirathipkumar R.[3]

[1]Final Year Siddha Medical Student, Unit of Siddha Medicine, Trincomalee Campus, Eastern University of Sri Lanka.
[2]Senior Lecturer, Unit of Siddha Medicine, Trincomalee Campus, Eastern University of Sri Lanka.
[3]Lecturer, Unit of Siddha Medicine, Trincomalee Campus, Eastern University of Sri Lanka.

***Corresponding Author: Abirame S.**
Final Year Siddha Medical Student, Unit of Siddha Medicine, Trincomalee Campus, Eastern University of Sri Lanka.
Email ID: abirameaa@gmail.com,

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ABSTARCT

This study was a comparative in vivo study, conducted in Wistar albino rats at Unit of Siddha Medicine, Trincomalee Campus, Eastern University of Sri Lanka. Thrombosis is the formation of a blood clot inside a blood vessel, obstructing the flow of blood through the circulatory system. When a blood vessel is injured, the body uses platelets (thrombocytes) and fibrin to form a blood clot to prevent blood loss. Thrombosis is a common pathology underlying ischemic heart disease, ischemic stroke and VTE Major burden of disease across low-income, middle-income and high-income countries. The plant *Bambusa arundinacea* was selected for this study. The plant *Bambusa arundinacea* belongs to the family Bambusaceae which popularly known as Bamboo in English and Mungil in Tamil and found throughout in India, Burma and Ceylon. In Ceylon it is common along river banks in the warmer parts of the island. It is a perennial tree with many stems tufted on a stout root stock; stems branching from the base, 24 —30 m high, 15—17.5 cm diameter, graceful, curving; nodes prominent (the lowest rooting), the lower emitting horizontal, almost naked shoots, armed at the nodes with 2 or 3 stout, recurved spines sometimes 2.5 cm long (Jayaweera, 2006). Adult female Wister albino rats of weighing around 200-250gm were obtained. The animals housed in cages under standard laboratory condition. The albino rats were divided into 2 groups 6 animals in each. Mean values of baseline bleeding time 84.50 ± 14.761sec in standard group and 88.50 ± 9.813 sec in test group after overnight fasting for 12 hours. After induced by intraperitoneal injection of the thromboplastin (#0203) - 0.48ml injection the mean values of bleeding time were subsequently decreased 69.50 ± 12.787 sec in standard group and 76.33± 9.688 sec in test group. Then the treatment was continued. Standard group administered with Phytomenadione (Vitamin K) and test group administered with *Bambusa arundinacea* leaves Powder **(**direction of medicine bid**).** After that rat's tail bleeding time was noted at 0 min, 30min and 60min after 3h and 24h. Study was tested for (using assessment (Time-Dependence Pattern**)**) anti-thrombotic activity. Mean value results in Standard group (**0 min:** 76.50 ± 10.083**s**, **30 min:** 74.20 ± 9.757**s** and **60 min:** 75.80 ± 8.786s**)** after 3h. Mean value results in test group (**0 min:** 89.00 ± 6.831**s**, **30 min:** 85.40 ± 10.526**s** and **60 min:** 84.83 ± 11.250**s**) after 3h. Mean value results in Standard group (**0 min:** 77.00 ± 8.485**s**, **30 min:** 81.80 ± 7.396**s** and **60 min:** 89.33± 6.377**s)** after 24h. Mean value results in test group (**0 min:** 103.33 ± 13.619**s**, **30 min:** 103.83 ± 13.586**s** and **60 min:** 105.17 ± 13.977**s**) after 24h.P value of bleeding time in standard group [0min: (0.391), 30min: (0.142) and 60min :(0.049) after 3h treatment. P value of bleeding time in test group [0min: (0.001), 30min: (0.001) and 60min :(0.000)] after 3h treatment respectively. P value of bleeding time in standard group [0min: (0.012), 30min: (0.007)] and 60min :(0.007) after 3h treatment. P value of bleeding time in test group [0min: (0.000), 30min: (0.000) and 60min :(0.000)] after 24h treatment respectively. In comparison of bleeding time, both the standard and test groups indicate significance of bleeding time (P value less than 0.05). The significance of bleeding time higher in the test group in comparison to standard group. Hence antithrombotic activity of *Bambusa arundinacea* leaves powder is higher compared to Phytomenadione (Vitamin K). *Bambusa arundinacea* leaves possess source of antithrombotic activity compounds for the management of various thrombogenic disorders.

KEYWORDS: Antithrombotic activity, *Bambusa arundinacea*, Phytomenadione (Vitamin K), Thrombosis, Thromboplastin, Bleeding time.

1. INTRODUCTION

This study is a comparative in vivo study. Haemostasis is the process that retains the blood within the vascular system during periods of injury. The coagulation mechanism may be thought of as a complex series of cascading reactions involving development of enzymes from their precursor (zymogens). Most of the substances which are necessary for coagulation are present in an inert form and must be converted to an activated state. Most adult cardiovascular disorders involving hypertension, cerebral hemorrhage, coronary thrombosis, arteriosclerosis and CHD are caused by problems in the blood circulatory system as blood clotting disorders which constitute a serious medical problem (Sliver et al, 1974). Anti- thrombotic include anticoagulants, anti-platelets and thrombolytic that decrease the rate of blood clotting in the body by dissolving already formed ones or prevent clot formation (Webster,2001). Oral anticoagulants have been used in the management of atherothrombotic stroke treatment (Donnan et al, 2008) which accounts for 61% of all strokes and have been relied upon for prevention and treatment for several decades.

About 80% world's population depends on traditional medicine for their health benefits. Traditional medicine contains a wide range of materials which is used to treat several disorders. There is a critical need to explore for effective and alternative anticoagulants and antioxidants from natural products with minimal effect. The plant *Bambusa arundinacea* is valued for many medicinal properties in extracts from different parts of the plant (Leave¸ Root ¸Seed¸ Uppu and Nodes) have been shown to possess Stimulant, Astringent, Tonic, Antispasmodic, Aphrodisiac activities (Murugesamudaliyar,2008). Leave has been proved to possess activities Emmenagogue, Anthelmintic (Murugesamudaliyar, 2008). Preparation from the plant leaves is popular and highly acclaimed used for manage the abdominal pain and intestinal pain Dr. K.S. Murugesamudaliyar(2008) has states that the leaves of the plant can be used for anti-thrombotic activity, but it has not been clinically proven yet.

The present study sought to investigate the antithrombotic activities of *Bambusa arundinacea* leaves using standard experimental models.

There are lots of herbal based interventions used for treating antithrombotic but, there's no final objective decision of the effectiveness of these medicines. So the positive results of this research may use for antithrombotic activity as a standard & upgrading the siddha medical system. In order the research has been plan to assess the antithrombotic activity of this plant.

2. OBJECTIVE

To evaluate the antithrombotic activity on leaves of *Bambusa arundinacea*.

3. MATERIALS AND METHOD

3.1 Study design: Comparative in vivo study.
3.2 Study area: Trincomalee Campus, Eastern University.
3.3 Study population: Wistar albino rats.
3.4 Inclusion criteria: Well-being rats weight 200-250g.
3.5 Exclusion criteria: Diseased rats weight below 200g

3.6. Plant material and chemicals

Bambusa arundinacea leaves were collected in Vavuniya district & the plant was identified and authenticated. Phytomenadione (Vitamin K) and Thromboplastin (#0203) were obtained from the General Hospital, Vavuniya.

3.7. Preparation of plant materials

The collected fresh leaves were dried and made into a finely powdered material. Finely powdered material 500 mg mixed with 5ml of distilled water. Then 60.810mg for each Wister albino rat twice a day.

3.8. Anti- thrombotic studies

Wister albino adult female rats weighing 200-250gm were obtained from animal house of Medical Research Institute, Colombo. The animals were grouped and housed in cages with under standard laboratory conditions and the rats were given 12h light and 12 h dark cycles. The animals were allowed to acclimatize to the environment for 7 days. They were fed with standard pellet diet and water.

The rats were divided into two groups of six rats each. Then rat's tail baseline bleeding time was noted overnight fasting (12H) female Wister albino rats. After that thrombus was induced in Wister albino rats by single

intraperitoneal injection thromboplastin (#0203) (0.48ml/225g) anesthetized by Ether. Thromboplastin (#0203) is thrombogenic agent. After 10min again bleeding time was noted.
Dose calculation for powder of test drug:
Animal equivalent dose calculation based on body surface area.

AED (mg/kg) =Human dose (mg\kg) ×Km ratio

The standard group of each 6 rats (R1,R2,R3,R4,R5,R6) administered with Phytomenadione (Vitamin K) 0.43mg/225g twice a day and the test group of each 6 rats (R7,R8,R9,R10,R11,R12) administered with 60.81mg/225g leaves powder of the plant *Bambusa arundinacea* twice a day. Rat's tail bleeding time was noted at 0 min, 30min and 60min after 3H and 24H of treat with test and standard doses.

All data obtained were analysed by compare means test using the Statistical Package for the Social Sciences (SPSS) version 21 at a statistical significance level of P.

4. RESULTS AND DISCUSSION

Table 4.1: Changes of bleeding time between standard and test groups.

Standard		R1	R2	R3	R4	R5	R6
Baseline		1 min 18sec	1 min 30sec	1min 03sec	1 min 25sec	1 min 48sec	1 min 23sec
Thrombogenic Agent (Thromboplasin)		1min 02sec	1min 15sec	51sec	1min 12sec	1min 29sec	1min 08sec
1st Dose Is Given							
After 3H	0min	ND	1 min 15sec	ND	1min 12sec	1min 31sec	1min 08sec
	30 min	1min 05sec	1 min 16sec	ND	1min 12sec	1min 30sec	1min 08sec
	60 min	1min 08sec	1 min 18sec	ND	1min 13sec	1min 30ec	1min 10sec
2nd Dose Is Given							
After 24H	0min	1 min 08sec	1 min 20sec	ND	1 min 15sec	1min 30sec	1 min 12sec
	30 min	1 min 16sec	1 min 25sec	ND	1 min 20sec	1min 33sec	1 min 15sec
	60 min	1 min 26sec	1 min 32sec	1 min 31sec	1 min 28sec	1 min 39sec	1 min 20sec
Test		R7	R8	R9	R10	R11	R12
Baseline		1 min 41sec	1 min 20sec	1 min 15sec	1 min 36sec	1 min 27sec	1 min 32sec
Thrombogenic Agent (Thromboplasin)		1min 28sec	1min 08sec	1min 04sec	1min 24sec	1min 12sec	1min 22sec
1st Dose Is Given							
After 3H	0min	1min 36sec	ND	ND	1min 32sec	1min 20sec	1min 28sec
	30 min	1min 38sec	ND	1min 10sec	1min 30sec	1min 21sec	1min 28sec
	60 min	1min 40sec	1 min 15sec	1min 10sec	1min 32sec	1min 22sec	1min 30sec
2nd Dose Is Given							
After 24H	0min	1 min 62sec	1 min 46sec	1 min 25sec	1 min 47sec	1 min 30sec	1 min 50sec
	30 min	1 min 62sec	1 min 46sec	1 min 25sec	1 min 48sec	1 min 31sec	1 min 51sec
	60 min	1 min 65sec	1 min 47sec	1 min 25sec	1 min 49sec	1 min 34sec	1 min 51sec

*R1, R2, R3, R4, R5, R6, R8, R9, R10, R11, R12 - Wister albino rats *ND - Not Defined.

Table 4.2: Bleeding time in standard group. Standard Group.

Standard Group		Mean	Std. Deviation	Std. Error of Mean	Paired 't'	Sig. (2-tailed)
Before Treatment		69.50	12.787	5.220	-	-
After 3h Treatment	0min	76.50	10.083	-1.000	0.391	5.041
	30min	74.20	9.757	-1.826	0.142	4.363
	60min	75.80	8.786	-2.804	0.049	3.929
After 24h Treatment	0min	77.00	8.485	-4.417	0.012	3.795
	30min	81.80	7.396	-5.177	0.007	3.308
	60min	89.33	6.377	-4.417	0.007	2.603

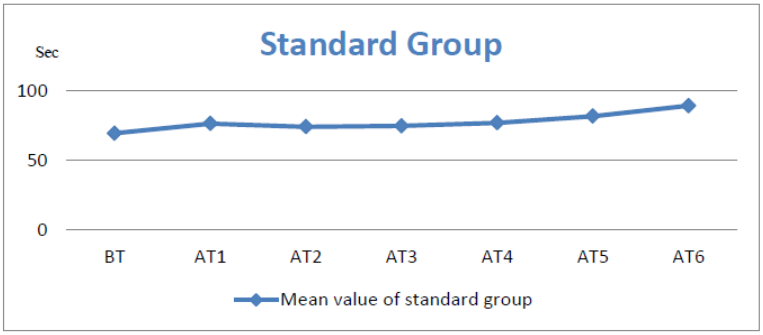

Figure 4:1 Mean value of standard group

Table 4.3: Bleeding time in test group.

Test Group		Mean	Std. Deviation	Std. Error of Mean	Paired 't'	Sig. (2- tailed)
Before Treatment		76.33	9.668	3.947	-	-
After 3h Treatment	**0min**	89.00	6.831	-15.000	0.001	3.416
	30min	85.40	10.526	-8.488	0.001	4.707
	60min	84.83	11.250	-9.604	0.000	4.593
After 24h Treatment	**0min**	103.33	13.619	-8.482	0.000	5.560
	30min	103.83	13.586	-8.977	0.000	5.546
	60min	105.17	13.977	-9.229	0.000	5.706

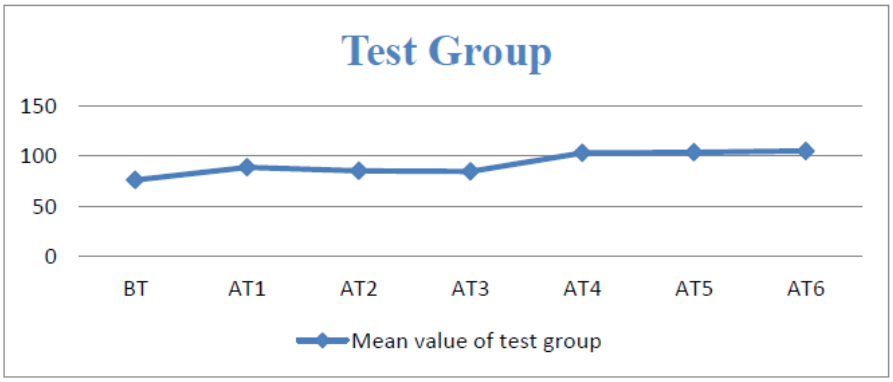

Figure 4: 2 Mean value of test group

BT:-Before Treatment
AT1:-After 3h treatment in 0min
AT2:-After 3h treatment in 30min
AT3:-After 3h treatment in 60min
AT4:-After 24h treatment in 0min
AT5:-After 24h treatment in 30min
AT6:-After 24h treatment in 60min

Discussion of antithrombotic activity based on Siddha System

- *Ilaaghu* gunam cosist of
  - Panchabhutha composition- *Akayam+Vayu+Anal*
  - *Suvai -Astringent, Bitter*
  - *Veeriyam- Ushanm*
  - *Vipakam –Astringent*
- *Ushnam* gunam cosist of
  - Panchabhutha composition (*Anal*)
  - *Suvai* (*Sour, Salt, Pungent*)
  - *Veeriyam* (Ushanm)
  - *Vipakam* (*Pungent*)
- *Ruksha* gunam cosist of
  - Panchabhutha composition (Vayu+Agni+Piruthuvi)
  - *Suvai* (*Pungent, Astringent, Bitter*)
  - *Veeriyam* (*Ushanm*)
  - *Vipakam* (*Pungent*) (Shanmugavelu, 2009).

Bambusa arundinacea has Suvai - Thuvarpu (Astringent)
Veeriyam - Vepam (Heat)
Pirivu - Kaarpu (Pungent)

Above concept explains that *Suvai*, *Veeriyam*, *Vipakam* indicating the action *Ilaaghu*, it has the properties of prevent stagnation of fluid. So *Bambusa arundinacea* prevent stagnation of blood and formation of blood clot. It acts as antithrombotic. Therefore quotation of

Bambusa arundinacea mentioned in Gunapadam text was proved scientifically through this study (Sastry, 2010).

5. CONCLUSION

As per this experimental study, the data reveals the plant possessing the antithrombotic activity. The quotation for general character of *Bambusa arundinacea* is scientifically proven from the above study for antithrombotic activity.

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