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IN VIVO STUDY OF EFFICACY OF *APARAJITA MULA* (ROOT OF *Clitoria ternatea*) IN SNAKE VENOM POISONING

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IN VIVO STUDY OF EFFICACY OF *APARAJITA MULA* (ROOT OF *Clitoria ternatea*) IN SNAKE VENOM POISONING

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ABSTRACT:

Visha (Poison) is a substance which after entering the body disturbs natural and physiological function of the body. The poison most difficult to treat among animal poisons is that of the snakes. Snake bite is an important and serious medico legal problem in many parts of the world, especially in south Asian countries. It has been estimated that 5million snake-bite cases occur worldwide every year causing about 100,000 deaths. Currently the only scientifically valid treatment for snake venom envenomation is Serotherapy i.e. polyvalent anti-snake venom serum (PVASVS). Available first aid measures in snake-bites are application of tourniquet, incision and suction but their effectiveness is limited and required trained person for that. In *Ayurved* various plant species are mentioned as Anti- ophidian. But their effectiveness in snake-bite couldn't investigate scientifically. So to investigate effectiveness of plant species as a first aid measure i have selected in vivo study. "*Aparajita mula churna*" was selected for the study. As it is mentioned in '*Eksaragana*' in *Sushruta Samhitaa*, also in various *Granthas* and *Nighantus Aparajita* is described as remedy for snake venom poisoning.

Key Words: Serotherapy, Anti-snakes venom serum, Antiophidian, *Eksaragana*

INTRODUCTION

Visha (Poison) is a substance which after entering the body disturbs natural and physiological function of the body. The poison most difficult to treat among animal poisons is that of the snakes. In India the large number of people stays in villages. The number of villages is in mountainous region covered by dense evergreen forest in which poisonous snakes are very common. Hence the snake bite cases are more common in villages. Poor access to health services in these setting and in some instances a scarcity of Anti-venom, often leads to poor outcomes & considerable morbidity and mortality. Many victims fail to reach hospital in time or seek medical care after a considerable delay because of lack of transport facility. Some even die before reaching hospital. In the rural tropics victims are often bitten in an agricultural field or jungle and in many instances the bitten species is not identified, so the mortality rate is also high in snake poisoning cases.

Currently 'Poly valent Anti snake venom serum' [1] is used for snake bite but which is not available every time. In snake bite cases it is necessary to give immediate treatment, but it is not possible every time in villages due to lack of transport facility. So that the period between snake bite and actual treatment increases; this may be one of the causes for poor prognosis in such circumstances.
In *Ayurved* many remedies are indicated for snake bite, but experimentally they are not proved. So now it is the need of our science to introduce our medicines all over the world with the help of research and experimental study. '*Aparajita mula*' is one of the drug remedy mentioned in certain classics of Ayurveda [2,3] and *Nighantu*, for the treatment of snake poisoning, but its efficacy has not been proved with the help of modern parameters. Present research was conducted with an intention to evaluate the efficacy of '*Aparajita mula'* in cobra and Russell's snake envenomation in Albino-mice.

AIMS & OBJECTS

1) To study the efficacy of *'Aparajita Mula'* (Root of *Clitoria ternatea*) in cobra venom poisoning as a first aid measure. (In Vivo).
2) To study the efficacy of *'Aparajita Mula'* (Root of *Clitoria ternatea*) in Russell's viper venom poisoning as a first aid measure. (In Vivo)
3) To study whether there is any adverse drug reaction between *'Aparajita Mula'* & Poly Valent Anti Snake venom Serum (PVASVS)

MATERIALS & METHOD

I) Review of Literature

I) Vedic *Samhita, Sangraha, Nighantus* & Modern literature
II) New researches related to this topic

II) Collection of Cobra Venom & Russell's viper Venom

The lyophilized snake venom was procured from 'Halfkin's Institute Mumbai'.

III) Collection of Polyvalent Anti Snake Venom Serum (PVASVS)

Lyophilized PVASVS was procured from 'Halfkin Institute, Mumbai'.

IV) Preparation of drug & Standardization of drug:- [4]

The dried powder of *'Aparajita Mula'* was used for experiment.

V) CPCSEA Guidelines were followed as per Ethical Committee instructions & Institutional Ethics Committee Clearance (IEC) was obtained from National Toxicology Center, Pune in the month of September 2009.

V) In Vivo Efficacy of Plan Drug

Animal Species : Albino-mice.
Place of Experiment : National Toxicology Center, Pune
Sex of Animal : Albino mice of either gender
Avg. wt. of Animals : 20 gm.
No of Animals : 3 mice of each group
No. of groups : 8
Route of Administration:
Snake Venom - Intramuscular route
Aparajita Mula Churna - Oral route
PVASVS - I.V. route

Table No 1: Groups of Animals

Group – I	Only common cobra venom
Group – II	Common cobra venom + *Aparajita Mula*
Group – III	Common cobra venom + PVASVS
Group – IV	Common cobra venom + *Aparajita mula* + PVASVS
Group – V	Only Russell's viper venom
Group – VI	Russell's viper venom + *Aparajita Mula*
Group – VII	Russell's viper venom + PVASVS
Group – VIII	Russell's viper venom + *Aparajita mula* + PVASVS

Group I & V - Control Group
Group III & VII - Standard Group
Group II, IV, VI & VIII- Experimental Group

OBSERVATION

Animals were observed for the following parameters

1. Severity & duration of paralytic signs [5] of cobra venom injected mice.
2. Severity & duration of convulsion [6] of cobra venom injected mice.
3. Duration of survival in control group [7].
4. Duration of survival in standard group.
5. Duration of survival in experimental group.
6. Any observable adverse reaction in mice treated with PVASVS.

Table No 2: Appearance of signs (average time in minutes)

Parameter	I	II	III	IV	V	VI	VII	VIII
Appearance of Paralysis	130	53	-	-	-	-	-	-
Appearance of Convulsion	148.33	109.33	-	-	-	-	-	-
Duration of Survival Period	151.33	112.66	-	-	2018.33	1655	-	-

ANALYSIS & RESULTS

Table No 3: Appearance of Paralysis

Test	Common Cobra control Group Gr.I	Aparajita Mula Gr.II
Mean	130	53
SE	8.737	14.5433
Median	135	50
SD	15.133	25.1694
Minimum	113	30
Maximum	142	80
Sum	390	160
Count	3	3

Comparison by Unpaired t-test (two tailed)

t = 4.567
P = 0.0106(significant)

Time of appearance of paralysis was significantly decreased by 77 min in *Aparajita Mula Churna* group.

Table No 4: Appearance of Convulsion

Test	Common Cobra control Gr. I	Aparajita Mula Gr. II
Mean	148.3333	109.3333
Standard Error	0.8819	19.33
Median	148	90
Standard Deviation	1.528	33.4861
Minimum	147	90
Maximum	150	148
Sum	445	328
Count	3	3

Comparison by Unpaired t-test (two tailed)

t =2.015
P =0.1141(not significant)

Time of appearance of Convulsion was decreased by 39 min in *Aparajita Mula Churna* group, but not significantly.

Table No 5: Duration of Survival Period

Test	Common Cobra control Group Gr.I	Aparajita Mula Gr.II
Mean	151.333	112.66
Standard Error	1.20	19.1871
Median	152	95
Standard Deviation	2.08	33.2321
Minimum	149	92
Maximum	153	151
Sum	454	338
Count	3	3

Comparison by Unpaired t-test (two tailed)

t = 2.011

P = 0.1146 (not significant)

Duration of Survival was decreased by 38.67 min in *Aparajita Mula Churna* group, but not significantly.

Table No 5: Observation in Viper Venom group

Out Come	Russell's Viper control Group Gr. V	*Aparajita Mula* Gr.VI
Survived	-	2 (66.67%)
Died	3(100%)	1 (33.33%)
Total	3	3

Comparison by Fisher's Exact test (Two Tail)

P =0.4000 (not significant)

In Control group all mice i.e. 3(100%) mice are died and in *Aparajita Mula* group 1(33.33%) mice is died and 2(66.67%) mice are survived but this is statistically not significant.

- **Group III (Standard Common cobra group):-**
  - All three mice survived completely without showing any signs.
- **Group IV:-**
  - All three mice survived completely without showing any signs.
- **Group VII (Standard Russell's viper group):-**
  - All three mice survived completely without showing any signs.
- **Group VIII:-**
  - All three mice survived completely without showing any signs.

RESULTS

- In Common cobra control group (Gr. I) appearance of paralysis was observed after **130min** (average) and that of drug group (Gr. II) it was after **53min** (average) i.e. time duration of appearance of paralysis was decreased by **77min** in *Aparajita Mula Churna* group, which is statistically significant. **P value is 0.0106** (Two tail).
- In Common cobra control group (Gr. I) appearance of convulsions was after **148.33min** (average) and in drug group (Gr. II) it was after **109.33min** (average) i.e. time duration of appearance of convulsions was decreased by 39 min in *Aparajita Mula Churna* group, but P value is not statistically significant. P value is 0.1141(Two tail).
- In Common cobra control group (Gr. I) duration of survival was **151.33min** (average) & that of drug group (Gr. II) duration of survival was **112.66min** (average) i.e. duration of survival was decreased by 38.67min in *Aparajita Mula Churna* group, but P value is not statistically significant**. P value is 0.1146**(Two tail).
- In Common cobra venom + PVASVS (standard group III) all mice survived completely without showing any signs
- In Common cobra venom + *Aparajita Mula Churna* + PVASVS group (Gr. IV) all mice survived completely without showing any signs. No adverse interaction between *Aparajita Mula Churna* and PVASVS was seen.
- In Russell's viper venom group (Gr. V) of survival was ***2018.3min*** (average) & In drug group (Gr. VI) only one(33.34%) mice was died and survival period was **1655min** and Two (66.67%) mice were survived, but which is statistically not significant. ***P value is 0.4000*** (Fisher's exact Test).
- In Russell's viper venom + PVASVS group (Gr. VII), all mice survived without showing any signs.
- In Russell's viper venom + *Aparajita Mula Churna* + PVASVS group (Gr. VIII), all mice survived without showing any signs. No adverse interaction between *Aparajita Mula Churna* and PVASVS was seen.

CONCLUSION

1) ***Aparajita Mula Churna* is not useful as a first aid measure in Common cobra Venom because,**
   - It does not delays the onset of symptoms and survival period in Common cobra venom poisoning,
   - Symptoms appear earlier when *Aparajita Mula* Churna was given after ingestion of Cobra Venom.

   Hence we can say that, adverse drug reaction appear due to administration of

Aparajita. So it is unfair to use *Aparajita Mula* Churna as a first aid measure in Cobra venom poisoning.

2) ***Aparajita Mula Churna*** **is not useful as a first aid measure in Russell's viper Venom because,**

- It does not delay the survival period in Russell's viper venom poisoning.
- Though 66.67% mice are survived but which is statistically not significant.

P Value (Fisher's exact test) = 0.4000 (Considered not significant)

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