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Acute Oral Toxicity Study Of *Vatsanabh* (Aconitum Ferox Wall)

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Abstract –
Vatsanabh(Aconitum ferox Wall) is one among the *mahavisha*s the potent toxic drugs explained by *Bruhattrayees*. After processing, it is also a powerful plant that is currently recognised by humanity and used to treat a few illnesses. Inappropriate use or accidental ingestion can result in various signs and symptoms that might lead to potentially fatal conditions. The main objective of this study is to determine the toxicity dose(LD50) of *Vatsanabh*. Swiss albino mice were used for the present study. The LD50 value of *Vatsanabh* deteremined according to the OECD 425: Up and Down Procedure. Various dosage levels were tested, starting from a concentration of 175mg/kg. The study employed close monitoring of motor activity, convulsions, and other physiological parameters to evaluate the drug toxicity. The research ultimately aimed to establish the lethal dose (LD50) of *Ashuddha Vatsanabh*. Following the toxicity assessment, it was determined that the LD50 dose was approximately 550 mg/kg. The quantitative analytical examination of *Vatsanabh* is suggested by this study to strengthen the toxicological analysis.
Keywords – *Vatsanabh*, LD50, oral toxicity

Introduction -

Herbal medicines are recognised as therapeutic options in alternative and traditional medicine to treat several diseases. A common component of Indian formulations (Ayurvedic) is aconite. One of the world's most toxic perennial herbs are aconites, which are members of the Ranunculaceae family and belong to the genus Aconitum[1]. These are also known as Monkhood, Wolf's bane, Leopard's bane, bikh, mithazahar. In the Indian markets, it is marketed as "*Vatsanabh*a," a blend of eight Aconitum species[2]. Aconitum Ferox (Indian Aconitum), in comparison to Aconitum napellus is far more dangerous and also widely recognised as the "king of poisons". The roots of aconite are found to be most powerful vegetable poison. It contains poisonous alkaloids such as aconitine, picroaconitine and pseudoaconitine[3]. The range of total alkaloid concentration is 0.8% to 1.2%. In *Ayurveda*, the *Vatsanabh*'s roots (Kand) are used to treat a variety of illnesses after being processed.

Vatsanabh is the primary ingredient in many formulations, such as *Sanjivani Vati, Mrutyunjaya rasa, Kaphaketu rasa* and others. Accidental poisoning can be caused either by improper preparation of formualtions or by taking Aconitum instead of horseradish roots[4]. The *Ashuddha Vatsanabha* (nonpurified drug) is much more toxic than the *Shuddha*[5]. In case of poisoning, it can cause symptoms like palpitations, tingling, vomiting, hippus and mydriasis. Many fatal poisoning cases with aconite have been described in case reports[6-10]. There are over 600 reported cases of poisoning in China alone up to 2006 and in Hong Kong it was estimated that 75% of Chinese herbal medicine related hospital admissions were related to aconite toxicity[11]. An Ayurvedic herbal preparation *Mahashankha Vati*, which contains eight ingredients along with *Vatsanabh*, has been linked to one case of poisoning[12]. An investigation of the toxicity of Aconitum Ferox extract in rats was published in 2019. The researchers observed that excessive amounts of the extract may harm the kidneys and liver, emphasizing the need for caution while using this plant medicinally[13]. Information about a drug's acute toxicity is particularly crucial for organizing phase III clinical trials and for predicting the consequences of possible overdose situations. In Ayurvedic literature, it is clearly mentioned that *Ashuddha Vatsanabha* can be used to treat severe cases of animal bites (*Jangam Visha*)[14]. Hence it is mandatory to know the LD50 value of the *Vatsanabh*. But it is seen that various research studies shown different LD50 value of *Vatsanabh*[15,16]. To ascertain whether a material or water sample is hazardous to living things, an acute toxicity test is utilized. This makes it feasible to assess the degree to which the toxicity test's goal has been met. Therefore, the main objective of this study is to determine the toxic dose of the *Vatsanabh*.

Materials and Methods

Drug preparation -

The source of *Vatsanabh* was Chamoli, Uttarakhand. The authentication of *Vatsanabh* was done at Botanical Survey of India, Pune. According to *Sharangdhar Samhita*[8], the fine churna of *Vatsanabh* was prepared.

For preparation of *Vatsanabh* churna, 50gms of well dried *Vatsanabh* roots (*Kand*) was taken and cut into small pieces. It was put into Khalva and ground until a fine powder was made. It was then sieved through 100 no. mesh. In this way *Vatsanabh* churna is prepared for the experiment. To ensure the quality of drug, it is necessary to standardize that drug before using in experiment. This study was done at Sudhatatwa Pharmacy, Dr. D. Y. Patil Ayurved college, Pune.

Standardization Report of '*Vatsanabh*' Churna:

- **Organoleptic characters of *Vatsanabh* churna:**

Parameters	Churna
Colour	Light Brown
Odour	Characteristic
Taste	Slightly Bitter

- **Physicochemical Parameters:**

Total Ash (% w/w)	1.19%
Acid Insoluble Ash (% w/w)	0.07%
Water soluble extractives (% w/w)	9.20%
Alcohol soluble extractive (% w/w)	29.31%
Foreign Matter (%)	0.1%
Loss on drying	11.33%

Animals and experimental design

The median lethal dose (LD50) of the aqueous solution of *Vatsanabh* was determined using Swiss albino mice. The LD50 for the aqueous solution of *Vatsanabh* was determined according to the OECD 425 : Up and Down Procedure and its value was calculated using the AOT statistical program (AOT 425, version 1.0, USEPA, Washington DC, USA)

The animal management protocol and the experimental design were approved by the Institutional Animal Ethical Committee, Haffkine Institute for Training, Research & Testing, Parel, Mumbai, registration number – HITRT/IARC/06/2023

All the animals were kept under acclimatization for 7days before dosing. The animal was marked with saturated picric acid solution in water for proper identification. They were

exposed to natural day and night cycles with ideal laboratory conditions in terms of ambient temperature, humidity.

Following protocol were used for animal selection –

Species	Swiss albino mice
Strain	Mus musculus
Housing	As per the standard guidelines' requirements
Acclimatization	For a minimum period of 7days
Feeding regime	Diet and Water adlibitum
Weight	18-20 grams
Sex	Female

A total of 9 healthy female of body weight range 18-20gm Albino mice were selected. The mice were housed in polypropylene cages (5 mice per cage) in a ventilated room with a controlled light(12h): dark(12h) cycle and temperature of range ($25^0C \pm 2^0C$). The animals were marked as head, neck, body tail, no mark, head & neck & forelimb. The animals were administered dosing sample using intragastric gavage.

Toxicity study –

Drug preparation (stock solution) of *Vatsanabh* root powder is taken $1/5^{th}$ of LD50 (powder/kg body weight) in 20ml of distilled water in test groups. Test drug was administered orally once on 1^{st} day in the morning session between 11am and 1pm and observed for 14 days, including experiment day.

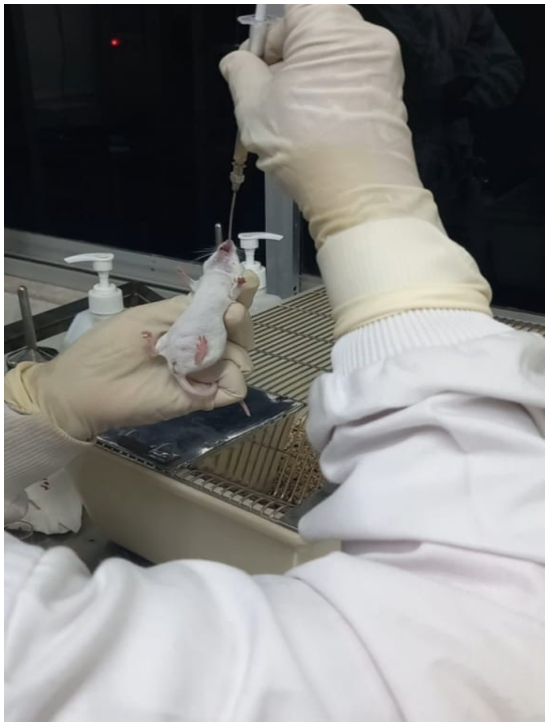
According to OECD 425, when no information is available, starting dose of 175mg/kg is recommended and after observation of single dose, animal progression factor 3.2 or one-half log have been used for further concentration. Using a consistent dose volume (1 milliliter per 100 grams of body weight), each animal received 1750 mg/kg, 550 mg/kg, 175 mg/kg, and 55 mg/kg.

Each animal receives a single dose.

After the dosage, the animals were observed closely for four hours. The animal was carefully observed from the side of the cage without interfering with its attention, and at the conclusion of each hour, it was taken to an open arena to record any behavioral changes, such as decreased or increased motor activity, catatonia, spasticity, opisthotonus, convulsions, straub's reaction, muscle spasm, hyperesthesia, muscle relaxation, anaesthesia, arching and rolling, lacrimation, salivation, diarrhoea etc.

Mortality :

Throughout the 14-day trial period, all animals were monitored for mortality at ½, 1, 2, 3, 4, 24, 48 hours after dosage, and then every day after that.



Result and discussion –

As per the guidelines 175mg/kg of *Vatsanabh* moola churna was administered to the animal dose of 0.525ml of solution. It is observed that mice was survived for 14 days. For the second mice 550mg/kg of *Vatsanabh* moola choorna was given as per the protocol and verified with AOT software. The animal dose of 0.534ml was administered. It was observed that mice was died in 4 hours due to respiratory distress.

For the next animal 175mg/kg of *Vatsanabh* moola churna was given. 0.520ml of solution was administered to the animal. It is observed that initially there was decreased motor activity, look pale but then after it survived till 14 days. Three mice were then given a dosage of 550 mg; two of them survived, while one died within 24 hours as a result of respiratory failure and reduced motor activity.

AOT software indicates that a dosage of 1750 mg/kg was chosen for two mice, one of whom survived and the other of whom died within 1 hour. Additionally, one lowered dose 55mg/kg was administered to one mice. The mice survived till 14 days. From the above observations of this study, it is concluded that the LD50 of the *Vatsanabh* is found to be 550 mg/kg for mice. The LD50 of *Vatsanabh* was determined to be 550 mg/kg for mice based on the aforementioned observations.

Sr. no.	Body weight of Mice	Dose	Mortality
1	26.3gm	175mg/kg 0.525ml	0/1
2	26.7gm	550mg/kg	1/1

		0.534ml	
3	26.1gm	175mg/kg 0.520ml	0/1
4	26.4gm	550mg/kg 0.528ml	0/1
5	27.7gm	1750mg/kg 0.554ml	0/1
6	27.9gm	550mg/kg 0.558ml	1/1
7	27.0gm	55mg/kg 0.540ml	0/1
8	26.1gm	550mg/kg 0.540ml	0/1
9	26.5gm	1750mg/kg 0.530ml	1/1

Conclusion –

The computed LD50 for *Vatsanabh moola choorna's* acute oral toxicity was 550 mg/kg bodyweight. The lethal dose at which 50% of the test population dies is known as the LD50 value, and it is commonly used to quantify toxicity. In addition to determining safe dosage ranges, acute toxicity studies serve as a basis for long-term research and other animal testing procedures by directing future investigations into chronic toxicity. By defining the therapeutic window in which toxic compounds can be used as medications without risk, these investigations aid in the preparation of safe *Ayurvedic* formulations. The quantitative analytical examination of *Vatsanabh* is suggested by this study to strengthen the toxicological analysis.

References –

1. Jasmin Kaur, Aconite Poisoning and its Status in India. International Journal of Forensic Science, 2020;3(2):123–127.
2. Sarkar PK, Prajapati P and Chauhan MG. Pharmacognosy of aconite sold under the name Vatsanabha in Indian market. Indian Journal of Traditional Knowledge 2012; 11(4):685–696.
3. R.N. Karmakar, Forensic Medicine and Toxicology;1st edition; Academic Publishers, Kolkata;2006;94
4. Parikh CK. Parikh's Text of Medical Jurisprudence and Toxicology; 6th edition; Delhi CBC Publishers and Distributors;1999; :10.66
5. Shastri K. Vishaoupavisha Vijinaniya Rasa Tarangini of Sadananda Sharma. 2012; 8th edition;Varanasi Motilal Banarasidas:653 Ch. 24., Ver. 66

6. Sharma M, Kumar S, Prashar A and Sharma A. Case Report: An interesting case of suicidal poisoning. Online Journal of Health and Allied Sciences 2010; 8(4):14.
7. Lowe L, Matteucci MJ, Schneir AB. Herbal aconite tea and refractory ventricular tachycardia. New England Journal of Medicine 2005; 353:14.
8. Moritz F. Compagnon P. Kaliszczak IG. Kaliszczak Y. Caliskan V. Girault C. Severe acute poisoning with homemade Aconitum napellus capsules: toxicokinetic and clinical data. Clinical Toxicology(Phila) 2005; 43:873–6
9. Munnecom THC, Van Kraaij DJW, Van Westreenen JC. Case report: Arrhythmia a deux: A poisonous salad for two. International Journal of Cardiology 2005; 152:e37–e39.
10. Bonnici K, Stanworth D, Simmonds MSJ, Mukherjee E, Ferner RE. Flowers of evil. Lancet 2010; 376:1616.
11. Imazio M, Belli R, Pomari F, Cecchi E, Chinaglia A, Gaschino G, etal., Malignant Ventricular Arrhythmias due to Aconitum napellus Seeds.Circulation.2000;102:29 07-08
12. Panda AK, Debnath SK. Overdose effect of aconite containing Ayurvedic Medicine ('Mahashankha Vati'). International Journal of Ayurveda Research 2010 Jul-Sep;1(3):183–186.
13. Wang X, Cao Y, Dong Y, Wang C, The Toxicity of Aconitum ferox Vahl.: A Comprehensive Review, Front Pharmacol. 2019 Sep 24; 10:1027. doi: 10.3389/fphar.2019.01027. PMID: 31619994; PMCID: PMC6771373.
14. Kaviraj Atridev Gupta, AshtangaSangraha by Acharya Vagbhat, edition,Varanasi, krishnadas Academy, 1993; p. 385; 48/2
15. Regmi, Prerok; Ravikrishna, S.; Bhat, Sudhakar; Hebbar, Chaithra; Jasphin, Shiny; Subacute Toxicity of Vatsanabha (*Aconitum ferox*, Ranunculaceae) Collected from Different Regions of India and Nepal. Journal of Ayurveda, Apr–Jun 2021, 15(2): 75-83.
16. Rakshith N N, Shrinidhi. R, Sudhakara Bhat and Chaithra Hebbar; Experimental Study on Acute Oral Toxicity of Vatsanabha (Aconitum ferox wall. ex Seringe) and Antidote Effect of Sweta Chitraka (Plumbago zeylanica Linn.) Patra Swarasa W.S.R to Haematological Parameters in Vatsanabha Induced Poison. International Journal of Ayurveda and Pharmaceutical Chemistry; 2020 ; Volume 13, Issue 3; 213-219