Journal of Ethnopharmacology 205 (2017) 138–146

Contents lists available at ScienceDirect

Journal of Ethnopharmacology

journal homepage: www.elsevier.com/locate/jep

Evaluation of processed borax as antidote for aconite poisoning

Prasanta Kumar Sarkar[a,*], Pradeep K. Prajapati[b], Vinay J. Shukla[b], Basavaiah Ravishankar[c]

[a] J.B. Roy State Ayurvedic Medical College and Hospital, West Bengal University of Health Sciences, 170-172, Raja Dinendra Street, Kolkata 700004, West Bengal, India
[b] Institute for Post Graduate Teaching and Research in Ayurveda, Gujarat Ayurved University, Jamnagar 361008, Gujarat, India
[c] S.D.M. Research Centre for Ayurveda and Allied Sciences, S.D.M. College of Ayurveda, Kuthpady, Udupi, Karnataka, India

ARTICLE INFO

Keywords:
Aconite
Acute protection
Borax
Cardiac activity
Neuro-muscular activity
Sub-acute protection

ABTBRACT

Ethnopharmacological relevance: Aconite root is very poisonous; causes cardiac arrhythmias, ventricular fibrillation and ventricular tachycardia. There is no specific antidote for aconite poisoning. In Ayurveda, dehydrated borax is mentioned for management of aconite poisoning.

Aim of the study: The investigation evaluated antidotal effect of processed borax against acute and sub-acute toxicity, cardiac toxicity and neuro-muscular toxicity caused by raw aconite.

Materials and methods: For acute protection Study, single dose of toxicant (35 mg/kg) and test drug (22.5 mg/kg and 112.5 mg/kg) was administered orally, and then 24 h survival of animals was observed. The schedule was continued for 30 days in sub-acute protection Study with daily doses of toxicant (6.25 mg/kg), test drug (22.5 mg/kg and 112.5 mg/kg) and vehicle. Hematological and biochemical tests of blood and serum, histopathology of vital organs were carried out. The cardiac activity Study was continued for 30 days with daily doses of toxicant (6.25 mg/kg), test drug (22.5 mg/kg), processed borax solution (22.5 mg/kg) and vehicle; ECG was taken after 1 h of drug administration on 1^{TB}, 15th and on 30th day. For neuro-muscular activity Study, the leech dorsal muscle response to 2.5 μg of acetylcholine followed by response of toxicant at 25 μg and 50 μg doses and then response of test drug at 25 μg dose were recorded.

Results: Protection index indicates that treated borax gave protection to 50% rats exposed to the lethal dose of toxicant in acute protection Study. Most of the changes in hematological, biochemical parameters and histopathological Study induced by the toxicant in sub-acute protection Study were reversed significantly by the test drug treatment. The ventricular premature beat and ventricular tachyarrhythmia caused by the toxicant were reversed by the test drug indicate reversal of toxicant induced cardio-toxicity. The acetylcholine induced contractions in leech muscle were inhibited by toxicant and it was reversed by test drug treatment.

Conclusion: The processed borax solution is found as an effective protective agent to acute and sub-acute aconite poisoning, and aconite induced cardiac and neuro-muscular toxicity. Processed borax at therapeutic dose (22.5 mg/kg) has shown better antidotal activity profile than five times more than therapeutic dose (112.5 mg/kg).

1. Introduction

Antidotes are the remedies to counteract the effects of poison. They are used because the poison may not have been completely removed by emesis or lavage or the poison is already absorbed or it has been administered by route other than ingestion. In popular fiction, miraculous properties are attributed to antidotes. They are usually projected as 'magic bullets', which fly to heart of the problems, affecting a dramatic recovery to the patients. This myth is still perpetuated. It is true that there are a few poisons for which genuine antidote exists, but the vast majorities unfortunately are without such magic remedies, aconite is one of them. Still there is no specific antidote for aconite poisoning in modern medicine (So et al., 2000; Paudel et al., 2008).

Aconite root is very poisonous. It is reported to have a depressant action on mammalian heart. It stimulates the vagus centre and slows the heart rate. It induces cardiac arrhythmias, ventricular fibrillation and ventricular tachycardia (So et al., 2000; Tai et al., 1992; Lin et al., 2004). It has a characteristic action on all the sensory nerves, at first the nerve endings are stimulated with the characteristic prickling and tingling sensation. After a short time the action is reversed and the

Abbreviations: ECG, Electrocardiogram; PSS, Physiological salt solution; SGOT, Serum glutamic oxaloacetic transaminase; SGPT, Serum glutamic-pyruvic transaminase

* Correspondent author.
E-mail address: prasantaksarkar@yahoo.com (P.K. Sarkar).

http://dx.doi.org/10.1016/j.jep.2017.04.014
Received 29 December 2016; Accepted 18 April 2017
Available online 26 April 2017
0378-8741/

nerve endings are paralyzed and symptoms like numbness occurs (Dzhakhangirov and Bessonova, 2002; Salgado and Saar, 2004).

Aconite is used in Ayurvedic system of medicine as a therapeutic entity after proper detoxification (*Shodhana*) treatment. Improperly treated aconite may cause various adverse effects in the body. Aconite poisoning due to ingestion of Ayurvedic medicines containing aconite is reported (Panda and Debnath, 2010). Many remedies to treat the patients of aconite poisoning are mentioned in the Ayurvedic system of medicine. One of such remedies contains processed borax and ghee (Shastri, 1995). Processed borax is also mentioned to be used with treated aconite for therapeutic uses and to detoxify the poisonous effect of aconite (Mishra, 1999). Processed borax is included in most of the formulations of aconite as an ingredient. The present Study was planned to evaluate processed borax as antidote for acute and sub-acute exposure to aconite poisoning.

In this Study, cardiac activity of raw aconite has been evaluated after sub-acute exposure (30 days) in albino rats. The cardiac activity of processed borax has been evaluated in the rats treated by raw aconite to observe any protection given by processed borax to the rats exposed to raw aconite.

The effect of antagonism can be assayed on isolated preparations. The neuro-muscular protection activity of processed borax has been evaluated on the raw aconite treated leech dorsal muscle to observe any antagonism effect produced by processed borax to muscle exposed to raw aconite.

2. Materials and methods

2.1. Drugs

Raw aconite (*Aconitum chasmanthum* Holmes ex Stapf., dry root) was purchased from Saket Market, Rajasthan, India, and was identified by the botanist of Institute of Ayurvedic Medicinal Plant Sciences, Gujarat Ayurved University, Jamnagar. The transverse sections of the specimen were studied; powder microscopy was carried out and HPTLC profile of the plant material was determined. The voucher specimen was kept in the department of Rasashastra and Bhaishajya Kalpana, Institute for Post Graduate Teaching and Research in Ayurveda, Gujarat Ayurved University, Jamnagar, India (Voucher Specimen No. RSBK/182/08). The dry roots of raw aconite (RA) were made into fine powder form (mesh size 120), and drug suspension was prepared by adding 1 ml 5% gum acacia solution in 100 ml distilled water. The therapeutic dose of aconite mentioned in classics of Ayurveda is 8 mg to 16 mg/d (Shastri, 2000). In the present Study, human dose of aconite has been decided to be 15 mg/d. The dose for experimental Study of the drugs was 1.25 mg/kg, calculated by extrapolating the human dose to animal dose based on the body surface area ratio.

Raw borax was purchased from local market of Jamnagar, India. It was heated and water for crystallization was evaporated. The raw borax and the treated borax were analyzed physic-chemically; and X-ray diffraction pattern of the samples was determined. The specimens of the raw and treated borax were kept in the department of Rasashastra and Bhaishajya Kalpana, Institute for Post Graduate Teaching and Research in Ayurveda, Gujarat Ayurved University (Voucher Specimen No. RSBK/187/08). The treated borax (TB) was made into fine powder form, and drug solution was prepared by dissolving the powder in distilled water. The therapeutic dose of treated borax mentioned in classics of Ayurveda is 125 mg to 375 mg/d (Shastri, 2000). In the present study, human dose of aconite has been decided to be 250 mg/d. The dose for experimental Study of the drugs was 22.5 mg/kg, calculated by extrapolating the human dose to animal dose based on the body surface area ratio.

2.2. Animals

Charles Foster strain albino rats of either sex weighing between 180 g and 250 g were used for acute and sub-acute protection and cardiac protection Studies. They were obtained from the Animal House attached to the pharmacology laboratory of Institute for Post Graduate Teaching and Research in Ayurveda, Gujarat Ayurved University. They were housed in breeding cages at an ambient temperature with a natural day and night cycles. The animals had free access to Amrut brand rat pellet feed supplied by Pranav Agro Industries and tap water.

Leech (*Hirudo medicinalis*) was used in in vitro neuro-muscular protection Study. It was collected from the Pharmacology Laboratory of Institute for Post Graduate Teaching and Research in Ayurveda, Gujarat Ayurved University. The dorsal muscle of leech was dissected and isolated and used for the experiment.

2.3. Acute and sub-acute protection Study

2.3.1. Experimental design

This part of Study has been carried out in two phases, acute and sub-acute protection Study. Poisoning was induced by administration of RA suspension. In acute protection Study, protective activity of treated borax was determined against lethal dose of RA (35 mg/kg) in rats. The dose was determined from an initial pilot study (Sarkar et al., 2008). And in chronic protection Study, protective effect of treated borax (TB) was studied in two dose levels, therapeutic effective dose (TB1) and five times of therapeutic effective dose (TB5), in rats after sub-acute exposure (30 days) to higher dose of RA (6.75 mg/kg). Protection profile TB1 and TB5 has been compared.

2.3.2. Study protocol for acute protection Study

Total 24 Charles Foster rats of either sex weighing between 180 g and 250 g were taken and divided randomly into 4 groups, each containing 6 animals. The treatment schedule was as follows: group I comprised of vehicle (tap water) treated control animals, group II was designated as toxicant control group and received suspension of lethal dose (35 mg/kg) of RA. Animals of group III were treated with 35 mg/kg, suspension of RA and 22.5 mg/kg, solution of TB1; group IV animals were treated with 35 mg/kg, suspension of RA and 112.5 mg/kg, solution of TB5. Solutions of test drugs were prepared in two different concentrations i.e. 4.5 mg/ml and 22.5 mg/ml. Suspension of RA was prepared in 8 mg/ml concentration. Single dose of toxicant and drugs was administered orally, and then 24 h survival of animals was observed. The protection index (PI) was calculated using the following formula (Dube et al., 2000):

$$\mathrm{PI} = \frac{\text{Percentage of death in raw aconite + Antidote}}{\text{Percentage of death in raw aconite group}}$$

2.3.3. Study protocol for sub-acute protection Study

Total 24 Charles Foster rats of either sex weighing between 180 g and 250 g were taken and divided randomly into 4 groups, each containing 6 animals, 3 male and 3 female. The treatment schedule was as follows, group I comprised of vehicle (tap water) treated control animals, group II animals were treated with 6.75 mg/kg, suspension of RA, group III animals received 6.75 mg/kg, suspension of RA and 22.5 mg/kg, solution of TB1 and animals of group IV were treated with 6.75 mg/kg, suspension of RA and 112.5 mg/kg, solution of TB5. Suspension of RA was prepared in 1.35 mg/ml concentration. And solution of TB1 and TB5 were prepared in 4.5 mg/ml and 22.5 mg/ml concentrations respectively.

The schedule was continued for 30 days with daily doses of toxicant, test drugs and vehicle. Gross case behavior was observed throughout the Study period. On 31st day rats were sacrificed by stunning, blood was collected from jugular vein for hematological and biochemical tests, the vital organs were collected for the histopathological Study and