Original Research Paper | Ayurveda | Volume : 6 ¦ Issue : 9 ¦ Sep 2021 ¦ e-ISSN : 2456-5040

EFFICACY OF *NIMBAPATRA AGADA* IN CISPLATIN INDUCED HEMOTOXICITY IN MALE SWISS ALBINO MICE

Anjana M.[1], P. A. Aboobaker[2], Ramadasan Kuttan[3]

[1]Assistant Professor, Dept. of AgadTantra, Ahalia Ayurveda Medical College, Palakkad, India.
[2]Guide, Associate Professor, Dept.of AgadTantra, V P S V Ayurveda College, Kottakkal, India.
[3]Co-guide, Research Director, Amala Cancer Research Centre, Trissur, India.

ABSTRACT

In the ecology of Kerala, vipers dominate over all the species of poisonous snakes. Viper venom has more hemotoxic contents. *Nimbapatra Agada* (Neem leaf paste and milk in ratio 1:5) mentioned in *viṣavaidya jyotsnika- maṇḍali viṣa sāmānya cikitsa prakaraṇa* in hematemesis due to viper envenomation. Traditional *viṣa vaidyās* in Kerala practicing this formulation in bleeding conditions due to viper bite. Cisplatin, a potent chemotherapeutic drug causing myelosuppression.

(hemotoxicity) nephrotoxicity and oxidative stress. If the efficacy of *Agada* proven by reverse pharmacology, it can be used in chemotherapy induced toxicity. So an experimental study has been planned to prove the efficacy of *Nimbapatra Agada* in Cisplatin induced Hemotoxicity.

KEY WORDS: Viper snake venom , Cisplatin, *Nimbapatra Agada,* Hemotoxicity.

INTRODUCTION:

Among 24 neglected tropical diseases (NTDs) and conditions listed by WHO, snakebite is among the top killers. Tens of thousands of people die each year as a result of snakebite envenoming; with close to 50,000 deaths in India alone.[1] Viper snake venom is hemotoxic in nature which affect cardiovascular system. This hemotoxic venom typically acts to lower blood pressure and blood clotting. It may also attack the heart muscle with the goal of causing death.[2] Coagulopathy is the commonest important, systemic clinical syndrome caused by snake envenoming in the world, and Venom-Induced Consumption Coagulopathy (VICC) is the most clinically important coagulopathy, because it can be complicated by serious and life-threatening haemorrhage . Venom-Induced Consumption Coagulopathy (VICC) is the commonest coagulopathy resulting from snakebite and occurs in envenoming by Viperid snakes, certain elapids, including Australian elapids, and a few Colubrid (rear fang) snakes. More recently, the term "venom-induced consumption coagulopathy" has been introduced because it provides a more general description of the coagulopathy.[3] Toxicology, known as *Agadatantra* in classical Ayurvedic treatises, contains clinical as well as forensic branches of toxicology; contrary to the specializations in modern medicine. The clinical toxicology of Ayurveda has a wide range of topics dealing with various types of poisons of animal, vegetable, and mineral origin. The effect of *Agada* mentioned in *Viṣacikitsa granthas* need to be evaluated for their capabilities to be adopted in the diversified field of toxicology. Cisplatin is a potent chemotherapeutic agent having proved efficacy against solid tumor[4], acting as cytotoxic agent. This benefit is however marked by its profound toxicity to multiple system of the body.[5] It is a very potent nephrotoxic agent which causes myelosuppression. There are also reports of this toxicity to gastrointestinal tract and central nervous system.[6] In this study Cisplatin was used to induce hemotoxicty. In order to provide relief to such patients who are on Cisplatin medication and its toxic effects, *Nimbpatra Agada* mentioned in *viṣavaidya jyotsnika- maṇḍali viṣa sāmānya cikitsa prakaraṇa* is being used in this study. *Nimbapatra* (*Azadirachta indica A.Juss*) *Kalka*(paste) with milk mentioned as an *Agada* in case of frank bleeding from gum and hematemesis due to viper envenomation.[7]

MATERIALS AND METHODS:

Nimbapatra Agada is a combination of *Nimbapatra Kalka* (*Azadirachta indica A. Juss* paste) and milk (*Kṣīra)* in the ratio of 1:5 (W/V). So the non toxic concentration of *Nimbapatra Agada* was calculated by using by extrapolating the therapeutic dose to mice dose on the basis ofbody surface area ratio by referring to the table of Pagets and Barnes, 1964. As per Ayurvedic texts, the normal human dose of *Kalka* is one *Karsha* (12gm). Based on this converted animal dose was administered in three doses, Lower dose, Medium dose and Higher doses receiving 50mg/kg b.wt, 100mg/kg b.wt,and 200mg/kg b.wt respectively.

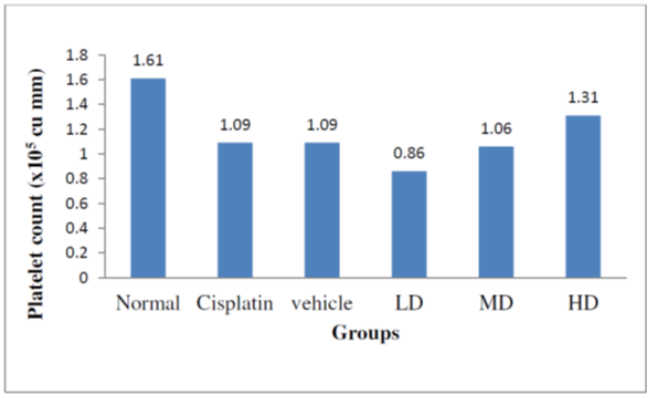
The Male Swiss Albino Mice (5-6 weeks old, 20-30 gm b. wt.) were used for conducting the study, which were randomly divided into 6 groups (6 animals in each group).

Groups	Functions
Group 1	Normal; control
Group 2	Cisplatin at a dose of 16mg/kg body weight, i.p
Group 3	Vehicle + Cisplatin at a dose of 16mg/kg b.wt i.p
Group 4	Nimbapatra Agada 50mg/kg b.wt (LD)+ Cisplatin at a dose of 16mg/kg b.wt i.p
Group 5	Nimbapatra Agada 100mg/kg b.wt (MD)+ Cisplatin at a dose of 16mg/kg b.wt i.p
Group 6	Nimbapatra Agada 200mg/kg b.wt (HD)+ Cisplatin at a dose of 16mg/kg b.wt, i.p

The animals in group- 1 were kept as Normal (without any treatment). The animals in group 2 were treated Cisplatin (intra peritoneal injection(i.p) alone. Animals in 3rd group were kept as vehicle group, ie., milk was administered as vehicle. The animals in group 4 to - group 6 were treated with *Nimbapatra Agada* (oral) in lower dose (50mg/kg b.wt), medium dose (100mg/kg b.wt) and higher dose (200mg/kg b.wt) respectively for 5 days. On the fifth day animal in the groups 2 – group 6 were administered with a single dose of Cisplatin as intraperitoneally (i.p). Oral administration of *Agada* was continued for 3 days. Animals in all groups were euthanized 72hours (3 days) after Cisplatin administration.

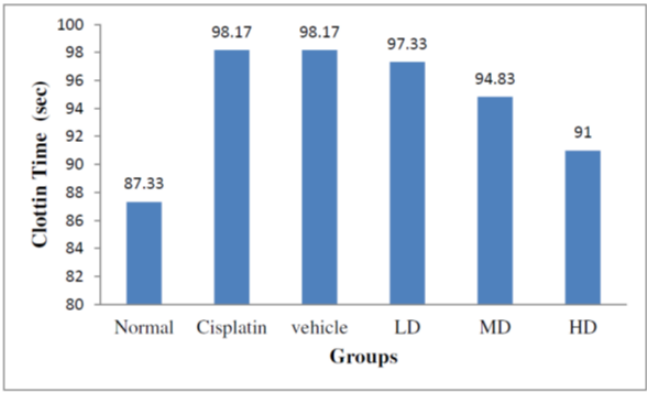
RESULTS AND DISCUSSION:

In animal experimental study, Hemoprotective, activity of *Nimbapatra Agada* were carried out by assessing hematological parameter . The primary function of bone marrow is hematopoiesis, the synthesis of red blood cells, platelets, and white blood cells.[8] Platelets plays an important role in blood clotting. Cisplatin causes a destruction of bone marrow results drastic reduction of platelet count. [9] Cisplatin induced reduction in total platelet count was significantly increased ($P<0.01$) by administering the higher dose of *Nimbapatra Agada* (200mg/kg b wt.). The increased platelet count may be related to the reported constituents of the neem leaf (flavonoids and quercetin), that have been shown to have hematopoietic properties.[10] Cisplatin induced elevation in clotting time was reduced in the higher dose of *Nimbapatra Agada* (200mg/kg b.wt) treated groups showed a significant reduction in clotting time ($P<0.01$). The leaves of *Azadirachta indica* enhance the production of IgM, and IgG, and having a immunomodulatory action.[11] *Nimbapatra Agada* is the combination of *Nimbapatra and kṣīra. Nimba* is a *tikta rasa pradhāna* drug. By this it will alleviate *pitta* and *kapha doṣas*. The leaves and bark of *nimba* possesess *vraṇapācana* and *vraṇaśodhana* property. *Tikta rasa* of *nimba* may be reducing the *dravatva* of *pitta* which included in the primary stage, in the pathogenesis of *raktapitta*. It will purify *rakta by* its *raktaśodhaka* property and will reduce the *śopha* produced due to *raktavikāras*. It is having *śīta vīrya* which will helps to stop the bleeding from various orifices.[12] Due to these properties *nimba* can be used in various *rakta vikāras*. *Kṣīra* is having *madhura rasa* and *vipāka*, *guru guna* and *śīta vīrya,* alleviates *vāta* and *pitta doṣās*.[20] *Kṣīra* is indicated in various bleeding conditions.



Group-1 Normal, Group-2 Cisplatin alone, Group-3 vehicle + Cisplatin
Group-4 LD-lower dose(50mg/kg b.wt) + Cisplatin, Group-5 MD- medium dose (100mg/kg b.wt)+ Cisplatin , Group-6 HD- higher dose (200mg/kg b.wt) + Cisplatin

Fig. 1: Effect of *Nimbapatra Agada* on platelet count in cisplatin treated animals



Group-1 Normal, Group-2 Cisplatin alone, Group-3 vehicle + Cisplatin
Group-4 LD-lower dose (50mg/kg b.wt) + Cisplatin, Group-5 MD- medium dose (100mg/kg b.wt)+ Cisplatin , Group-6 HD- higher dose (200mg/kg b.wt) + Cisplatin

Fig. 2: Effect of *Nimbapatra Agada* on clotting time in cisplatin treated animals

CONCLUSION:

Trial drug is the combination of *Nimbapatra* (*Azadirachta indica A.Juss*) *Kalka* and milk in the ratio 1:5(Weight/Volume) mentioned as an *Agada* in case of frank bleeding from gum and hematemesis due to viper envenomation. In this study, only higher dose of *Nimbapatra Agada* (200mg/kg b.wt) is effective in total platelet count and in clotting time. So the trial can be used in various bleeding disorders with altered platelet count and clotting time.

REFERENCES:

I. Alirol E, Lechevalier P, Zamatto F, Chappuis F, Alcoba G, Potet J ntivenoms for Snakebite Envenoming: What Is in the Research Pipeline, PLoS Negl Trop Dis 2015 9(9): doi:10.1371/journal.pntd.0003896 Available http://journals.plos.org/plos-ntds/article?id=10.1371/journal.pntd.0003896

II. http://www.wisegeek.org/how-does-snake-venom-affect-the-human-body.htm

III. Kalana Maduwage and Geoffrey K Isbister. Current Treatment for Venom- Induced Consumption Coagulopathy Resulting from Snakebite. PLoS Negl Trop Dis. 2014 Oct; 8(10): doi: 10.1371/journal.pntd.0003220 Available - http://www.ncbi.nlm.nih.gov/pmc/articles/PMC4207661/ Published online 2014 Oct 23.

IV. Prestavko W D' Asoust JC Issel BF, and crooke ST Cisplatin (cisdiamminedichloroplatinum) cancer Treat Rev 1979; 6; 17-39

V. Zahid H- Cisplatin mode of action and molecular basis of resistance. Oncogene 2003; 22: 7265-7279

VI. Taguchi T, Nazneen A, Abid MR, and Razzaque MS. Cispatin associated nephrotoxicity and pathological events, contrib. Nephrol 2005; 148:107-121

VII. Narayana, Viṣavaidya jyotsnika K Mahadeva Sasthri editor, Sri Vanci Setu Laksmi Granthavali series,Printed and published as per the order of Sree Chithirathirunal Maharaja,TVM,1958 sloka.28 Adhikaranam-6 p.29

VIII. Domen J, Wagers A and Weissman I, Bone marrow (Hemopoietic stem cells, p-14 available http://stemcells.nih.gov/staticresources/info/scireport/PDFs/D.%20CHAPTER%202.pdf

IX. Monica Bartucci et al., Prevention of Chemotherapy-Induced Anemia and Thrombocytopenia by Constant Administration of Stem Cell Factor Clin Cancer Res Oct1, 2011; 17; 6185. doi: 10.1158/1078-0432.CCR-11-1232 available-http://clincancerres.aacrjournals.org/content/17/19/6185.full

X. Raja SB Kumar NK, Devaraj SN. Isolation and partial characterisation of a novel lectin from aegle marmelos fruit and its effect on adherence and invasion of shigellae to HT29 cells. PloS One 2011;6:e16231

XI. K mukhaerjee et al immunomodulatory leads from medicinal plants, Indian journal of tradition knowledge. 2014 vol.13,(2) p 235-236

XII. Sharma PV, Dravyaguna-Vijnana, Choukhambha Bharati Academy Varanasi, reprint 2005, vol-2, 2/57, p-149-152.

XIII. Vagbhata, Ashtanga Hrdayam, Murthy KRS (trans.), Chowkhamba Krishnadas Academy Varanasi, Reprint 2006, vol-1, Sutrasthana, 5/20-21,p.58