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EVALUATION OF THE HEPATOPROTECTIVE ACTIVITY OF FORMULATION CONTAINING ANDROGRAPHOLIDE ON HEPATOTOXICITY INDUCED ALBINO WISTAR RAT

MAJUMDAR M*, FAZAL M, BISWAS DS, GHOSH T, CHATTERJEE A, ROY P, ROY S AND PAUL S

Department of Pharmacology, Netaji Subhas Chandra Bose Institute of Pharmacy, Chakdaha, Nadia, 741222, West Bengal, India

***Corresponding Author: Dr. Mrityunjoy Majumdar: E Mail: majumdar777@gmail.com**

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ABSTRACT

Every plant on earth has been used for humans and other animals, according to ancient Indian literature. The primary bodily organ that controls homeostasis is the liver. It is engaged in practically all metabolic pathways that are connected to growth, the prevention of disease, the delivery of nutrients, the production of energy, and reproduction. *Andrographis paniculata* is a plant that is widely cultivated, and as more evidence emerges in favour of its several therapeutic benefits, its significance as a medicinal plant is increasing. With careful consideration of the plant's beneficial properties, it can be recommended as a secure, crucial medicinal herb for people. Numerous chemical components, primarily lactones, diterpenoids, diterpene glycosides, flavonoids, and flavonoid glycosides, are present in this herb. The main objective of this experiment was to find out the hepatoprotective activity of the formulation which is made up of *Andrographis paniculata* in albino wistar rats by investigating the various parameters like body growth and the liver function tests like Serum Glutamic Oxaloacetic Transaminase (SGOT), Serum Glutamic Pyruvic Transaminase (SGPT), and Alanine Transaminase (ALT) on Carbon tetrachloride (CCl_4) induced experimental model.

Keywords: *Andrographis paniculata*, Andrographolide, Silymarin, Carbon tetrachloride, Alcohol, Hepatoprotective

INTRODUCTION:

Hepatic disorders, a primary cause of death, have become more prevalent in many developing countries. The use of herbal product-based remedies is very much accepted for such hepatic disorders. For this, medicinal herbs have been used and some facilitated the discovery of active compounds which helped the new synthetic drug development against hepatic diseases **[1]**.

Andrographis paniculata is an important herbal agent to treat various disease, the physiological effects of *Andrographis paniculata* shows due to its various biologically active ingredients (Neoandrographolide, Panicoline, Paniculide), and their potential mechanisms of action. Some of the most important active substances are andrographins and andrographolide, the actions of which reduce bilirubin and normalize the levels of relevant serum enzymes indicative of liver pathophysiological conditions **[2]**. *A. paniculata* (*Andrographis paniculata*) has a wide range of benefits, mostly related to immune system-enhancing properties. In native India, the traditional use has primarily included supporting the immune system. During the twentieth century, use of *Andrographis paniculata* spread outside India to Europe and beyond **[3]**. *A. paniculata* act as an immune support ingredient, this is one of the most popular herbs that's related to the herb that has the ability to restore normal balance in the bacterial flora of the gut **[4]**.

Benefits of *Andrographis paniculata* **[5]**

- Enhances immune function
- Restoration of normal bacterial flora of the gut
- Supportive liver function
- Enhanced bile flow

Other than the Acanthaceae family there are other families of the plant which provide liver protective activity for example; Euphorbiaceae (*Phyllanthus niruri*), Asteraceae (*Silybum marianum*), Combretaceae (*Terminalia chebula*) and etc. **[6]**. *Andrographis paniculata* (A.P) showed protective effects in the activity of superoxide dismutase, catalase, glutathione peroxidase, and glutathione reductase, as well as the level of glutathione **[7]**. The major component of A.P is andrographolide and andrographin **[8]**. All Acanthaceae family previously showed a greater response in hepatoprotective activity and thus this is our targeted drug. Therefore, our study aimed to analyse the effects of a formulation containing andrographolide that is found from *Andrographis paniculata* or well known as kalmegh, on liver protection, and a comparative study was also executed to check the efficacy of formulation containing andrographolide against the standard drug Silymarin (100mg).

MATERIALS AND METHODS:

Materials:

Amar Pvt. Ltd. provided an *Andrographis paniculata* extract that contained 10% w/w andrographolide (as determined by IP assay method). Neo Science, Kolkata, donated choline chloride. From Erba Mannheim (Parent organization-Transasia Bio-Medicals LTD), SGOT, SGPT, and ALP kits were procured. Silymarin was procured from Micro Lab.

Method:

In accordance with the formula shown in **Table 1**, a hepatoprotective formulation was developed by dissolving an extract of *Andrographis paniculata* that contains 10% andrographolide (by IP Assay method) as well as choline chloride.

Table 1: Formula of *Andrographis paniculata* extract containing 10% andrographolide (by IP Assay method) and choline chloride

Ingredient	Amount for 100ml	Amount in a dose of 0.5ml hepatoprotective formulation
Andrographis paniculata extract (contains 10% andrographolide)	1.6 gm	8 mg
Choline chloride	9.4 gm	47 mg
Water up to	100 ml	0.5 ml

Animal and treatment:

Observations were executed according to the OECD guidelines. Thirty young albino wistar rats of mixed gender were purchased from CPCSEA authorized breeder and supervised under controlled conditions. Adult albino wistar rats, weighing 150g-200g, were used in this research. The rats were augmented with a reverse natural light-dark cycle and temperature of 22 ± 2°C, a relative humidity of approximately 55%, as practiced in the Animal House of Netaji Subhas Chandra Bose Institute of Pharmacy. All the rats were maintained on standard rat pallets and tap water ad libitum. They were acclimated to laboratory conditions for 1 week before starting treatment. Their condition of health was also monitored during this period. Only healthy animals showing no clinical signs were used for the study. One day before the onset of treatment, rats were assigned to treatment groups (n=6 each) by a randomized procedure in a way that mean body weights were identical in all groups.

Experimental model: [9, 10]

CCl_4 induced model: Total 30 no. of adult rats were selected and divided into 5 groups: Group 1- Untreated, Group 2- CCl_4, Group 3- Silymarin (100mg/kg) + CCl_4, Group 4- AP (311mg/kg) + CCl_4, Group 5- AP (311mg/kg) p.o,

Group 2, 3, 4 received CCl_4 (0.5ml/kg), p.o for a period of 15 days. The body weights of all the rats were measured regularly.

Alcohol induced model: Group 1- Untreated, Group 2- Alcohol, Group 3- Silymarin (100mg/kg) + Alcohol, Group 4-

AP (311mg/kg) + Alcohol, Group 5- AP (311mg/kg) p.o,

Group 2, 3, 4 received Alcohol (3.75ml/kg), p.o for a period of 25 days. The body weights of all the rats were measured regularly.

Thiopentone sodium induced sleeping time: Thiopentone sodium (40mg/kg, ip) was administered on the sixteenth day. Twelve hours prior to the administration of thiopentone sodium, food was withheld. The loss of the righting reflex in the rats was observed. Thiopentone sodium sleeping time was determined to be the period of time between the loss and recovery of the righting reflex. This parameter was used to examine the liver's metabolic activity.

Marker enzymes of liver damage: 1 ml of blood was drawn from each rat using the retro-orbital technique after 24 hours of thiopentone sodium-induced sleep time, and it was then allowed to coagulate for 30 minutes at 37°C. Centrifugation was used to separate the serum for 15 minutes at a speed of 2500 rpm, and SGPT, SGOT, and ALT levels were measured.

RESULT AND DISCUSSION:

Untreated animals and AP treated group has shown all the reading like SGOT, SGPT, ALP in the normal range, CCl_4 used to damage the liver and all the test of lever function reading have proved it. The Group that received silymarin along with CCl_4 and A.P has shown moderate improvement in liver functions compared with only CCl_4 treated group. In the A.P treated group the increase in body weight was even better than the untreated group, but the other 3 groups have shown negative growth compared with the untreated control.

Table 2: Effect of Andrographolide of *Andrographis paniculata* on Thiopentone sodium Induced sleeping time, body weight, SGOT, SGPT, ALP

Treatment	Thiopentane Sodium Induced Sleeping time	Increased body weight (%)	SGOT	SGPT	ALP
Untreated	42 ± 1.58	20%	50.11 ± 1.843	41.22 ± 0.441	83.11±1.416
CCl_4	101 ± 3.27	-11%+++	162.70 ± 0.0697	196.77 ± 373	153.33±1.015
CCl_4+ Silymarin	81±2.08***	5%+++	80.00 ± 1.843***	166.52±4.603***	121.11±1.211***
CCl_4+ AP	72±6.57***	2%+++	101.44±3.367***	136.81±2.123***	101.24 ± 1.416
AP	46±1.99***	27%	36.98 ± 0.323***	39.73 ± 1.213**	89.34 ± 1.821

All values are mean ± SEM, n=6, p*<0.001 while compared with alcohol treated Group, p+++<0.001, p+<0.05 while compared with untreated control**

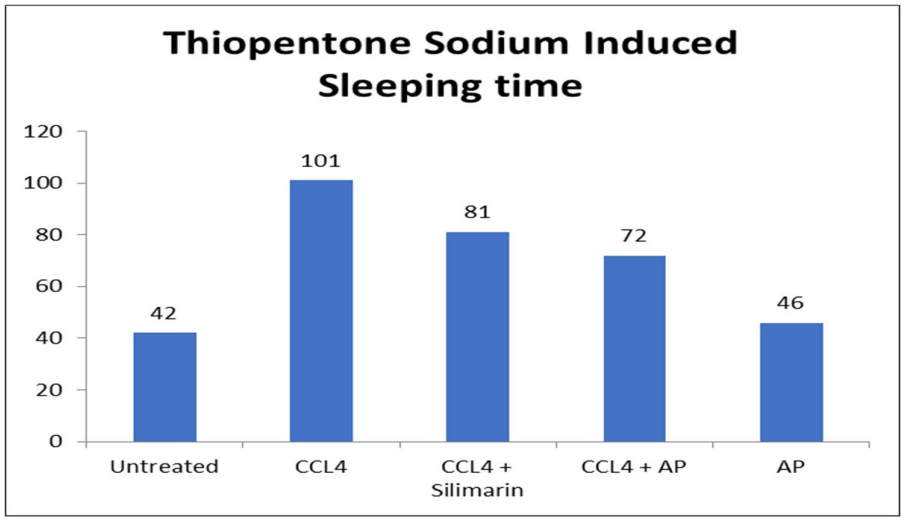

Figure 1: Effect of Andrographolide of *Andrographis paniculata* on Thiopentone sodium Induced sleeping time

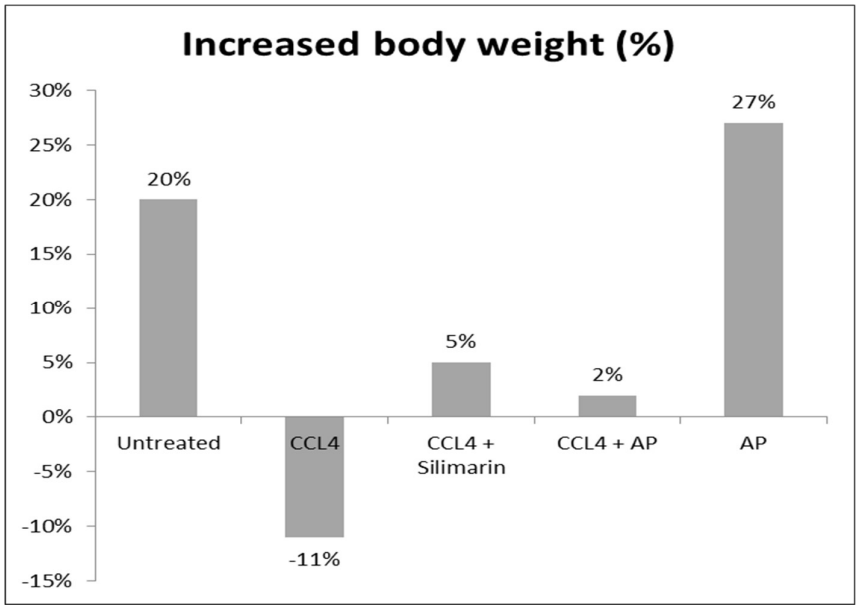

Figure 2: Effect of Andrographolide of *Andrographis paniculata* on increased body weight

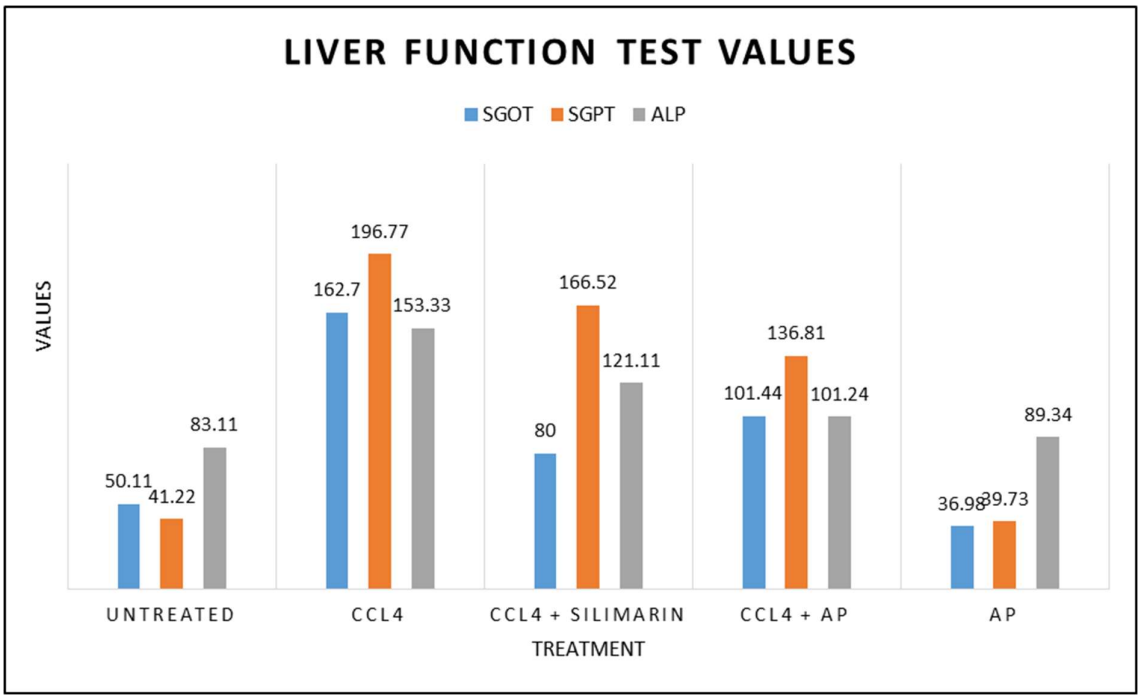

Figure 3: Effect of Andrographolide of *Andrographis paniculata* on liver function test values

Alcohol Induced Model:

Table 3: Effect of Andrographolide of *Andrographis paniculata* in case of alcohol induced model on Thiopentone sodium Induced sleeping time, body weight, SGOT, SGPT, ALP.

Treatment	Sleeping time	Sgot	Sgpt	Alp	Increase in body weight
Control	42±1.58	50.11±1.843	41.22±0.441	83.11±1.416	20%
Alcohol	98±2.44***	133.8±2.167***	172±9.11***	140.0±2.17***	-8.5%+++
Alcohol + sil	75±1.51***	88.2±1.767***	160.29±0.25	111.91±0.56***	5%+++
Alcohol+ ap	71±4.21***	96.2±3.133	143.12±2.71***	114.51±0.73***	8%+++
Ap	39±2.01***	47.21±0.395	45.31±3.13***	78.21±5.21***	32%+++

All values are mean ± SEM, n=6, p*<0.001 while compared with alcohol treated Group, p+++<0.001, p+<0.05 while compared with untreated control**

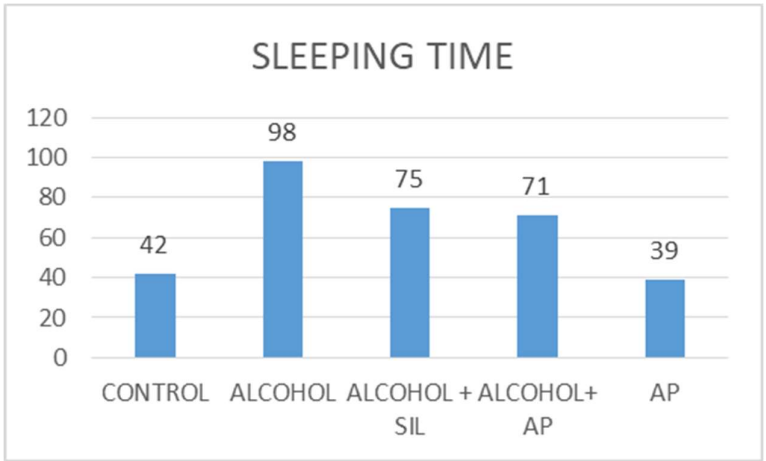

Figure 4: Effect of Andrographolide of *Andrographis paniculata* in case of alcohol induced model on Thiopentone sodium Induced sleeping time

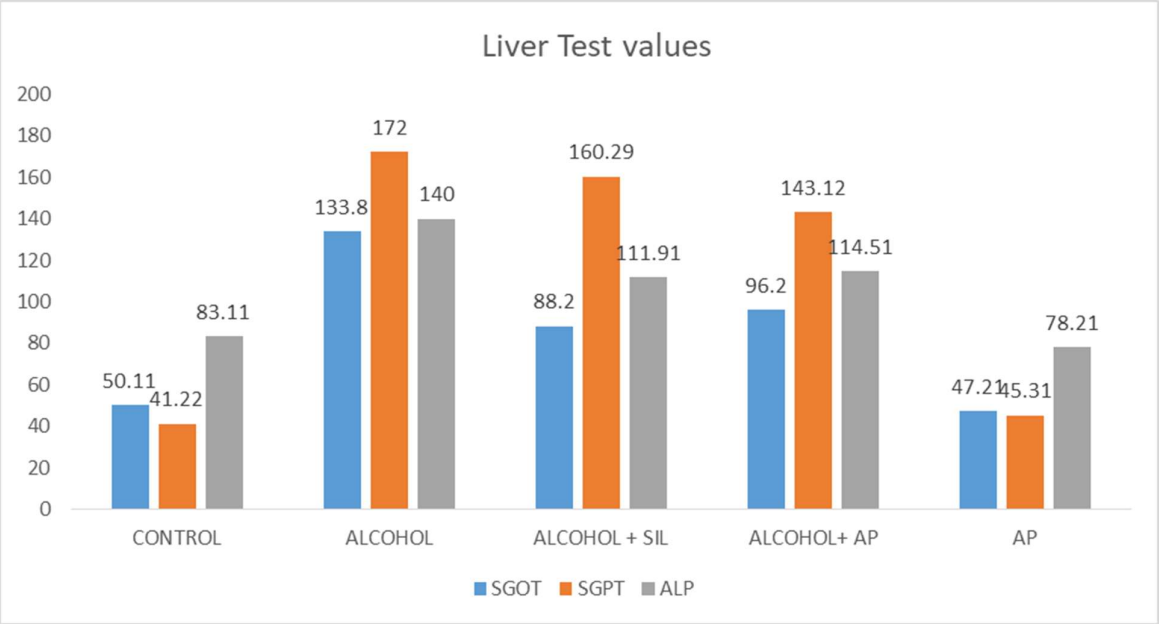

Figure 5: Effect of Andrographolide of *Andrographis paniculata* in case of alcohol induced model on liver

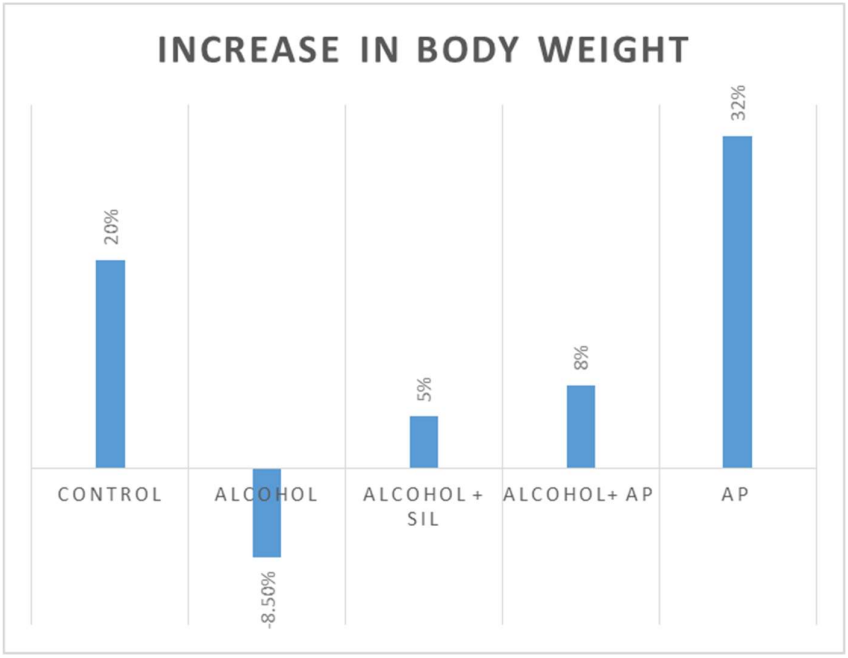

Figure 6: Effect of Andrographolide of *Andrographis paniculata* in case of alcohol induced model on body weight

CONCLUSION:

After performing this experiment, we came to a conclusion that herbal formulation processed *kalmegh* herb made from *kalmegh* IP (*Andrographis paniculata*), containing andrographolide 10% by IP assay, supplied by Amar Pvt limited Indore, and A.P. has shown significant improvement in liver functions and also increased the growth rate in an animal. This formulation can be used as a regular medicine in the poultry industry for

commercial benefit as well as for the health of mankind is a concern.

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REFERENCES:

[1] Rangnathrao TS, Shanmugasundaram P. Antioxidant and Hepatoprotective activity of Ehretia laevis Roxb against paracetamol induced acute Hepatotoxicityin wistar rats. Research Journal of Pharmacy and Technology. 2019 Dec 30;12(12):6143-8.

[2] Zachariah SM, Aleykutty N, Viswanad V, Halima OA. An overview on hepatoprotective activity of natural products. Research Journal of Pharmacy and Technology. 2012 Mar 1;5(3):317.

[3] Jannathul Firdous, Norain Ab. Latif, Resni Mona, Rehanna Mansor, Noorzaid Muhamad. Andrographis paniculata and its Endophytes: A Review on their Pharmacological Activities. Research J. Pharm. and Tech 2020; 13(4): 2027-2030. doi: 10.5958/0974-360X.2020.00365.0

[4] Andrographis. Vital Plan. 2018 [cited 22 May 2018]. Available from: https://vitalplan.com/ingredients/andrographis?gclid=Cj0KCQjwuYTYBRDsARIsAJnrUXBCCs3LEEUoQcIUe9Fkpztvmy18B7wOkML3vVkT4VvwzzFXPTugztkaArFrEALw_wcB

[5] Avbunudiogba John Afokoghene, Okafo Sinodukoo Eziuzo, Nwobi Chidimma Lynda. Antimicrobial investigation, Formulation and Evaluation of Andrographis paniculata aqueous herbal cream for topical application. Research Journal of Pharmacy and Technology. 2022; 15(8):3553-8. doi: 10.52711/0974-360X.2022.00596

[6] Home - PMC - NCBI [Internet]. Ncbi.nlm.nih.gov. 2018 [cited 22 May 2018]. Available from: https://www.ncbi.nlm.nih.gov/pmc/

[7] Vetriselvan S, Victor R, Parimala D, Arun G. Comparative evaluation of hepatoprotective activity of andrographis paniculata and Silymarin in ethanol induced hepatotoxicity in albino wistar rats. Der Pharmacia Lettre. 2010; 2(6):52-9.

[8] Tumewu L, Pamungkas IR, Ilmi H, Hafid AF, Tantular IS, Wahyuni TS, Widyawaruyanti A. The role of

andrographolide in Andrographis paniculata as a potential analgesic for herbal medicine based drug development. Research Journal of Pharmacy and Technology. 2021 Dec 28;14(12):6269-74. doi: 10.52711/0974-360X.2021.01084

[9] Anusha M, Venkateswarlu M, Prabhakaran V, Taj SS, Kumari BP, Ranganayakulu D. Hepatoprotective activity of aqueous extract of Portulacaoleracea in combination with lycopene in rats. Indian journal of pharmacology. 2011 Sep;43(5):563.

[10] Achliya GS, Wadodkar SG, Dorle AK. Evaluation of hepatoprotective effect of AmalkadiGhrita against carbon tetrachloride-induced hepatic damage in rats. Journal of Ethnopharmacology. 2004 Feb 1;90(2-3):229-32.