



HEPATOPROTECTIVE EFFECT OF ASPARAGUS RACEMOSUS AQUEOUS EXTRACT ON PARACETAMOL INDUCED STRESS IN THE FISH ANABAS TESTUDINEUS

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ABSTRACT

Present study clearly demonstrates the ameliorating effects of *Asparagus racemosus* extract on hepatic injury caused by repeated paracetamol dosing in fishes. Histopathological observations and quantitative estimation of serum alkaline phosphatase activity were used as indices for studying the damage caused by paracetamol and the ameliorative effect of *Asparagus racemosus*. Repeated administration of paracetamol caused a sharp increase in the serum alkaline phosphatase activity as indicated by the high values in treated group of fishes in comparison with control group. Histopathological study also revealed altered liver architecture as evidence by swollen aopptotic hepatocytes with coarse granular cytoplasm and compressed sinusoids. Severe lymphocytic infiltration was also noticed. In *Asparagus* treated group liver architecture and alkaline phosphatase activity were normal. When the paracetamol injected group was treated with *Asparagus* extract there was a significant decline in elevated ALP level. The hepatic tissue architecture also showed signs of cure as treated with *Asparagus*. When fishes pre treated with *Asparagus* was later treated with paracetamol there was lesser degree of damage to liver tissue and only slight variation in alkaline phosphatase activity was recorded. All these result clearly indicates that the plant *Asparagus* has promising ability to cure hepatic damage.

KEYWORDS : Hepatoprotective, Paracetamol, *Asparagus*, *Anabas*

INTRODUCTION

Liver toxicity or hepatotoxicity is the reason for many liver disorders and one of the most important worldwide health problems in recent years. Death due to liver damage is alarmingly increasing now a days. Liver damage is caused by ever increasing rate of atmospheric pollution and also as result of modern life style. Over use of medicines is related to many unhealthy practices of modern life style. Most of the hepatotoxic chemicals, including some drugs, damage liver cells mainly by inducing oxidative damage and lipid peroxidation in liver. Acetaminophen or paracetamol is one of such therapeutic chemical which otherwise has an excellent safety profile under prescribed dosage. Because of its common and wide usage it is used by large number of common people even without prescription by a medical practitioner. Due to scarcity of metabolically safe hepato-protective drugs, there is increasing interest in the alternative medicines for the treatment of liver diseases and associated metabolic ailments which include herbal remedies and dietary supplements. Chopra and Nayar (1986) have reported the medicinal value of many Indian medicinal plants.

MATERIAL AND METHODS

Healthy, uninjured specimens of *Anabas testudineus* were collected from Kuttanad region of Kerala State. They were brought to the laboratory and were acclimatized to lab conditions for one week. From this stock adult specimen weighing 16-21 gms and having snout - vent length 11-12.5 mm were selected for experimental purposes. Acclimatized specimens alone were used. They were fed with pelleted artificial fish feed. Special care was taken to provide proper aeration and keeping the tank clean from excreta and other waste materials.

Experimental protocol

Animals were divided into five groups of six fishes each and treated as below for 15 days.

Group-I: served as normal control received distilled water injection for 15 days.

Group-II: served as negative control received paracetamol injection 500mg/kg body weight for 15 days.

Group-III : served as experimental group received paracetamol injection as in group-II and then *Asparagus* extract 500 mg/kg body weight administered orally for 15 days.

Group IV: served as experimental group pre-treated with *Asparagus* extract orally for 15 days and then received paracetamol injection for 15 days.

Group V: Served as experimental control received *Asparagus* extract alone.

Animals were sacrificed under light ether anaesthesia 24 hour after the last dose. Blood was collected by cardiac puncture using a syringe and liver was removed, rinsed in cold saline and fixed in neutral buffered formalin for 48 hours. It was subsequently washed in distilled water and processed through graded series of alcohol, cleared in xylene and embedded in paraffin wax. Sections of 7 micron thickness were cut; stained with Harris haematoxylin and Eosin and mounted in DPX. Stained sections were examined with research microscope for histopathological changes.

RESULT AND DISCUSSION

The results obtained clearly revealed the hepatoprotective ability of *Asparagus*. After receiving paracetamol injection for ten days, some behavioural abnormalities like reduced activity level and heavy secretion of mucus were noticed in the fishes. Upon treatment with *Asparagus* for fifteen days fishes returned to their normal behavioural patterns. Fishes pre-treated with *Asparagus* extract before getting paracetamol injection also showed no behavioural abnormalities.

BIOCHEMICAL CHANGES:

Quantitative estimation of alkaline phosphatase activity in the blood of paracetamol treated fishes showed a sharp increase in levels of this enzyme when compared with the control fishes. In normal fishes the value was 75.5±0.6707. In fishes those received paracetamol at a dose of 500mg/kg body weight this value increased up to 140±3.1727. In fishes those received *Asparagus* extract for fifteen days increased alkaline phosphatase values showed a declining trend reaching up to 84.33±1.085. Fishes pre-treated with *Asparagus* extract before getting paracetamol injection also showed nearly normal values.

No of fish	Normal Control	Treated with paracetamol	Paracetamol + <i>Asparagus</i>	Pre treated with <i>Asparagus</i> + paracetamol	<i>Asparagus</i> extract alone
1	76	144	96	83	77
2	78	129	102	80	80
3	75	148	99	87	76
4	76	132	96	84	76
5	75	141	106	85	77
6	73	146	102	87	76
Mean & SE	75.5 ± 0.6707	140 ± 3.1727	100.5 ± 1.4548	84.33 ± 1.085	77 ± 0.6324

t value	t=19.889 35 P<0.1	t=14.2150 1 P<0.1	t=6.92352 P<0.1	t=1.6269 8 P<0.1
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HISTOPATHOLOGICAL CHANGES

The normal histological architecture of liver, various histopathological changes produced by paracetamol and the hepatoprotective ability of *Asparagus* as evidenced from histopathological studies in this organ of the fish are explained below.

HISTOLOGY OF NORMAL LIVER:

The liver of *Anabas testudineus* is a solid glandular organ, made up of polyhedral hepatic units called hepatic lobules. The surface is covered by a serous membrane and the hepatic lobules are made up of hepatic cells which are roundish polygonal in shape containing a clear spherical nucleus. They are located among sinusoids forming cord like structures known as hepatic cell cords. There is a central vein which drains blood away from the lobules. The wall of sinusoids contains phagocytic Kupffer cells. Sandwiched between the lobules, there are strands of connective tissue called portal canals carrying branches of portal vein, hepatic artery, bile duct and lymphatics.

GROSS PATHOLOGY OF THE LIVER:

When the liver samples of the treated fishes were examined grossly, they were found to be slightly enlarged but with severe congestion.

Histology of liver after *Asparagus* administration for 15 days.

Treatment of *Asparagus* alone caused no alterations in structure. Liver appeared as normal as in control.

Histopathology of liver under paracetamol administration:

Injection paracetamol 500gm / kg body weight of the fish for 15 days induced pronounced pathological changes in the liver tissue of *Anabas testudineus*. There were changes like swelling, loosening and clumping of hepatocytes and hemorrhage. Hyalinization and lymphocytic infiltration was also noticed.

Histopathology of liver under paracetamol administration and treatment with *Asparagus*.

After treating with *Asparagus* extract it was seen that hepatic lesions induced by paracetamol injection was markedly reduced.

Histopathology of liver under paracetamol administration with pre-treatment with *Asparagus*.

Pre-treatment with *Asparagus* caused little liver damage even after the administration of paracetamol for 15 days. Liver appeared almost normal except for some mild lymphocytic infiltration.

DISCUSSION

In the present study, activity of serum Alkaline phosphatase in the paracetamol treated group had a sharp and significant increase as compared to a healthy control group indicating successful induction of hepatotoxicity by repeated dosing of paracetamol. Yakout *et al.*, (2015) also have reported the medicinal properties of *Asparagus racemosus*. Histopathological studies also indicated significant alterations in liver architecture in paracetamol treated animals compared to the normal control group, further indicating liver damage. The healthy control group displayed normal liver histology while the paracetamol treated group showed swollen or occasionally apoptotic hepatocytes with coarse granular cytoplasm and compressed sinusoids. *Asparagus racemosus* treated groups exhibited strikingly normal liver histology without any anatomically detectable anomalies. In them the alkaline phosphatase activity also recorded normal range. When the paracetamol injected group was treated with *Asparagus racemosus* extract there was promisingly sharp decline in elevated ALP level. The hepatic tissue architecture also showed signs of cure as treated with *Asparagus racemosus*. Fasalu *et al* (2011) reported the hepatoprotective properties of *Asparagus racemosus* roots against paracetamol-induced hepatotoxicity in rats.

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