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# Role of Aqueous Bark Extract of *Terminalia Arjuna* on Serum Enzymes in Cigarette Smoke Exposed Rats

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**ABSTRACT:** Cigarette smoking is the leading cause of mortality and a major public concern. The aim of our study to investigate the role of aqueous bark extract of *Terminalia arjuna* on serum enzymes in cigarette smoke exposed rats. A total fifteen (15) adult healthy Wistar rats from 100-158 were divided into three groups (A, B and C) each containing five rats. Group (A) serving as control without exposure to cigarette smoke, Group (B) and (C) served as the experimental groups. The group (B) was exposed to Cigarette smoke (6 Cigarettes) for one hour/day for 30 days and experimental group (C) was exposed to cigarette smoke along with oral administration of aqueous bark extract of *Terminalia arjuna* (5mg/rat/day) for 30 days. The results indicate that, a significant increase in Alanine amino transferase (ALT) and Aspartate amino transferase (AST) after exposure to cigarette smoke in comparison to control group while, a significant decrease in ALT and AST level after cigarette smoke exposure along with oral administration of aqueous bark extract of *Terminalia arjuna* in comparison to Cigarette smoke exposed rats due to antioxidant defence mechanism.

KEYWORDS: Albino rat, Antioxidant, Cigarette smoke, Serum enzymes, Terminalia arjuna.

#### INTRODUCTION

Cigarette smoking is an important predisposing factor to many diseased conditions such as hepatic, respiratory and cardiovascular disease. It is one of the leading cause of preventable ill health in the world. Cigarette smoking is now increasing rapidly throughout the developing world and biggest threats to current and future World health<sup>[1]</sup>. It is a complex chemical composition with many toxic and mutagenic compounds. Cigarette smoke contains over 4000 compounds e.g. Nicotine, Tar, Carbon Monoxide, Ammonia, Butane, Cadmium, Arsenic, hydrogen cyanide which are harmful to the health<sup>[2]</sup>. It generates a range of oxidant, free radicals, reactive oxygen species (ROS). These free radicals are highly unstable and capable of undergoing complex interaction in biological system, make oxidative stress.<sup>[3]</sup> About 1014 free radicals enter in the lungs in every inhalation of cigarette smoke and it can directly or indirectly produce oxidative stress in the body<sup>[4]</sup>.

Cigarette smoke is able to cause tissue oxidative stress at various levels. Reactive oxygen species and free radicals damage DNA, proteins, carbohydrates, lipids and affect serum enzymes activity.<sup>[5]</sup> Serum enzymes- Alanine amino transferase (ALT) and aspartate amino transferase both are highly concentrated in the liver. ALT is localised solely in the cytoplasm and AST is present in the both cytosine and mitochondria of hepatocytes<sup>[6]</sup>. Cigarette smoke cause oxidative stress, which involves disorders in the balance between pro-oxidant and antioxidant agents<sup>[7]</sup>. The antioxidant play a key role in curbing the hazardous free radical formation and reduce the oxidative stress. *Terminalia arjuna* used as an antioxidant in the present study is an important medicinal plant and commonly known as *arjuna* belongs to combretaceae family. It is about 60-80 feet in hight. The bark of *Terminalia arjuna* contains numerous bioactive compounds such as glycosides, flavonoids, tannins and minerals which have antioxidant, anti-inflammatory ,hypocholesterolaemic, hepatoprotective and cardioprotective properties due to presence of these compounds<sup>[8]</sup>. Thus the present study, designed to evaluate the role of aqueous bark extract of *Terminalia arjuna* on serum enzymes in cigarette smoke exposed rats.

### MATERIALS AND METHODS

The adult and healthy wistar albino rats of both the sexes (100-158g) have been selected for the present study. They were kept in polypropylene cages and maintained at standard laboratory conditions of temperature  $21\pm0.5$  and relative humidity  $60\pm5$  % with a photoperiod 12 hours/day. The experimental protocol was in accordance with institutional ethics committee. Threats were fed on commercial food pellets (Golden feed, New Delhi) and water *ad libitum*. The experimental animals were acclimatized for one week prior to the experiment.

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The capstan pilot (a filtered cigarette of 64 mm length) ITC limited, kolkata was selected for the present study.

#### Plant Material and extraction:

The *Terminalia arjuna* bark was collected from the botanical garden university campus, Dr. Bhim Rao Ambedkar University Agra and Identified by a plant taxonomist. Heat distillation process was used. The stem bark powder was boiled in distilled water at 1:5 ratio at 100°C for 30 minutes<sup>[9]</sup>. After 30 minutes the mixture was filtered and the filtrate was stored in a refrigerator until use.

#### **Experimental design:**

The albino rats were divided into three groups one control (A) and two experimental groups (B and C) each group containing five rats.

Control Group (A): Unexposed to cigarette smoke

Experimental Group (B) : Exposed to cigarette smoke 6 cigarettes/hr in a day

(1 cigarette /10 minutes) for 30 days.

Experimental group (C): Exposed to cigarette smoke along with oral administration of

aqueous bark, extract of Terminalia arjuna (5mg/rat/day) for

30 days.

#### **Exposure to Cigarette Smoke:**

Mini exposure cabinet (60 cm x 30 cm x 30 cm) manufactured by Precision Instrument, Varanasi is used for cigarette smoke exposure. The experimental rats were kept in an isolated smoke chamber with their cages for whole body exposure to cigarette. Smoke of a filtered cigarette (6 cigarettes/ hour in a day) for 30 days.

#### **Blood Collection:**

At the end of exposure duration of 30 days, all threats of control group (A), and experimental group (B and C) were sacrificial under light anaesthesia (diethyl ether). The blood samples were collected by cardiac puncture of the dissected rats with the help of 5.0ml sterilized disposable syringe and transferred into plain sterilized centrifuge tubes for separation of serum.

#### Separation of Serum:

For the separation of serum, blood samples were allowed to stand for one hour undisturbed and centrifuges at 2500 rpm for 30 minutes. The supernatant serum was separated for the estimation of serum enzymes.

#### **Estimation of Serum Enzymes:**

Serum enzymes ALT and AST were estimated by UV (IFCC) kinetic assay kit method (Span diagnostic Ltd., Sachin) described by Schumann *et al.*, 2002<sup>[10]</sup>.

#### Statistical calculations:

The data were expressed as mean Em. They were signified by using 't' test by KpKy plot ( Version 3.0).

#### **RESULTS AND DISCUSSION**

The results of the present study for serum enzymes ALT and AST in control and experimental rats are given in (Table-1). After 30 days exposure to cigarette smoke, there was a significant elevation is serum ALT and AST level in comparison to control group while, a significant decrease in, serum ALT and AST level after exposure to cigarette smoke along with oral administration of aqueous bark extract of *Terminalia arjuna* in comparison to cigarette smoke exposed rats.

Cigarette smoke generates free radicals which reduces endogenous antioxidant, produce oxidative stress that inducing lipid peroxidation cause alternation in membrane permeability results elevation in ALT and AST level. Cigarette smoke increases the level of some enzymes such as ALT and AST which are capable of inducing alterations in membrane permeability properties of the liver<sup>[11]</sup>. ALT and AST are present in hepatocytes liver parenchyma cells and elevation in ALT and AST level are due to injury of these cells results in the elevation of these enzyme caused by cigarette smoke<sup>[12]</sup>. Cigarette smoke increase the activities of SGOT and SGPT and these enzymes leak out from the liver cytosol into the blood stream which gives an indication of hepatotoxic effect<sup>[13]</sup>.



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Cigarette smoke increases the levels of AST and ALT<sup>[14]</sup>. Smoking significantly increased the level of aspartate amino transferase(AST)<sup>[15]</sup>. Arsenic is a component of cigarette smoke increase AST and ALT levels indicating liver dysfunctions and cardiac disorders in rats<sup>[16]</sup>. Cadmium which is present in cigarette smoke caused a significant increased in activity of SGOT which is an indicator of cardiac injury in rats<sup>[17]</sup>.

<u>Terminalia arjuna</u> is a deciduous ever green tree found all over in India. Its stem bark possesses glycosides, large quantities of flavonoids, tennis, triterpenoids and minerals. The aqueous bark extract of *Terminalia arjuna* protect against oxidative stress and reduce the level of lipid per oxidation. It has various components e.g. polyphenols, flavonoids, glycosides, steroids triterpenoids, proanthocyanidins, gallic acid, ellagic acid and minerals. These compounds exert antioxidant, anti-inflammatory activities due to free radicals scavenging properties<sup>[18]</sup>. Aqueous bark extract of *Terminalia arjuna* acts as effective antioxidant augments endogenous antioxidant compounds that reduced the oxidative stress and lipid per oxidation<sup>[19]</sup>. The bark of *Terminalia arjuna* has flavonoids *and* tannins these compounds facilitate the regeneration of hepatic cells and stabilization of the plasma membrane and reduced the level of SGOT and SGPT in rats<sup>[20]</sup>.

#### CONCLUSION

From the findings of our present study revealed that aqueous bark extract of *Terminalia arjuna* has tremendous potential to provide protection against cigarette smoke induced alterations in serum enzymes ALT and AST level. The aqueous bark extract of *Terminalia arjuna* has antioxidant activity due to presence of numerous phytochemicals components which scavenge free radicals and reduced the level of ALT and AST.

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#### **Conflict of Interest:**

There are no conflicts of interest.

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**Table-1:** Effect of Aqueous bark extract of *Terminalia Arjuna* on ALT and AST level after exposure to cigarette smoke in Albino rats.

Groups	Treatment	30 days (Mean ± S.Em)	
		ALT(IU/L)	AST(IU/L)
Control Group A (5)	Ambient air	40.72 <u>+</u> 1.532	163.83 <u>+</u> 3.299
Experimental Group B (5)	Cigarette Smoke	48.142 <u>+</u> 3.412 <b>↑</b> *	169.16 <u>+</u> 3.286 ▲**
Experimental Group C (5)	Cigarette smoke + Aqueous Bark extract of T. <i>arjuna</i>	40.950 <u>+</u> 2.233 <b>↑</b> NS <b>↓</b> **	164.884 <u>+</u> 2.966 <b>↑</b> NS <b>↓</b> **

(5) = No. of Albino rats = NS- Non significant

\* - Significant \*\* -Highly significant f increase decrease.

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