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TOXICOLOGICAL STUDIES *OF MELIA AZEDARACH* L. (FLOWERS AND BERRIES)

ZAKIR-UR-RAHMAN, SHAHNAZ AHMAD, SHAMIM QURESHI, ATIQ-UR-RANMAN and YASMEEN BADAR
Pharmaceutical and Fine Chemicals Research Centre, P.C.S.I.R. Laboratories Complex, Karachi-75280, Pakistan

ABSTRACT

Toxicological evaluation of *Melia azedarach* Linn. was carried out in lower laboratory animals, i.e. rats and mice by oral and intravenous routes. Aqueous and alcoholic extracts were found to be non-toxic instil a dose of 1500 mg/kg orally in mice and rats. When injected aqueous extract intravenously, LD_{50} was 395, 500mg/kg (flowers) and 700, 925mg/kg (berries) respectively in mice and rats. It was also observed that alcoholic and aqueous extracts *of Melia azedarach* have a mild CNS sedative effect.

Introduction

Melia azedarach Linn. (Meliaceae, synonym *Melia orientalis* Roam.), commonly known as "Persian Lilac", "Bakain" or "think" grows wild through out the sub-himalayan tract. It is cultivated in India and Pakistan for both ornamental and medicinal purposes (Dymock, 1890; Watt, 1893; Kutikar and Basu, 1933; Nadkarni, 1954; Chopra, 1958; Watt and Brandwijk, 1962; Abdulla, 1972).

Literature *survey* reveals that in many parts of the world preparations of *Melia azedarach* a*re* being used locally and systematically for curing many diseases (Watt and Brandwijk, 1962; Baquar, 1989). The plant is considered as resolvent, deobstruent and alexipharmic. Locally flowers, leaves, fruits/berries and bark are used for curing many diseased skin conditions, such as eczema, ulcerative wounds, syphilitic ulcers, leprosy, scrofula etc. in the form of lotion, ointment or poultice. Systematically it is used as an emetic, cathartic, anthelmintic, antipyretic, expectorant and diuretic (Dymock, 1890; Watt, 1893; Chopra, 1958; Baquar, 1989). Commercially the oil of *Melia azedarach* is used in soap and cosmetic industries.

Chemical composition reveals the presence of alkaloids, gum, resins, tannins, meliotannic acid, benzoic acid, vanillic acid, cinnamic acid, β-sitosterol, phenol, coumarin, teranor-triterpenoid, triterpenes, glycoside, bakayanin and margosine (Khalid *et al*, 1979; Schultz *et al*., 1979; Khalid *et al*, 1981; Oelrichs *et al.*, 1983; Srivastava and Savitri, 1987; Baquar, 1989). Oil of *Melia azedarach* was found to be deficient in Sulphur (Rao *et al*, 1984).

Ample data is available on the toxicity of *M. azedarach* to bovine, sheep, goat and the poultry (Hoti *et al.,* 1976), and the toxic constituent responsible for toxicity is a melatonin (Oelrichs *et a1.*, 1983, 1984). No report or data is available on its toxicity on lower laboratory animals, *i.e.* rats and mice etc. Therefore, it was thought necessary to determine its toxicity to lower laboratory animals, *i.e.* rats and mice. This paper covers the results achieved by using extracts (alcholic and aqueous) of *Melia azedarach* flowers and berries.

Materials and Methods

Fresh flowers and berries of *Melia azedarach* were taken. Stalks were removed from the flowers, and berries were crushed slightly. About 1 kg each of flowers and berries were soaked in 4 liters of 95% ethyl alcohol for 96 hours. The semi-solid solvent was then decanted and concentrated *in vacuo*. Resultant mass obtained was referred to as "alcoholic extract". Half of the alcoholic extract was partitioned with petroleum ether and water (1:2, v/v). Aqueous layer was then separated and concentrated under reduced pressure at room temperature into a semi-solid mass and was referred to as "aqueous extract".

Toxicity studies

Healthy albino rats of Sprague Dawley strain and mice (male and female) reared at PCSIR Animal House, weighing 100.120 g and 25 - 30 g respectively, were selected for oral as well as parenteral route investigations. The animals were kept under optimal experimental conditions. Animals used for testing were housed in standard plastic cages having the dimension of 12.0 x 8S inches at top, 10S x 8.0 inches at bottom and 63 inches high. Normal routine feed was given to animals and water was supplied freely by means of inverted bottles. To facilitate the movement of animals saw dust was spread in cages. Cages were marked with their respective doses. Each dose was repeated thrice to confirm the results.

Oral route

Doses in the range of 100-1500 mg/kg body weight of alcoholic and aqueous extracts of *Melia azedarach* flowers and berries was administered by means of a gavages in a single dose to a group of rats and mice of either sex A group of 10 animals was used for each dose level. All animals were observed carefully for a period of five days for gross physical and behavioural changes and mortality rate was noted daily. A few rats and mice were killed after 24 hours and microscopy done. Control group was run simultaneously and were given the same volume of the vehicle, i.e. 95% ethyl alcohol: water (1:2, *v/v)* by means of a gavage.

Parenteral route (intravenous route)

The intravenous toxicity was done by injecting the aqueous extract (flowers and berries) through tail vein of either sex in a group of 8 for each dose level in both *rats* and mice. Inject able volume and the time taken for injecting the drug was kept constant in order to avoid volume and time variation effect. The animals were closely and continuously observed for gross physical and behavioural changes. The animals, which survived, were kept under observation for a period of 5 days. Mortality rate was noted and autopsy was carried out in order to observe any drug related gross change.

Results and Discussion

Assessment of the toxicological manifestation of the two different extracts, alcoholic and aqueous, of *Melia azedarach* flowers and berries through oral and intravenous route respectively in rats and mice reveals that their is marked variation in the severity and depth of symptoms by both routes. Furthermore, signs and symptoms observed by both routes are dose dependent.

Oral route toxicity

A single dose regimen of *Melia azedarach* flowers and berries (aqueous and alcoholic) both in rats and mice upto a dose range of 100-1500 mg/kg did not show any mortality. However, a dose of 510 mg/kg (alcoholic) and 650 mg/kg (aqueous) extracts showed mild CNS sedation. Signs and symptoms became more marked and intensed with increasing dose. Gross physical and behavioural changes include vasodilatation followed by vasoconstriction, deep shallow respiration, piloerection, catching and gagging, increased heart rate (tachycardia), loss of writhing renews, sedative mood and dried mouth. Necroscopic finding revealed no drug related gross changes.

Intravenous toxicity

No mortality was observed in mice and rats uptil a dose of 380, 520 mg/kg body weight (flowers) and 550, 800 mg/kg body weight (berries) respectively. The LD_{50} was found to be at a dose of 395, 580 mg/kg body weight (flowers), and 700, 925 mg/kg body weight (berries) in mice and rats, whereas LD_{100} was 405, 602 mg/kg body weight (flowers) and 825, 965 mg/kg body weight (berries) respectively in mice and rats as shown in Table-1

Table 1

Intravenous toxicity of *Melia azedarach*

Test animal	Flowers			Berries		
	E.D. mg/kg	LD_{50} mg/kg	LD_{100} mg/kg	E.D. mg/kg	LD_{50} mg/kg	LD_{100} mg/kg
Mice	380	395	405	550	700	825
Rats	520	580	602	800	925	965

In this case also the depth and severity of signs and symptoms are dose dependent. Major gross physical and behavioural changes observed were vasodilatation followed by vasoconstriction, depression of respiration, papillary constriction, piloerection, marked chill, muscle stretching, hyperirritability, gagging, tachycardia, dried mouth, micturation decreased physical activities and bradykinesis. The animal lied flat on its abdomen and initiation of movement became difficult, ponderous and extremely inefficient. In some cases this leads to paralysis of hind limb.

Amelioration of all these signs and symptoms takes few minutes (5-40) in lower doses, while in higher doses recovery time was enhanced up to 90-110 minutes. The drug was excreted in urine 20-40 minutes after intravenous injection. In doses where mortality was observed it was preceded by tremor, cessation of respiration followed by asystole. In cases where animals survived it was observed that the aqueous extract of *Melia azedarach* (flowers and berries) has elevated the pain threshold. Some animals exhibited a sign of urination before death. Autopsy finding did not exhibit any particular drastic change in the body except few hemorrhagic spots on heart, liver and stomach. Urinary bladder was swollen.

Conclusion

The flowers and berries of *Melia azedarach* are toxic also to lower laboratory animals, i.e. rats and mice. Toxicity depends not only on dose but also on the route of administration. Higher concentration of the extracts depresses markedly the respiratory centre by both routes, oral as well as parenteral. This may be due to the direct action on the respiratory centers. In doses where mortality was observed it was noted that death occurs due to the cessation of respiration. If at this stage oxygen was made available, the

animal recovers. In doses where animal survived beside respiratory depression, analgesic activity was observed without the loss of consciousness.

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