Myodegeneration in Rats Fed Mindi *(Melia azedarach)*

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Abstract

The tree, *Melia azedarach L*, known locally as mindi and as white cedar in Australia, has been linked to poisoning in man and animals. In West Java in 1985 the leave of *Melia* were proposed as a traditional medicine for a variety of human diseases. Leaves are fed to ruminants in Bali during the dry season. To study the toxicity of *Melia*, young rats were divided into 6 groups of 5 rats. Rats in group 1 to 4 were fed daily a diet containing 25 % *Melia* leaves (dry) for 7, 14, 21 and 28 days, while groups 5 and 6 received a control diet. Sixty % (3/5) rats in group 3 died before 3 weeks of treatment, and 4/5 rats in group 4 died before 28 days treatment. Signs including anorexis, weakness and mild incoordination were observed in most rats in groups 1 to 4 but not in the controls. Paralysis of rear limbs and recumbancy were also seen in groups 3 and 4. On histological examinations, skeletal muscle changes were seen in all rats in groups 3 and 4. These lesions were characterized by necrosis and fragmentation of fibres, hyaline degeneration, proliferation and invasion by sarcolemmal nuclei and macrophages. These muscle changes have not been reported to be caused by the tetranortriterpene meliatoxins previously isolated from fruit of Australian *Melia azedarach* and shown to be responsible for an acute nervous syndrome and death in pigs.

Introduction

The tree *Melia azedarach* L, (Meliacea) is known locally in Indonesia as mindi, gringging or intaran, and as white cedar, cape lilac or Chinaberry in Australia and else where. Although now found in many parts of the world, *Melia* is thought to be a native of Persia, China and India. In Indonesia *Melia* is commonly planted as a fast-growing shade tree in villages and along roadsides.

Melia azedarach has been used medicinally in many countries for the treatment of a variety of human disorders (Watt and Breyer-Brandwich, 1962), but there are many early records of the plant beng poisonous to man. All parts of the plant have been shown to be toxic, but most cases occur from eating fruits. The ripe fruit was more toxic than the immature one or other parts of the plant (Vahrmeijer, 1981). Poisoning of pigs, cattle, sheep, goats and poultry has been reported (Everist, 1981; Watt and Breyer-Brandwich, 1962).

Observations from the Animal Research Institute in Queensland have suggested that toxicity may vary with location or stage of growth, and may be entirely absent in some trees (Oelrichs, et al, 1983, 1985).

In Indonesia there is no information about the toxicity of *Melia* for man and livestock. Nevertheless in Bali *Melia* is commonly consumed by cattle (Ressang, 1984), and in West Java in 1985 the leaves of Melia were proposed as a traditional medicine for a variety of human diseases. Earlier experiments (Sani, 1985, unpublished), in which *Melia* at 50% in rat diets induced multiple gastrointestinal haemorrhage and muscle pathology, stimulated the present study.

Materials and methods

Plant material was collected from roadsides in Bogor area, West Java. The leaves were separated, dried at 50° C in an over for 48 hours and milled to a fine powder. The powdered leaf was added at the rate of 250 g per kg standard diet (25% Melia in a diet). After mixing the constitutents, water was added to obtain a homogenous mix which was extruded into cylindrical pellets by a meat grinder. The pellets were dried at 50° C in oven for 48 hours and stored at room temperature.

Thirty 3 month-old rats housed individually were divided into 6 groups of 5 rats. The rats were alotted to cages at random and were weighed at the start of feeding trial and every week thereafter. Four groups (group 1 to 4) of rats were provided a diet containing 25 % melia leaves for 7, 14, 21, and 28 days respectively, while groups 5 and 6 (the control groups) received a control diet for 14 and 28 days respectively. Feed and water were available *ad libitum* throughout the treatment period. During the feeding period clinical symptoms were observed and dead animals necropsied. At the end of the feeding period all remaining rats were killed and examined.

Animals were killed under ether anaesthesia and examined for gross abnormalities. Samples of liver, kidney, stomach, small intestine, and skeletal muscle were fixed in 10 % neutral formol saline. The tissues were trimmed and embedded (blocked) in paraffin wax, and sections of 6 um were prepared and stained with haematoxilin and eosin using standard procedures. The slides were examined by light microscopy.

Results and discussion

No rats in group 1 and 2 had died after 1 and 2 weeks of feeding trials, but the animals showed weakness, anorexia and weight loss. These rats were killed and post mortem examination carried out. No gross pathology was obvious at necropsy.

All rats in group 3 and 4 showed clinical symptoms such as anorexia, weakness, weight loss, and mild incoordination. Signs including paralysis of rear limbs and recumbancy were also seen in groups 3 and 4. Three of 5 rats in group 3 died at 16, 17, and 19 days of treatment, and 4 of 5 rats in group 4 died at 17, 20, 21 and 26 days of treatment.

Grossly pale skeletal muscles in rear limbs were observed in most of rats in group 3 and 4. A few rats showed ulcerative lesions in the gut. No changes were seen in other organs.

On histological examination, skeletal muscle changes were seen in all rats in groups 3 and 4. These lesions were characterized by necrosis and fragmentation of fibres, hyaline degeneration, proliferation and invasion by sarcolemmal nuclei and macrophages. The characteristic gut pathology reported by Oelrichs, et al (1985) and fatty degeneration and hyperaemia of the liver and kidney (Kingsbury, 1964) were not observed in our rats.

Muscle changes such as we observed have not been reported to be caused by the tetranortriterpene meliatoxins when fed to pigs (Oelrichs, et al, 1983, 1985). Few phytotoxins or other chemicals are known to cause muscle fibre degeneration under experimental conditions. *Cassia* spp have been

reported to cause myopathy, usually accompanied by liver lesions, in horses, cattle and small ruminants in Southern United States (Bailey, 1985; Rowe et al, 1987). Skeletal muscle degeneration is also reported as one of the lesions caused by the mycotoxin, cyclopiazonic acid, in chicks (Cullen et al, 1988). Selenium/vitamin E deficiencies and excess exertion can also cause myopathies (Jubb et al., 1985).

Another member of the family Meliacea, *Azadirachta indica* or neem tree, has been reported both to be toxic (Ali, 1987) and to share tetranortriterpenoid constituents with *Melia azedarach* (Kraus et al, 1987). Ali (1987) described haemorrhages, congestion and degeneration in a variety of organs but did not list skeletal muscle among the tissues taken for histopathology.

Further toxicological study of *Melia azedarach* is waranted to determine the most sensitive organ in livestock consuming *Melia* and to address other issues, such as the possibility of non-toxic tress discussed by Oelrichs et al (1985).

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