SUMMARY & CONCLUSION
CHAPTER V

SUMMARY AND CONCLUSIONS

Exposure to high levels of arsenic has been recognized for many decades in some regions of the world. Millions of people are at risk of cancer and other diseases because of chronic arsenic exposure. Studies on this metal are carried out for hazard identification and safety assessment to protect the health of exposed life. The therapeutic effects of AP and MLT were also explored in view of the widespread arsenic induced health hazards the world over.

Adult Swiss female mice were administered arsenic trioxide at a dose of 0.5 mg/kg body weight (low dose) and 1.0 mg/kg body weight (high dose) for 30 days to investigate the gravimetric, histological, biochemical changes and arsenic concentration in certain endocrine organs (ovary, adrenal, pancreas and thyroid gland). The above doses were established on the basis of LD$_{50}$ values of arsenic trioxide. Role of Andrographis paniculata extract (andrographolide) and melatonin (MLT) were also investigated in this study. The control and treated animals were maintained on standard chow and water ad libitum. At the end of all the treatments, the animals were sacrificed on 31$^{st}$ day along with control and utilized to study various parameters. The study can be summarized as follows.

Arsenic trioxide treatment brought about a significant decline in the body and organs weights in a dose dependent manner which could be attributed to low intake of food due to the treatments. A decline in the levels of total sulphydryl in pancreas and reduced levels of total proteins in endocrine
organs might be due to binding of arsenic to proteins (–SH group) making it less available for other metabolic processes and protein synthesis impairment. Hence enzyme activities, structural proteins and secretions of the concerned organs would be affected. Cholesterol and activities of 3β and 17β dehydrogenases are directly involved in the biosynthesis of steroid hormones. A hypercholesterolemic condition was induced in ovary and adrenal with simultaneous decrease in the activities of 3β and 17β hydroxy steroid dehydrogenases by arsenic treatment indicating altered steroidogenesis in ovary and adrenal gland. The total ascorbic acid levels decreased in ovary and adrenal suggesting stress imposed by arsenic. Glutathione levels declined indicating reduced detoxification and altered ascorbic acid metabolism in endocrine organs. Arsenic treatment also caused formation of free radicals in above mentioned organs. The lipid peroxidation levels recorded a marked increase in arsenic treated groups. Exposure further led to a significant depletion in the activities of superoxide dismutase, catalase and glutathione peroxidase in treated mice. The observed alterations might be attributed to the utilization of these antioxidants to alleviate free radical induced oxidative stress.

Alteration of normal blood glucose levels revealed that exposure of arsenic possibly induces diabetes mellitus. Arsenite has high affinity for sulphydryl groups and thus can form covalent bonds with the disulfide bridge in the molecules of insulin, insulin receptors, glucose transporters and enzymes involved in glucose metabolism. As a result, the normal functions of these molecules can be hampered. Serum amylase and lipase are group of enzymes found primarily in the pancreas. Arsenic administration also elevated
the serum level of amylase and lipase due to impairment of acinar tissues indicated pancreatic dysfunction. Treatment of arsenic further caused significant reduction in the level of cholesterol and protein in the serum of mice as compared to controls. Existence of arsenic levels in all the endocrine organs reflects its accumulation. Histological alterations were also found after arsenic treatment in the endocrine organs which could be correlated with the alterations in biochemical indices.

To study the beneficial effects of *Andrographis paniculata* (AP) and melatonin (MLT), the antidotes were administered separately to the animals. *Andrographis paniculata* and melatonin were given at the dose of 50 mg and 10 mg/kg/animal/day respectively. AP supplementation reversed the changes caused by arsenic in biochemical and oxidative stress related parameters like protein, cholesterol, total –SH groups, blood glucose levels, ascorbic acid, lipid peroxidation and glutathione. Further the activities of serum amylase, lipase, 3β and 17β hydroxyl steroid dehydrogenases, superoxide dismutase, catalase, and glutathione peroxidase were unaffected. Arsenic levels in all organs were comparable to control values. No or minimum changes were observed after the treatment with AP indicating its role in arsenic toxicity.

Melatonin, a secretory product of the pineal gland, is now known to be produced in multiple cells and organs. Besides its ability to directly neutralize a number of free radicals and reactive oxygen and nitrogen species, it stimulates several antioxidative enzymes which increase its efficiency as an antioxidant. The results showed that MLT supplementation modified the change caused by arsenic in endocrine organs. The levels of protein, cholesterol, 3β and 17β dehydrogenases, total sulphhydryl and blood glucose
were remain almost unaffected. The activities of antioxidant enzymes, glutathione and lipid peroxidation were also unaltered after the combined treatment. No histopathological changes were observed after the treatment with MLT combination. Arsenic levels in the mentioned organs were also equivalent to control values.

In conclusion, the present data thus revealed that arsenic is toxic by inhibiting defense mechanisms and functions of endocrine organs of adult Swiss female mice. The results summarized here document the ability of AP and MLT to reduce oxidative damage induced by arsenic. Amelioration seems to be similar for these compounds.
FUTURE LINES OF WORK

1. Effects of arsenic on hormonal parameters of endocrine glands.

2. Ultrastructural investigations would be helpful to understand the
damage caused to cell organelles.

3. Detection of arsenic levels in other biological tissues and groundwater.

4. Similar studies as in this thesis need to be carried out on males and
other animal models.

5. Investigations are called for their effects on signal transduction and
apoptosis.

6. Research on antifertility effects needs to be done.

7. Teratogenic effects of arsenic also need to be studied.

8. Role of combination of AP and MLT need to be done.

9. Reversibility studies are also necessary.

10. Interaction of arsenic with other metals or combination metal studies is
an urgent need to be investigated.
ABSTRACT PUBLISHED


RESEARCH PUBLICATION:
