Summary & Conclusions
Selenium is a trace element that is essential in small amounts, but can be toxic in larger amounts. Soil is the pre-eminent source of most biologically active trace elements, which reach man through plants and animals. Since selenium continually enters into the human body from soil, water and air it gets accumulated in the body. This condition may lead to toxicity. Selenium is known to exist in the atmosphere from sea spray, windblown mineral dust, biogenic materials, volcanic effluvia, or coal combustion emissions. The use of selenium in photocopiers may create elevated airborne levels in the workplace. Anthropogenic activities such as mining, smelting, the burning of coal and oil and application of fertilizers may contribute to local selenium deposition on soil.

There is a strong correlation between low levels of selenium and high occurrence of cancer and atherogenesis. High amounts of selenium-enriched food stuffs have been prescribed as dietary essentials for prevention of cancer. It has been widely used as an antioxidant by many medical practitioners and it might lead to selenium toxicity. Selenium also assists the immune system, by increasing the antibody response to infection. It has been recognized as an essential element in biology and medicine. Although selenium is required for health, high doses can be toxic. Acute and fatal toxicities have occurred with accidental or suicidal ingestion of gram quantities of selenium. Chronic selenium toxicity (selenosis) may occur with smaller doses of selenium over long periods of time. Chronic selenium toxicity (selenosis) leads to reduced haemoglobin levels in humans. Selenium is teratogenic in birds and possibly also in domesticated animals (pigs, sheep and cattle). High doses of selenium
diet alters or decrease the spermatozoal quality characteristics of sperm count and motility in human beings. Deficiency of Se leads to Keshan disease, congestive cardiomyopathy, anaemia, enhanced cancer risk, immune system alterations, hair and nail changes, AIDS and Se abnormalities in thyroid hormone metabolism.

It is not easy to distinguish between Se toxicity and Se deficiency since they can induce similar effects, including anaemia, hair loss, infertility and impaired growth. The present study has attempted to establish in mice a sublethal concentration of Se that is essential for adequate nutrition and does not interfere with clinical parameters in chronic studies. Xenobiotic accumulation and their possible implications in biotic community is gaining importance at present. The teratological studies for three generations of rats were carried out by Srivastava and Raizada (2000) with hexachlorocyclohexane. The study for the three generation mice is to emphasize the core effects of selenium in the offsprings.

The toxic potentiality of selenium was determined through probit graphical method by Finney (1964), graphical method by Litchfield and Wilcoxon’s (1949), probit arithmetical method by Finney (1971), Karber’s method (1931) and cumulative mortality method by carpenter (1975). The results obtained for LD50 dose by the respective methods are 8.128, 8.128, 8.097, 8.60, 8.158. The mean apparent LD50 dose is 8.2065 (mg/kg body weight) for 48 hrs (Tables 1.1 to 1.6; Figs. 1.1 to 1.3). Since selenium is not traced in the contaminated natural ecosystems, 1/10th LD50 was selected as sublethal concentration for the present investigation. In the present
investigation, the sub-acute toxic effect of sodium selenite has been carried out studying energy metabolism, haematological, teratological, histological, spermatozoal and detoxification aspects in albino mice.

The healthy albino mice were divided into two batches having 5 (3 female + 2 male) mice each. The first batch served as control and they received isovolumetric quantities of distilled water. The second batch termed as the experimental mice were fed with 1/10th of sublethal dose of selenium in water.

The offsprings of the second batch experimental mice were called as first generation mice. The second generation was obtained from the first generation mice and the third generation from the second. All the three generations were fed selenium in water until they were sacrificed.

Teratological studies were carried from these offsprings. Adult mice from the three generations were subjected to haematological, biochemical, histological and sperm changes studies. The control mice were also subjected to the above mentioned studies.

In order to understand the energy related alterations in glycolysis and Krebs cycle, the activity levels of the selected dehydrogenases were estimated (Tables 2.1-2.3; Figs. 2.1-2.3). The NAD+ dependent lactate dehydrogenase (NAD+ - LDH) activity showed increase in the tissues. Decrease in dehydrogenases of Krebs cycle like isocitrate dehydrogenase (NAD+ - ICDH) and succinate dehydrogenase indicated the prevalence of hypoxic condition.
This reduced oxidative metabolism of mitochondria is due to selenite stress leading to binding of selenium to -SH group containing enzymes and mitochondrial swelling. The increased levels of LDH and decreased levels of ICDH and SDH confirms a shift in normal balance of glycolysis in favour of anaerobiosis.

Though the tissue mitochondrial oxidative enzymes were inhibited, yet glycolysis and other alternative enzyme systems, mostly confined to the cytosolic fractions were elevated. Since breakdown of glycogen and allied precursors require ATP, the enzyme systems namely total ATPases and phosphatase systems have been assayed in the tissues of albino mice exposed to sodium selenite.

In the present study the total ATPase activity is inhibited (Table 2.4; Fig. 2.4). This might be due to the action of selenium which affects the mitochondrial integrity as well as the enzyme systems localized in the mitochondria, leading to less synthesis of ATP molecules. The tissue acid and alkaline phosphatase activities were elevated in the present study (Tables 2.5 & 2.6; Figs. 2.5 & 2.6). Comparatively, alkaline phosphatase appears to be more active than the acid phosphatase. The alkaline phosphatase activity has a role in mineral metabolism, bone metabolism and synthesis of mucopolysaccharides, acid phosphatase with lysosomal activity. A decrease in the total ATPase system is well compensated by an increase in the tissue phosphatase system.
Detoxification is a process of continuous reactions on a particular chemical. Exposure of cells to oxygen radicals may cause lipid peroxidation in cell membranes which in turn may generate species that damage cell proteins and promote their degradation. Elevated XOD activity levels in the present study in the tissues of selenite treated mice (Table 3.1; Fig.3.1) indicate increased oxyradical production (superoxide radicals) which is responsible for oxidation of poly unsaturated fatty acids (PUFAs) in successive generations. It also indicates the effective detoxification of ammonia channeling the same towards uric acid synthesis, thereby maintaining nitrogen balance in the tissues under selenite stress. Increased catalase activity in the present investigation (Table 3.2; Fig. 3.2) suggests its active involvement in decomposition of superoxide radicals, generated by XOD. Catalase activity showed an increase indicating its active involvement in the decomposition of superoxide radicals and \( \text{H}_2\text{O}_2 \) thereby decreasing the toxicity of selenium. Glutathione plays an important role in the detoxification of xenobiotics. In the present study, tissues showed an elevated levels of glutathione S-transferase (GST) activity (Table 3.3; Fig. 3.3) under selenite toxicity, indicating active detoxification of selenium through enzymatic processes.

Histopathological changes were observed in liver tissue under sodium selenite treatment. Since, liver is the major metabolic centre to detoxify the chemical, the experimental mice under sodium selenite have shown moderate degenerative changes in cytoplasm, necrotic changes, cellular disarray and severe necrotic changes in hepatocytes. Thus, selenium feeding for three generations of mice resulted in dramatic cellular damage (Plates 3.1 to 3.3; Figs. A to F). These changes might be the possible reasons for significant changes in detoxifying enzymes investigated in the present study.
In the present investigation, haematological parameters (Table 4.1; Fig. 4.1) were altered in sodium selenite intoxicated albino mice (Table 4.1; Fig. 4.1). Sodium selenite produced a significant decrease in RBC, WBC count and Hb, PCV per cent and also decrease in MCV, MCH and MCHC. Selenium could produce anaemia indirectly by interfering with iron metabolism. Sodium selenite administration led to microcytic hypochromic anaemia.

Decrease in red blood cells count may suggest selenite fed development of haemolytic anaemia in mice. Significant decrease in WBC count due to selenium administration leads to lymphocytopenia.

In differential count also a gradual decrease was noticed in lymphocytes and monocytes. Increase was found in neutrophils, eosinophils, due to Se intoxication. The changes in haemogram indicates that cellular disorders are possible in blood corpuscles.

In the present investigation few teratological changes were observed in sodium selenite intoxicated albino mice (Table 5.1; Fig. 5.1) (Plates 5.1-5.4; Fig. A-H). The changes include retardation in litter number, size and litter weight. The reduction in skeletal and visceral weights indicates that the sodium selenite used in the present investigation might have inhibited the organogenesis and foetal development. Morphologically and teratologically there were no major malformations found in offsprings of first and second generation mice. But in third generation dead litters had black spots and wrinkled skin on the stomach region. In few cases of litters syndactyly was observed (Plate 5.3; Figs. E & F). Light malformations in skeletal and visceral
changes suggest the possibility of sodium selenite being foeto toxic. These results led to the conclusion that sodium selenite was lightly teratogenic to mice offsprings up to dose level of 0.82065 mg/kg body weight.

In the present investigation sodium selenite resulted in decrease of spermatozoal quality characteristics (Table 6.1; Fig. 6.1) of sperm count and motility in experimental mice. It seems that selenium affects the oxidoreductase activity of glutathione and resulting in oxidative damage to testis. Oral administration of sodium selenite produced a gradual decrease in sperm count and motility. Cumulative effect of Se could be seen in the sperm morphology, production and testicular damage.

The experimental mice testis have shown reduced seminiferous tubules, increased lumen, necrotic condition in seminiferous tubules and much degenerative changes in spermatids of seminiferous tubules (Plates 6.1 to 6.3; Figs. A-F).

The discrete pathological changes observed in the present investigation, might be the possible reasons for decrease in sperm count in all generations of mice under sodium selenite. However, the low levels of Se in general is beneficial to enhance the metabolic activity and the chronic and cumulative amounts of Se certainly cause derrangement in metabolic activities and cellular damage, making the test species less fit for better survival.